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Differentiation of 2,3-Methylenedioxyamphetamine from 3,4-Methylenedioxyamphetamine

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ABSTRACT: The 2,3- and 3,4-methylenedioxyamphetamine isomers can be distinguished using the sulfuric acid color test, gas chromatography, infrared spectroscopy, mass spectrometry, and ^{13}C nuclear magnetic resonance.

KEYWORDS: toxicology, 3,4-methylenedioxyamphetamine, identification systems, synthesis

The hallucinogenic drug, 3,4-methylenedioxyamphetamine (3,4-MDA), is currently listed under Schedule I of the Federal Comprehensive Drug Abuse Prevention and Control Act of 1970 [*1*]. During the course of its analysis by the Virginia Bureau of Forensic Science, it was questioned if the tests routinely employed by the drug analysis laboratory would be capable of distinguishing 3,4-methylenedioxyamphetamine from its only ring-modified positional isomer, 2,3-methylenedioxyamphetamine (2,3-MDA). A search of the literature revealed no information pertaining to the characterization of this isomer even though it is considered included under the above Act [*2*]. This paper reports the synthesis and analytical data necessary for the unambiguous differentiation of 2,3-methylenedioxyamphetamine from 3,4-methylenedioxyamphetamine.

Materials and Methods

The 3,4-MDA was an old sample obtained from Aldrich Chemical Co., Inc. (Milwaukee, WI) as β-(3,4-methylenedioxyphenyl)-isopropylamine. It was further purified by distillation under reduced pressure to a colorless oil and the hydrochloride salt was prepared as described in the experimental procedure for 2,3-MDA. Color test reagents were of standard composition [*3*]. Melting points were taken on a Mettler FP-2 melting and boiling point apparatus (Highstown, NJ). Thin-layer chromatography (TLC) was performed on Analtech precoated silica gel plates (Newark, DE). Gas chromatography was performed on a Bendix Model 2600 instrument (dual column) with a dual flame ionization detector (FID) (Lewisburg, WV). Coiled glass columns (152.4 by 3.175 mm [6 ft by $\frac{1}{8}$ in.] inner diameter) packed with 3% or

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5% OV-7 or 10% OV-1 on Gas Chrom Q, 100-120 mesh, were used. The operating conditions were as follows: nitrogen was the carrier gas (30 mL/min), the oven temperature was 150 or 200°C, the injection port was 260°C, and the detector temperature was 260°C. Gas chromatography/mass spectrometry (GC/MS) was carried out using a Hewlett-Packard Model 5993 GC/MS system (Palo Alto, CA). Infrared spectra were obtained with a Beckman Acculab 8 and ultraviolet spectra were obtained on a Beckman Model 25 (Berkeley, CA). The proton and carbon spectra were obtained on a JOEL FX900 11 with a multinuclear probe and a JEC-980B computer system FAFT 26/27/28 software system (Cranford, NJ). The ^{1}H and ^{13}C chemical shifts were referenced to sodium 2,2-dimethyl-silapentane-5-sulfonate or tetramethylsilane with a sweep width of 10 000 Hz and an 8-K transform, a pulse angle of 90°, and a spectrometer frequency of 22.6 (^{13}C) or 80 mHz (^{1}H).

2,3-Methylenedioxybenzaldehyde

To 1.49 gm (10 mmol) of 2,3-dihydroxybenzaldehyde dissolved in 30 mL of dimethylformamide containing 7.93 gm (50 mmol) of anhydrous cesium fluoride was added to 1.0 mL (14.2 mmol) of dibromomethane according to the procedure of Clarke et al [*4*]. A yellow oil or white crystalline solid was obtained in 44% yield, melting point 32°C (literature 34°C) [*5*].

