Technical Note

Levamisole: An Analytical Profile

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ABSTRACT: Levamisole, an antineoplactic cancer medication used in the treatment of colon cancer, has been identified in numerous submissions of illicit cocaine hydrochloride. Analytical methodologies and data (gas chromatography, capillary electrophoresis, infrared spectroscopy, mass spectroscopy, and proton nuclear magnetic resonance spectroscopy) are presented.

KEYWORDS: Levamisole, Cocaine, (l)-Tetramisole, Ergamisol, Ketrax, Solaskil, Forensic Chemistry

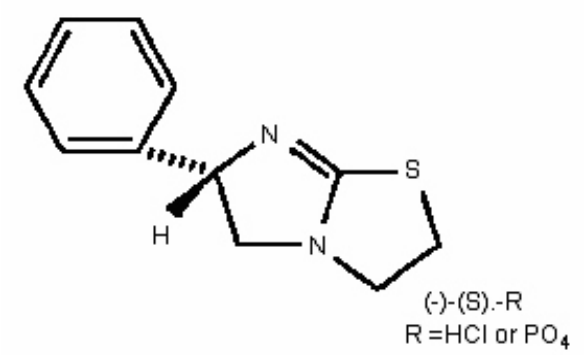

Figure 1: Structure of Levamisole

Introduction

Over approximately the past two years, this laboratory has received numerous cocaine submissions containing various amounts of levamisole, (S)-2,3,5,6-tetrahydro-6-phenylimidazo[2,1-b]thiazole [1,2]. Levamisole is the *levo* enantiomer of tetramisole, and is a synthetic imidazothiazole derivative that has been widely used in the treatment of worm infestations in both humans and animals. In 1990 the U.S. Food and Drug Administration approved the use of levamisole in combination drug therapy with another cancer drug, fluorouracil, for patients to treat some advanced cases of colon cancer [3]. Analytical data for levamisole is provided.

Experimental

Levamisole: $C_{11}H_{12}N_2S$; mw = 204.3 amu [2]

Source: Sigma-Aldrich, Inc. (St. Louis, MO); Lot #073K3602

Gas Chromatography

Instrument	Agilent 6890N with a flame ionization detector
Column	HP-5, 30 m x 0.25 mm x 0.25 μm film thickness
Injector Temperature	270 ^{O}C
Oven Temperature	215 ^{O}C for 5.5 min, 45 ^{O}C to 250 ^{O}C for 1.4 min
Carrier Gas	Helium ramped flow 2.7 mL/min for 5.5 min to 5 mL/min
Split Ratio	75:1

The retention time for levamisole is 2.99 minutes under the above experimental parameters. The retention time relative to cocaine is 0.58.

Capillary Electrophoresis

Instrument	Agilent HP^{3D} CE Capillary Electrophoresis System with a diode array detector
Column	Bare fused silica capillary, 50 μm ID, 40 cm LEF
Run Buffer	Microsolve DEA Custom Chiral, Phenethylamine and Propoxyphene Buffer containing 78.8 mg/mL 2-hydroxypropy-ß-cyclodextrin
Detector	200 nm, reference 480 nm
Voltage	20 kV
Cassette Temperature	15 ^{O}C
Precondition	Flush 1.0 min 0.1 NaOH
Flush Sequence	1.0 min Water; 1.0 min Microsolve CElixir A; 2.0 min Microsolve DEA Custom Chiral, Phenethylamine and Propoxyphene Buffer containing 78.8 mg/mL 2-hydroxypropy-ß-cyclodextrin
Injection Parameters	Pressure 35.0 mbar, 2.0 sec sample vial Pressure 35.0 mbar 1.0 sec water
Injection Solvent	3.75 mM Sodium Phosphate solution, pH 3.2

Note: The above instrumental parameters enables resolution of *dextro*- and *levo*- enantiomers of tetramisole [4].

