TECHNICAL NOTE

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A Method for Identification of Lysergic Acid Diethylamide (LSD) Using a Microscope Sampling Device with Fourier Transform Infrared (FT/IR) Spectroscopy

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ABSTRACT: The identification of lysergic acid diethylamide (LSD) has posed an analytical challenge for forensic science laboratories. In those cases in which a few doses are seized, only microgram quantities are available, often in forms which make isolation of the miniscule amount of LSD difficult. A method is described which yields small crystals of pure LSD in a form well-suited for analysis using a microscope sampling device with a Fourier transform infrared (FT/IR) spectrometer. These crystals produce excellent spectra from samples containing less than 50 μg of LSD. Distinguishing between LSD, iso-LSD, and lysergic acid *N*-methylpropylamide (LAMPA) poses no problem with the spectra obtained. This scheme combines preparative thin-layer chromatography (TLC) followed by wick evaporation, an old but not well-known technique for separating soluble components from high-solid mixtures without filtration.

KEYWORDS: toxicology, lysergic acid diethylamide (LSD), spectroscopic analysis, Fourier transform infrared spectroscopy

The identification of lysergic acid diethylamide (LSD) has posed a considerable problem for forensic laboratories for many years. Where only a few dosage units are seized, only microgram (μg) quantities are present. Further, the LSD is mixed with large amounts of excipients or in forms which make isolation of such minuscule amounts difficult.

In recent years, challenging drug analysis problems have usually been solved using gas chromatography/mass spectrometry (GC/MS). Unfortunately, LSD is infamous for its poor chromatographic characteristics, which makes GC/MS unsuitable under most conditions. Even with silica capillary columns it is by no means trivial to subject LSD consistently to gas chromatography (GC) successfully. Since the decomposition of LSD on a GC column appears to be easily catalyzed, successful chromatography can depend on the immediate past history of the column.

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Clark has shown [*1*] that under carefully controlled conditions it is possible to differentiate LSD from lysergic acid *N*-methylpropylamide (LAMPA) and iso-LSD by a rigorous examination of their mass spectra. However, because the differentiation is based on subtle changes in the intensity ratios, the entire process must be done with great care. Applying this method to casework samples would seem to be difficult.

Where identification is needed and GC/MS is not well suited, infrared spectroscopy (IR) is usually an attractive alternative. The forensic science literature provides several schemes for the microsampling of LSD [*2–4*], but obtaining usable infrared spectra of LSD was quite difficult with dispersive instruments.

The advent of Fourier transform infrared spectroscopy (FT/IR), with its much improved sensitivity, has made IR a more practical solution to the LSD analysis problem. The sample preparation methods in the literature, however, are laborious, and obtaining good-quality spectra from micro potassium bromide pellets requires considerable patience and skill.

We have developed a scheme which yields small crystals of pure LSD in a form well suited for direct analysis using a microscope sampling device with an FT/IR. Crystals which produce excellent spectra can be obtained from samples containing less than 50 μg of LSD. This scheme combines preparative thin-layer chromatography (TLC) on 1 by 3-in. (25.4 by 76.2-mm) TLC plates followed by an old, but not well-known, technique called wick evaporation [*5*]. Wick evaporation is used to separate soluble components from a solution with large amount of undissolved solids, without filtration.

Experimental Procedure

The method accommodates samples with 20 to 100 μg of LSD. If direct extraction of dosage units is to be the source, use two dosage units, if available, and extract with 2 mL of methanol. Protect from bright light during extraction. To the solution obtained, add two drops of concentrated ammonium hydroxide and gently evaporate to dryness, excluding bright light.

Redissolve the material in about 1 mL of methyl *tert*-butyl ether (MTBE) and streak the entire solution across two 1 by 3-in. (25.4 by 76.2-mm) TLC plates. Plates can be developed using any solvent system which gives resolution between LSD, LAMPA, and iso-LSD. We have used the following two systems: diethyl ether, cyclohexane, acetone, diethylamine (35:40:35:6) and chloroform, acetone (4:1).

A test plate with known LSD, LAMPA, and iso-LSD should be developed with the same solvent system to determine the Rf value for LSD under the conditions employed and demonstrate resolution of the other compounds. After the streaked plates are developed, the LSD band is located using the known Rf value and an ultraviolet (UV) light box. The LSD band is marked and the band of silica containing the LSD is scraped from both plates with a razor blade and collected. Place the silica obtained in a small vial which has been carefully cleaned and rinsed. A 2-mL portion of methyl *t*-butyl ether (MBTE) is added to the vial, which should be protected from light while being sonicated or shaken for about 5 min.