1-(2,3-Methylenedioxyphenyl)-2-Nitropropene

To 660 mg (4.4 mmol) of 2,3-methylenedioxybenzylaldehyde dissolved in 25 mL of benzene, 6 mL of nitroethane and 380 mg (4.9 mmol) of ammonium acetate were added. The mixture was refluxed for 12 h (overnight), cooled, washed with 30 mL of brine three times, dried over anydrous magnesium sulfate, and evaporated to dryness to give a yellow solid. Recrystallization from methanol gave 415 mg (75% yield) of 1-(2,3-methylenedioxyphenyl)-2-nitropropene as dark yellow crystals. The melting point was 74 to 77°C. Infrared data (with KBr pellets) were these: 1660 (m), 1520 (s), 1455 (s), 1315 (s), 1250 (s), 1065 (s), 960 (s), 930 (m), 795 (m), 770 (m), 720 (m) cm^{-1} (where m is moderate and s is strong). Proton nuclear magnetic resonance (NMR) data (in deuterochloform) were these: δ 8.02 (s, 1H, vinyl), 6.88 (s, 3H, Ar), 6.33 (s, 2H, CH_2), 2.40, 2.17 (s, 3H, CH_3) (where δ is the chemical shift in parts per million relative to the internal reference and s is singlet). The ^{13}C NMR data (in deuterochloform) were these: 148.6 (C-O), 127.1, 121.9, 109.3 (Ar), 101.4 (CH_2), and 14.5 (CH_3). The mass spectrum contained the parent peak at *m/z* 207, base peak at 103, and other prominent peaks at 147, 77, and 51.

2,3-Methylenedioxyamphetamine Hydrochloride (2,3-MDA)

To 500 mg (2.4 mmol) of 1-(2,3-methylenedioxyphenyl)-2-nitropropene dissolved in tetrahydrofouran was added 200 mg (5.3 mmol) of lithium aluminum hydride (LAH) and the mixture was refluxed for 1 h. Excess LAH was destroyed [*6*], and the ether filtrate was dried over anhydrous sodium sulfate and evaporated to a yellow oil. The oil was further purified by distillation under reduced pressure to give a colorless oil. The oil was dissolved in anhydrous ether and the hydrochloride salt was prepared by passing hydrochloride gas through the ether. Recrystallization in ethanol/ether gave 96 mg (19% yield) of 2,3-methylenedioxyamphetamine. The melting point was 158 to 160°C. Proton NMR data (in deuterium oxide) were these: δ 6.73 (m, 3H, Ar), 5.84 (s, 2H, OCH_2O), 3.56 (q, 1H, NC-H), 2.79 (d, J = 7.3, 2H, CH_2), and 1.18 (d, J = 6.6, 3H, CH_3) (where q is quartet, d is doublet, and J is a coupling constant in hertz). The ultraviolet data (with 0.04% in 0.1*N* sulfuric acid) were these: maxima 283 nm (ϵ = 2258), inflection at 233 nm, and minima at 126 nm (where ϵ is molecular extinction coefficient). The mass spectrum contained the parent peak at *m/z* 179 (1.4%) and a base peak at *m/z* 44 ($C_2H_6N^+$). What was expected to be found for 2,3-MDA hydrochloride was 55.69 (C) and 6.54 (H). What was found was 55.48 (C) and 6.60 (H).

Results and Discussion

The capacity to detect and differentiate 2,3-MDA from its only other ring-modified positional isomer using the routine analytical tests employed by the forensic drug chemist is possible. Of the color tests evaluated, only the sulfuric acid test gave a clear and distinctive difference when compared with 3,4-MDA. In the sulfuric acid color test 2,3-MDA gave a rose/pink color whereas the 3,4-MDA gave a deep purple color. In the other color tests evaluated (Marquis, Mecke, Froehde, Mandelin, and nitric acid) a positive color response was observed, however, the difference in color when compared with 3,4-MDA was not significantly different.

Thin-layer chromatography using commonly employed solvent systems (Table 1) would not differentiate the two isomers. However, analysis by gas chromatography gave a significant different in retention time of 1.0 and 0.5 min when compared to 3,4-MDA (Table 2).

