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Colorimetric Determination of 3,4-Methylenedioxyamphetamine (MDA)

MDA (α-methyl-3,4-methylenedioxyphenethylamine) is one of several arylalkylamine hallucinogens appearing in the illicit drug market. A simple, sensitive method for detecting and estimating the compound is thus desirable.

The ultraviolet absorption spectrum of MDA in dilute acid consists of two maxima of unremarkable intensity at 234 and 286 nm. The native fluorescence of MDA (excitation maximum 285 nm, emission maximum 325 nm; both uncorrected) provides adequate sensitivity but special equipment, not readily available in many laboratories, is required. Moreover, another illicit hallucinogen, 2,5-dimethoxy-4-methylamphetamine (STP), exhibits similar fluorescence characteristics and may interfere in the identification and determination of MDA.

Inspection of the structure of MDA reveals the presence of the $-OCH_2O-$ group, which characteristically loses a mole of formaldehyde when warmed in strong sulfuric acid. The liberated aldehyde may be determined with good sensitivity by its reaction with a variety of color reagents including chromotropic and gallic acids [*1*]. This behavior provided the basis for the development of the method described in this report.

Experimental

Reagents

Chromotropic acid (recommended for quantitative estimation)—One gram of the disodium salt of 4,5-dihydroxy-2,7-naphthalene disulfonic acid is dissolved in 100 ml of concentrated sulfuric acid.

Gallic acid (preferred for spot testing and as a thin-layer chromatography (TLC) spray reagent)—One gram of gallic acid is dissolved in 100 ml of concentrated sulfuric acid.

Procedures

Preparation of Calibration Curve—A stock reference solution containing 1 mg of authentic MDA per ml of 1 N H_2SO_4 is diluted with the same solvent to provide working solutions in the range 0 to 100 μg/ml. To each of 0.2-ml aliquots of the working reference solutions is added 1 ml of the chromotropic acid reagent. The mixtures are heated for 10 min in a heating block set at 80 C. After the solutions have cooled to room temperature, 4 ml of glacial acetic acid are added to each and the absorbances of the violet colors are measured photometrically at 575 nm against a reagent blank. The data are used to plot a calibration curve.

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The same procedure is applied if gallic acid is used as the color reagent, but the absorbance of the resulting blue pigment is measured at 595 nm.

Contraband Materials—A weighed portion of the confiscated substance is dissolved in 10 ml of water. The solution is made alkaline with 2 drops of 50 percent NaOH and extracted with 15 ml of ether. The ether is washed once with water and back-extracted with 1 ml of 1 N H_2SO_4. A 0.2-ml aliquot of the acid layer is treated identically as were the reference solutions.

Biologic Specimens—Ten ml of plasma or urine are made alkaline with 50 percent NaOH. Extraction of MDA and color production are performed as previously described for contraband materials.

Results and Discussion

The reaction between MDA and gallic acid furnishes a vivid blue pigment with an absorption maximum at 595 nm. This reaction is most useful as a spot test for rapid presumptive identification or as a chromogenic spray for TLC plates. The product of the reaction between formaldehyde liberated from MDA and chromotropic acid is the well-known violet pigment with a maximum absorbance at 575 nm.

Both absorbances follow Beer's Law over the range 0 to 4 μg of MDA per ml of final solution. The slope of the calibration curve for the gallic acid pigment is approximately 0.08 absorbance unit per microgram of MDA while that for chromotropic acid is approximately 0.12 absorbance unit per microgram. The latter represents approximately a sixfold increase in sensitivity over the ultraviolet spectrophotometric method in which 35.4 μg of MDA per ml of dilute acid exhibits an absorbance of 0.7 at 286 nm [*2*].

Both colors are stable at room temperature for at least 2 h, and show no perceptible decrease in absorbance during that period.

The mechanisms for both reactions are probably similar. The color production probably is the result of initial condensation between the aromatic hydroxy compound and formaldehyde, yielding a colorless hydroxydiphenylmethane derivative. This step is followed by oxidation to a *p*-quinoidal pigment [*3*].

To our knowledge neither the biologic half-life nor the metabolic fate of MDA in humans has been elaborated. But, since the customary dose necessary for producing the hallucinatory experience in adults is around 150 mg [*4,5*], it does not appear unreasonable that microgram quantities of the drug should be detectable in plasma for at least short periods of time after administration. Detectability in urine by the procedure described will depend on preservation of the integrity of the —OCH_2O—group. If the compound undergoes extensive deamination it may be necessary to search the acid and neutral fractions for metabolites. We have yet to encounter a case in which positive findings were observed in either plasma or urine. However, average recoveries of 50 μg of MDA added to 10 ml of water, urine, and plasma are 85, 85, and 80 percent, respectively.

Two reported precursors of MDA, piperonal and 3,4-methylenedioxy-1-(2-nitropropenyl)benzene [*6*] would be expected to yield positive results by the method described as would myristicin, methysticin, and dihydromethysticin, and safrole, the pharmacologically active principles of nutmeg, kava kava, and sassafras. These substances, however, do not contain an amine and would be extractable as neutral compounds. The only compound of this group available to us was piperonal. Tests on piperonal with both gallic and chromotropic acids provided results which were identical to those for MDA.

Among the many commonly encountered drugs tested for interference only thioridazine furnished a stable greenish-blue pigment. This color is due, not to the gallic (or chromotropic) acid, but rather to the acidic reaction medium. Accordingly, a parallel test in which

the reactive component is omitted from the reagent will reveal the presence of thioridazine. Other phenothiazines yield their characteristic reddish colors in acid, but even in proportions of 10 parts chlorpromazine to one part MDA, the blue color is evident when the gallic acid spot test is applied. Codeine, quinine, mescaline, and STP, all bearing the methoxy function, do not provide positive results under the experimental conditions described above. On the other hand, narcotine, a constituent of raw opium, contains the —OCH_2O— group and furnishes positive results.

Summary

A method for the detection and determination of the hallucinogen, MDA, is described. The method takes advantage of the color produced when the compound is heated with either gallic or chromotropic acid in concentrated sulfuric acid. Sensitivity and specificity of the reaction are discussed as are recoveries of the drug from urine and plasma.

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