A prepared Fiberglas wick (see below) is placed in the vial (Fig. 1), which is protected from direct light, and the solvent is allowed to wick evaporate to dryness in a fume hood. This will take about 4 h with MTBE as the solvent. The process can be speeded using an enhanced flow of dry gas past the wick. If the humidity is low, diethyl ether can be used as the solvent and wick evaporation is much faster. If the humidity is high, diethyl ether will cool the wick so much upon vaporization that ice crystals form on the wick.

To prepare the wick, twist a small amount of glass wool to make a narrow rope about 2 mm in diameter. The wick should be about 2 in. (50 mm) long. It is convenient to

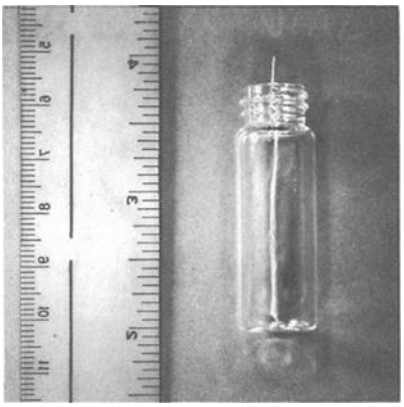

FIG. 1—*The prepared Fiberglas wick.*

make one wick about 4 in. (10 cm) long with both ends tapered and cut it in half. Since all nonvolatile ether soluble material will be concentrated at the tip of the wick, the method is extremely sensitive to contamination. Therefore, it is critical to insure that the wick and vial are free of even trace contamination. Contamination is often evidenced by an unwanted ester carbonyl peak at about 1720 cm^{-1} in the IR spectra obtained.

To clean the wick, soak it in methanol (preferably for several hours) and drain. Using forceps, transfer wick to a glass microscope slide and use another glass slide, perpendicular to the first, to hold the tip of the wick against the first slide. In that way the wick is touched only by clean glass slides for the final washing step. Wash thoroughly using a microsyringe or small pipette, first with fresh methanol (stored in glass) and then with ether. Make sure that the entire length of the wick, including both ends, has been thoroughly rinsed with both solvents. Wicks should then be air dried and stored in a clean, dry container for future use. Good wicks can be cleaned and reused, but the wick-making process, although sounding complicated, is really quite simple with a little practice, and fresh wicks are recommended.

When evaporation is complete, crystals should be found just below the tip of the wick, usually in a circular pattern around the wick at the point the wick emerges from the top of the vial. Crystals are best transferred by placing the wick directly on an IR sampling crystal [barium fluoride (BaF_2) or potassium chloride (KCl)] and using a fine forceps or needle to dislodge tiny crystals from the wick on to the IR crystal. Locating and transferring the crystals is best done on the stage of a stereo microscope since they are barely visible to the unaided eye.

Discussion

Often the crystals formed on the wick are thin enough so that an IR spectrum can be obtained directly using the microscope sampling device. If there are no crystals thin enough to yield reasonable transmission, the crystals can be flattened with a miniature roller or a needle probe to yield excellent spectra (Fig. 2). The spectra obtained directly on the crystals have all the major characteristics of literature spectra obtained using potassium bromide (KBr) pellets. The crystal spectra, however, appear to be a little sharper and better resolved. Even with very small amounts of material, spectra can be obtained which provide unambiguous identification of LSD. Further, the infrared spectra obtained allow ready differentiation of LAMPA or iso-LSD from LSD (Figs. 3 and 4).

It should be emphasized that the addition of ammonium hydroxide to the initial extract is critical. LSD tartrate does not wick evaporate well: it produces oily globs which are