Infrared Spectroscopy

Instrument	Thermo-Nicolet Nexus 670
Number of Scans	16
Resolution	4.000 cm^{-1}
Wavenumber Range	4000 cm^{-1} to 650 cm^{-1}

Data was obtained by the use of an attenuated total reflectance (ATR) attachment on FTIR [Figure 2]. The data was not corrected.

Mass Spectrometry

Instrument	Agilent 5973
Column	HP-5 MS, 30 m x 0.25 mm x 0.25 μm film thickness
Injector Temperature	255 ^{O}C
Oven Temperature	90 ^{O}C for 1.35 min, 35 ^{O}C/min to 290 ^{O}C
Carrier Gas	Helium with a 35:1 split ratio
Scan Range	40 - 550 amu

The electron impact mass spectrum is presented in Figure 3.

Nuclear Magnetic Resonance Spectroscopy

Data was obtained using 1D proton Nuclear Magnetic Resonance on a Varian Mercury 400 MHz NMR. The sample was prepared at 25.2 mg/mL in deuterated methanol (CD_3OD) containing TMS (tetramethylsilane) as the reference at 0 ppm. The proton spectrum of the standard was obtained with 8 scans using a 1.0 second delay, 45

degree pulse, and a 2.99 second acquisition time. Data from sweep width of 6410 Hz was stored in 32K data points [Figure 4].

Results and Discussion

The presence of pharmacologically active adulterants and inactive diluents found in illicit cocaine seizures is common. Many of these adulterants cause pulmonary and systemic reactions, and therefore may contribute to the toxicity of the cocaine. However, after a brief internet inquiry concerning adulterating illicit cocaine with levamisole, it is unclear as to why this relatively expensive compound is being used.

References

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3. http://www.fda.gov/bbs/topics/news/new00051.html

4. Lurie IS, Bethea MJ, McKibben TD, Hays PA, Pellegrini P, Sahai R, Garcia AD, Weinberger R. Use of dynamically coated capillaries for the routine analysis of methamphetamine, amphetamine, MDA, MDMA, and cocaine using capillary electrophoresis. Journal of Forensic Sciences 2001;46(5):1025-1032.