The infrared spectra of 2,3- and 3,4-MDA (Fig. 1) were similar except for the out of plane bending frequencies which are usually indicative of the aromatic substitution pattern [7]. The 2,3-MDA has a moderate absorption band at 780 cm which is characteristic for a 1,2,3-substituted aromatic ring whereas 3,4-MDA has strong absorption bonds at 820 and 870 cm which is characteristic of a 1,2,4-substituted aromatic ring. The 3,4-MDA also exhibits a strong absorbance at 1500 cm^{-1} which is very weak in the 2,3 isomer.

The ultraviolet spectrum of 2,3-MDA in 0.1*N* sulfuric acid was quite similar to 3,4-MDA except for a small hypsochromic (283 versus 286 nm) and hypochromic shift (ϵ = 2258 versus ϵ = 3064) for the B-band absorption. Also, the absorption at 234 nm was much weaker and appeared only as a shoulder.

Comparison of mass spectra (Fig. 2) indicates that both isomers gave a base peak at *m/z* 44 because of cleavage next to the amine and loss of the relatively stable methylenedioxybenzyl radical [8]. In addition, the 3,4-MDA gave a prominent ion at *m/z* 136 (15.7%) because of hydrogen rearrangement and elimination of a neutral amino olefin whereas this ion was very weak in 2,3-MDA (1.8%).

Although it was anticipated that proton NMR could clearly distinguish the aromatic substitution pattern this was not observed in that both isomers gave a poorly resolved ABC aromatic pattern that was similar. Indeed, unless one was specifically expecting the 2,3-MDA isomer it could easily be identified as 3,4-MDA using proton NMR. ^{13}C NMR is currently being evaluated as a tool for forensic drug analysis because of its ability to clearly distinguish

TABLE 1—*Thin-layer chromatography on silica.*

Solvent	2,3-MDA	3,4-MDA
NH_4OH/CH_3OH (1.5/100)	0.36	0.33
$CHCl_3/CH_3OH$ (1/1)	0.19	0.19
$CHCl_3/CH_3OH$ (9/1)	0.14	0.14
NH_3 sat $CHCl_3/CH_3OH$ (18/1)	0.42	0.36

TABLE 2—*Gas chromatography* (t_R, *min*).

Conditions[a]	2,3-MDA	3,4-MDA
5% OV-7, 150°C	5.4	6.3
10% OV-1, 150°C	3.8	4.3
3% OV-7, 200°C	0.6	0.7

[a]Conditions are described in the experimental section.