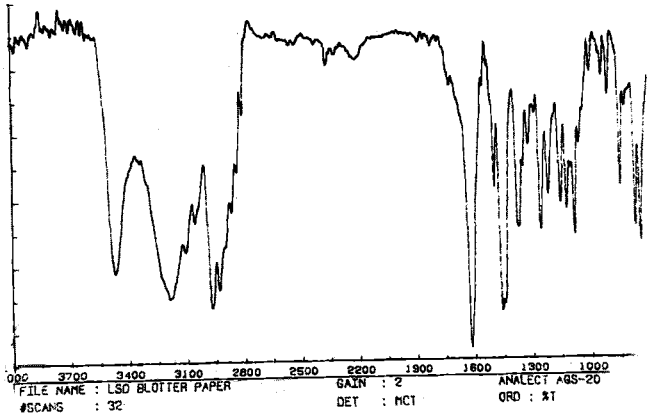

FIG. 2—*IR spectra of crystals from the wick.*

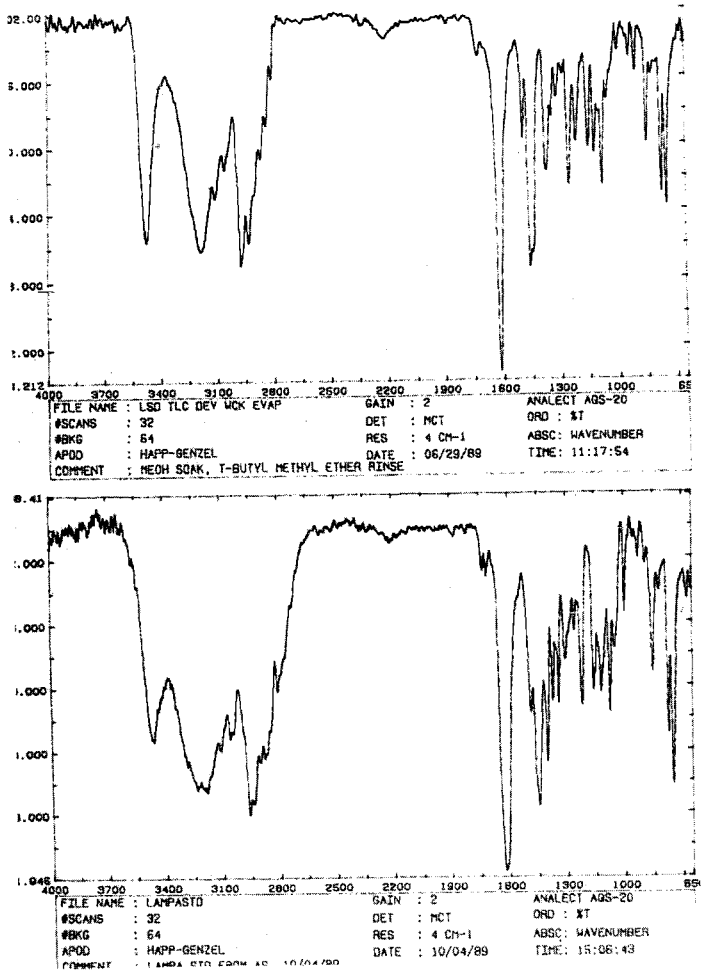

FIG. 3—*IR spectra of LSD* (top) *compared with that of LAMPA* (bottom).

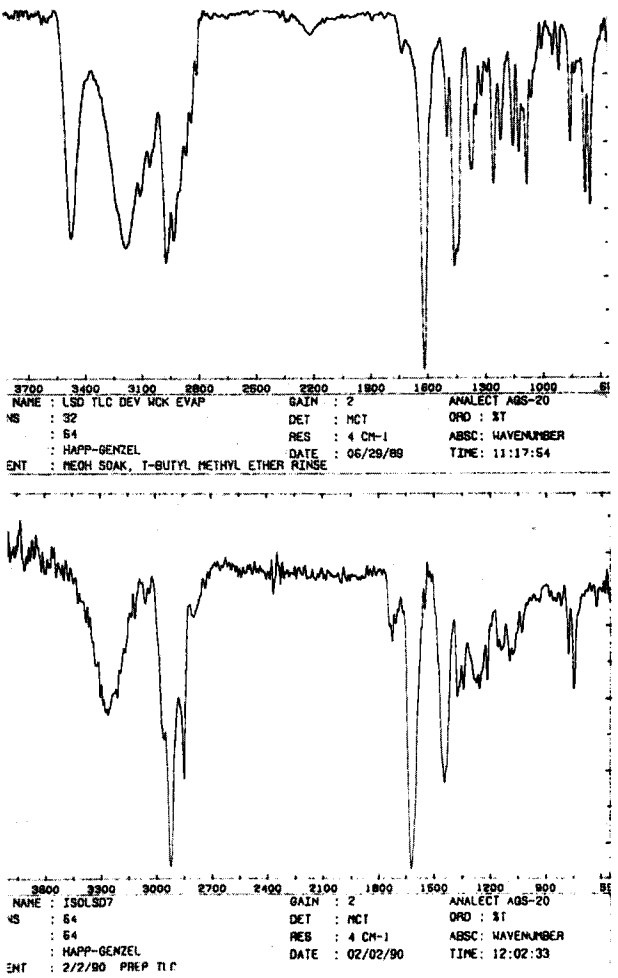

FIG. 4—*IR spectra of LSD* (top) *compared with that of iso-LSD* (bottom).

difficult to remove from the wick. It is hoped that the combination of preparative TLC and wick evaporation to produce microcrystals for IR identification will prove widely applicable. Several other drugs we have subjected to the process have also produced excellent spectra.

Our initial attempts to extract the LSD and obtain usable spectra after wick evaporation without the preparative TLC step were not successful. We obtained spectra of stearic acid from tablet dosage forms and other unidentified impurities from paper spots.

It should be mentioned that the method produces crystals of purified material which can be subjected to other tests. Even the crystals used to obtain the IR spectra can be removed from the IR sampling crystal and preserved for later use.

References

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