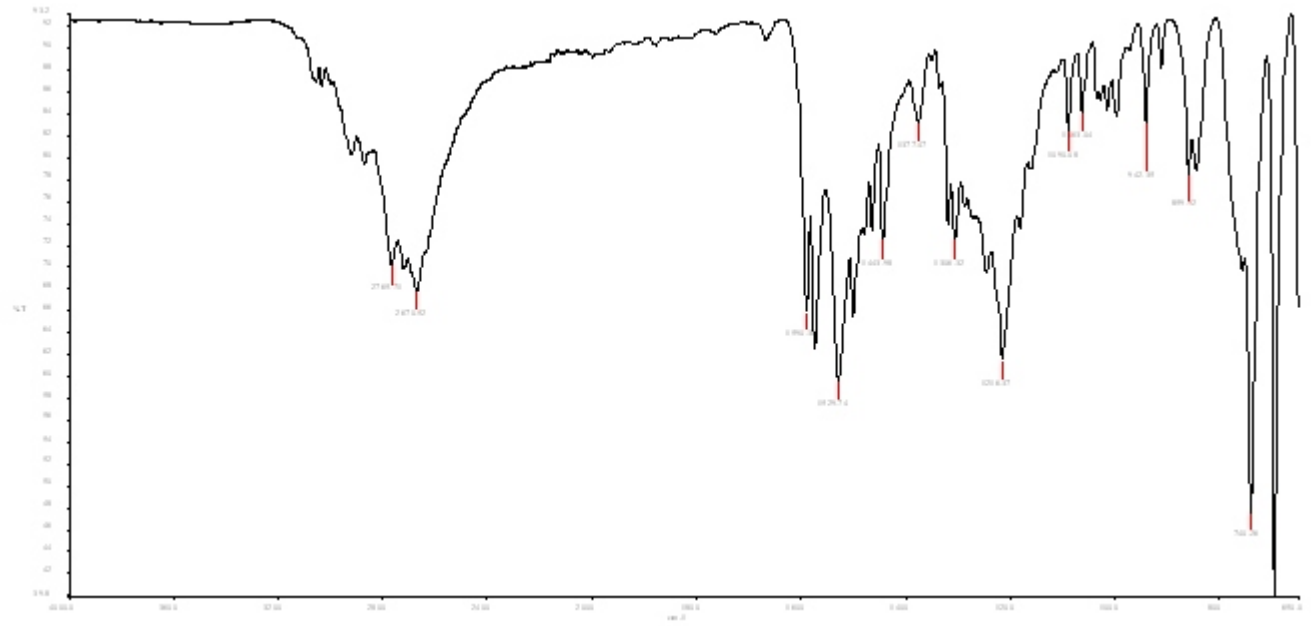

Figure 2: Uncorrected FTIR-ATR Spectrum of Levamisole Hydrochloride.

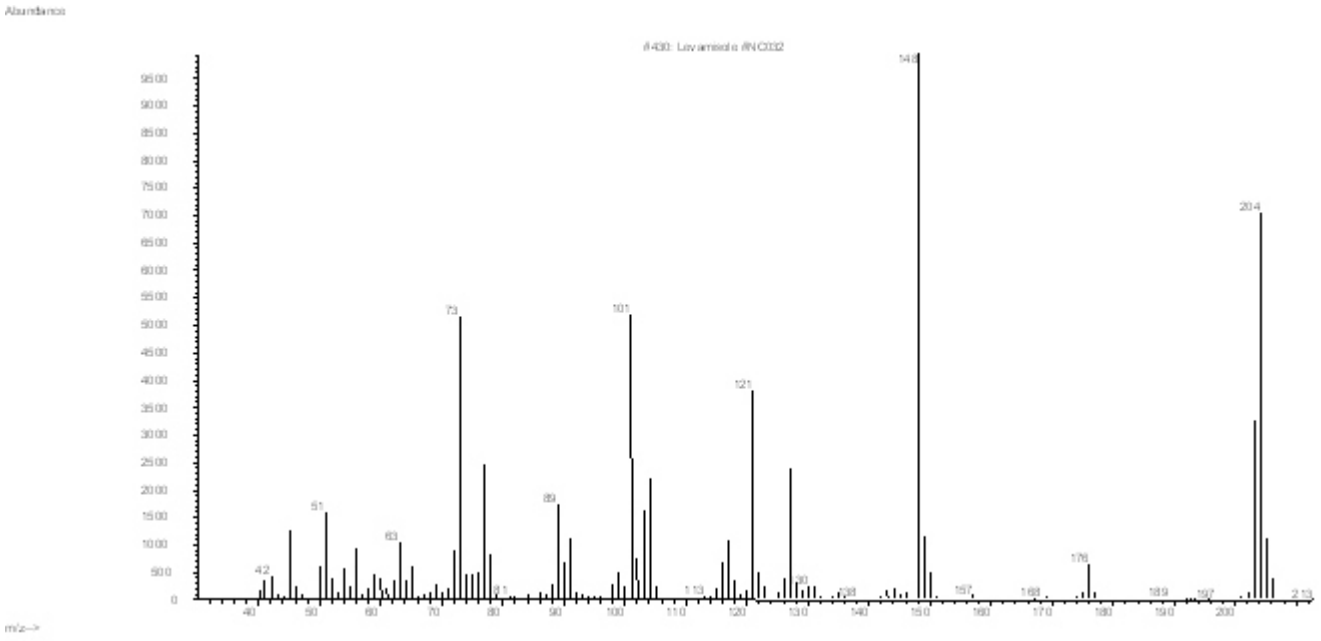

Figure 3: Electron Impact Mass Spectrum of Levamisole.