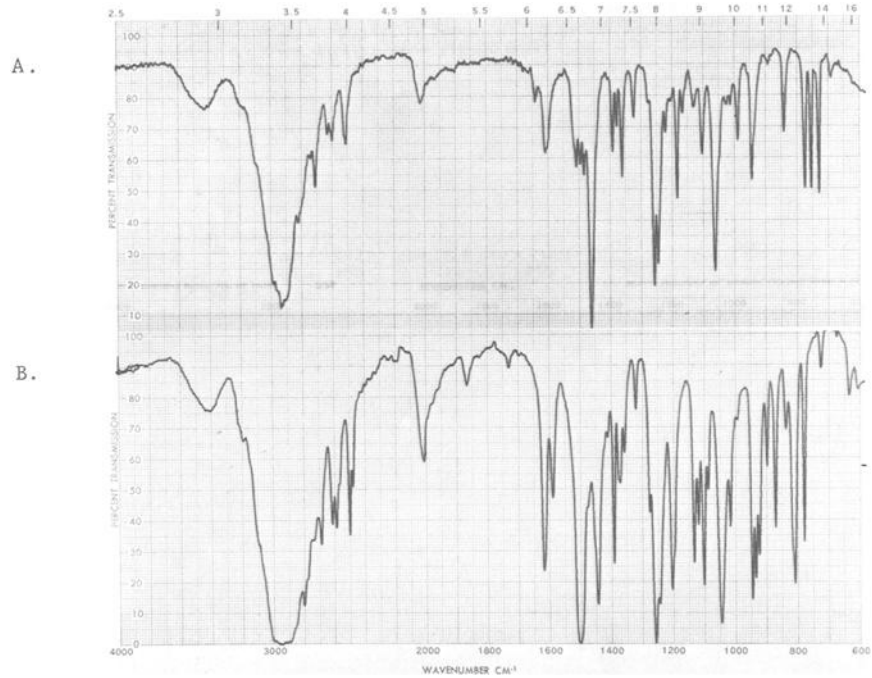

FIG. 1—*Infrared spectra of* (a) *2,3-MDA* · hydrochloride and (b) *3,4-MDA* · *hydrochloride. All as KBr pellets.*

TABLE 3—*Data from the* ^{13}C *NMR spectra of methylenedioxyamphetamines.*

	2,3-MDA[a]	2,3-MDA · HCl[b]	3,4-MDA[a]	3,4-MDA · HCl[b]
C-1	121.1	120.0	132.9	132.5
C-2	145.9[c]	148.3[c]	107.5[d]	111.4[d]
C-3	147.0[c]	149.5[c]	147.1[c]	150.1[c]
C-4	106.7	110.7	145.5[c]	148.9[c]
C-5	123.3	126.2	108.9[d]	112.4[d]
C-6	121.4	124.8	121.5	125.4
α	40.3	36.6	45.7	42.4
β	47.2	50.6	47.9	51.8
γ	23.5	20.1	22.8	20.0
$-OCH_2O-$	100.3	103.6	100.3	103.8

[a]Solvent: $CDCl_3$.
[b]Solvent: D_2O.
[c,d]These assignments may be reversed.

isomers [*9,10*]. In comparison of the 2,3-MDA to 3,4-MDA (Table 3), all of the carbons gave a unique and characteristic absorbance. The actual aliphatic carbon absorbances for both the free base and hydrochloride salt forms were in excellent agreement with what would be predicted from Bailey and Legault's studies on the mono-, di-, and tri-methoxyamphetamines [*9,10*]. The aromatic carbon absorbances for both of the methylenedioxyamphetamines were further upfield (2 to 5 ppm) but in the same relative order as that observed with the corresponding methoxyamphetamine. This provides further evidence for the usefulness of ^{13}C NMR as a unique analytical tool for forensic drug identification.

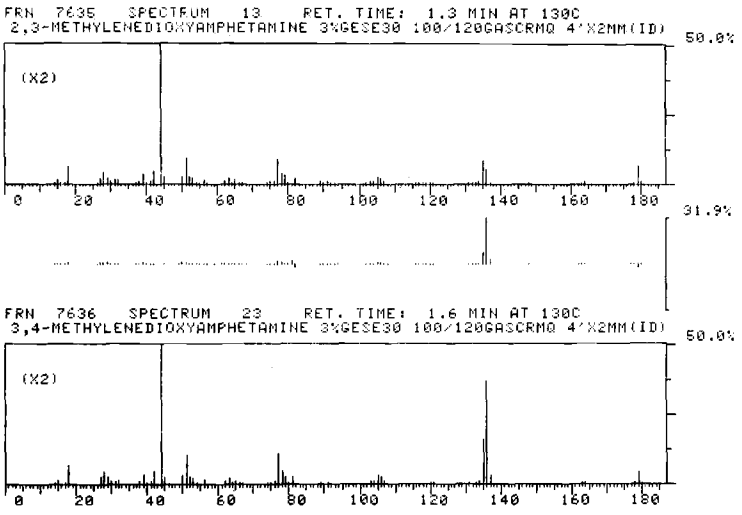

FIG. 2—*Mass spectral (electron impact) comparison of 2,3-MDA and 3,4-MDA.*

Conclusion

In conclusion, it would appear from the analytical data that a sulfuric acid color test, gas chromatography, infrared spectroscopy, mass spectrometry, and ^{13}C NMR provide distinctive methods for differentiating the two possible positional isomers of methylenedioxyamphetamine.

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