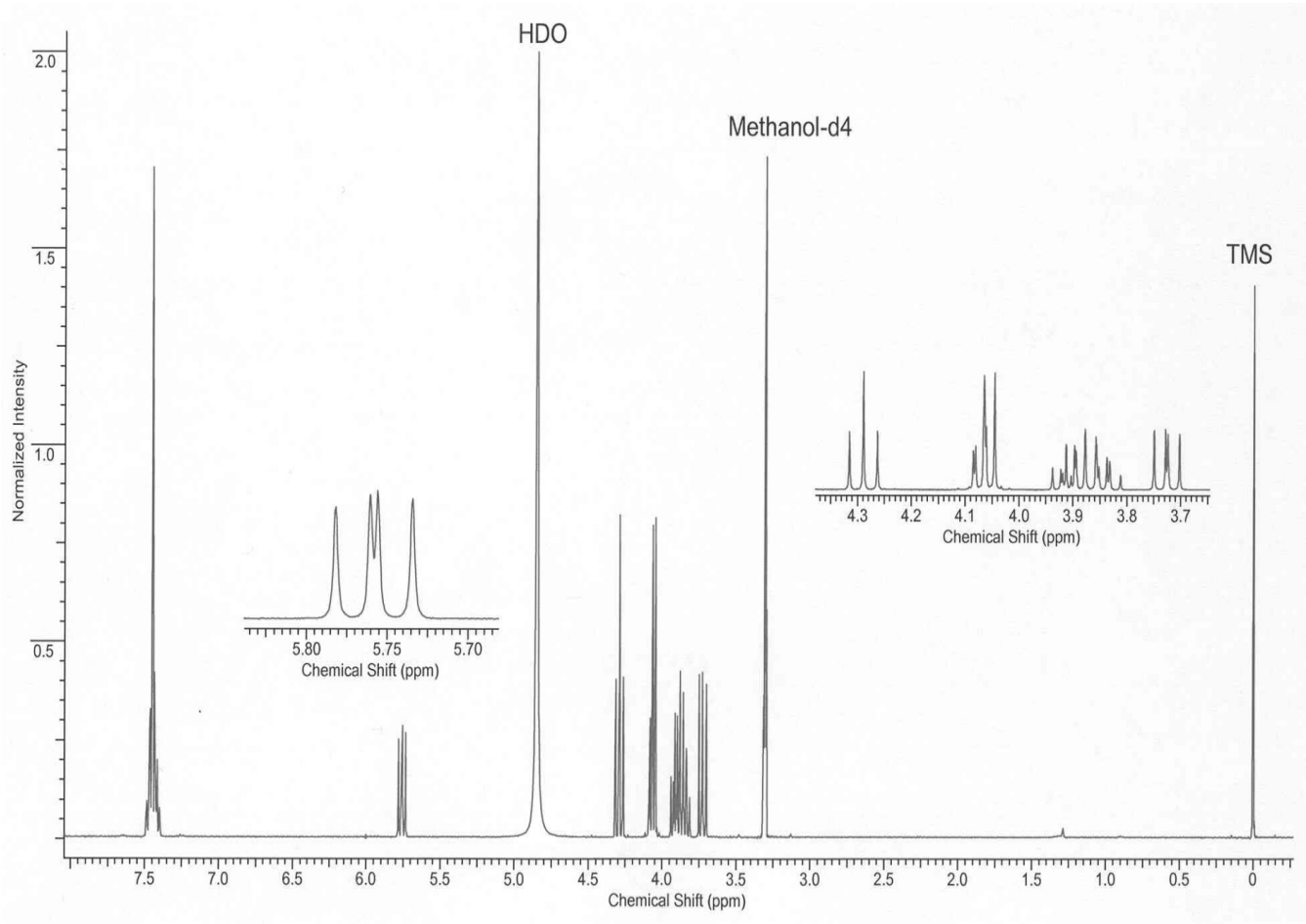

Figure 4: 400 MHz Proton NMR Spectrum of Levamisole Hydrochloride in CD_3OD.