method than the column chromatographic method cited by A.O.A.C. for the analysis of A.P.C. tablets[11].

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Identification of LSD and other indole alkaloids by ultraviolet degradation products

Many forensic analysts use thin-layer chromatography (TLC) to identify LSD but have found two or more systems necessary to separate it from closely related compounds[1,2]. In using TLC, some workers[2–4] have observed degradation spots on the developed chromatograms. GENEST AND FARMILO[3] applied a degradation principle to the identification of LSD, using a two-hour acid hydrolysis followed by prolonged ultraviolet (U.V.) irradiation. Although this procedure is adequate for identification, it is quite time consuming. A relatively simple technique, using only one TLC system and requiring only one TLC plate, has been investigated for the identification of LSD.

The method involves controlled degradation of the alkaloids in a chloroform solution under U.V. irradiation. The degraded material is then chromatographed on a thin-layer plate by the procedure of ROMANO[5]. A series of colored spots on the thin-layer plate is then detected with an U.V. lamp. This series of spots, numbering as many as eight or nine, has been shown to be unique for each of the compounds tested.

Experimental

A 2–10 ml portion of chloroform solution containing 10 μg of the alkaloid in question is placed in an open container (20 ml glass beaker is satisfactory) and the

liquid is exposed to long wavelength (near 366 mμ) U.V. radiation. The exposure time and distance depend on the intensity of the source and must be experimentally determined. The irradiation conditions should be such that, after irradiation, the parent spot is present and is of an intensity near that for 1 μg of unirradiated material. In our laboratory, using a Chromato-Vue lamp Model XX-15C, irradiation was carried out for 13 min in a 20 ml beaker at 13 cm from the lamp. After degradation, the chloroform is evaporated without heat and the entire residue, with the aid of small amounts of acetone, is spotted on a glass plate coated with 250 μ of Silica Gel G. This plate is then developed; chloroform–acetone (1:4) mixture is used both for developing solution and for atmosphere saturation. This system separates LSD from *iso*-LSD[6]. Spots are then detected under short wavelength U.V. light. A 1 or 2 μg portion of the unirradiated material is spotted similarly to establish R_F value and intensity of parent spot.

Discussion

Irradiation in a solvent was found to produce more consistent spot patterns than irradiation on the TLC plate after spotting. Spots of degradation poducts will begin appearing for solutions that have been irradiated only a few minutes, and the number of spots will increase as time increases to near optimum conditions; then the number of spots will decrease with additional exposure. Some of the first spots to appear will diminish in intensity and eventually disappear with greater irradiation time.

Table I is a compilation of R_F values of ten related indole-containing compounds. Fig. 1 shows the relative positions of each spot as well as relative intensities. Irradiated portions represent 10 μg and unirradiated portions represent 2 μg of each compound except ergotoxin for which the amounts are 30 μg and 6 μg, respectively. In Table I, values marked with an asterisk represent R_F values for spots with intensities equal to

TABLE I

R_F VALUES FOR SOME INDOLE ALKALOIDS BEFORE AND AFTER IRRADIATION

Compound	*R_F of each observable spot*[a]							
LSD	0.27*							
LSD (irradiated)	0.0*	0.06*	0.27*	0.34*	0.39	0.55	0.63	0.70
Lysergic acid	0.0*							
Lysergic acid (irradiated)	0.0*	0.34	0.52	0.60	0.71			
Ergometrinine	0.19*							
Ergometrinine (irradiated)	0.0*	0.02*	0.17*	0.20*	0.24	0.44	0.51	
Ergotaminine	0.58*							
Ergotaminine (irradiated)	0.0	0.05	0.51*	0.57*	0.63*			
Ergocristine	0.50*							
Ergocristine (irradiated)	0.0*	0.15	0.45*	0.50*	0.57	0.62		
Ergotoxine	0.0	0.07	0.17	0.22	0.46*	0.49		
Ergotoxine (irradiated)	0.0*	0.14*	0.20	0.39*	0.45*	0.56	0.66	
Methysergide	0.09*							
Methysergide (irradiated)	0.0*	0.08*	0.10*	0.22				
Ergonovine	0.07*							
Ergonovine (irradiated)	0.0*	0.01*	0.06*	0.08*	0.19	0.50	0.59	
Methylergonovine	0.10*							
Methylergonovine (irradiated)	0.0*	0.02*	0.09*	0.12*	0.24	0.56	0.62	
Ergotamine	0.22*							
Ergotamine (irradiated)	0.0*	0.06*	0.17*	0.22*	0.36	0.55		

[a] Asterisk indicates spot is of equal or greater intensity than 1 μg unirradiated LSD.

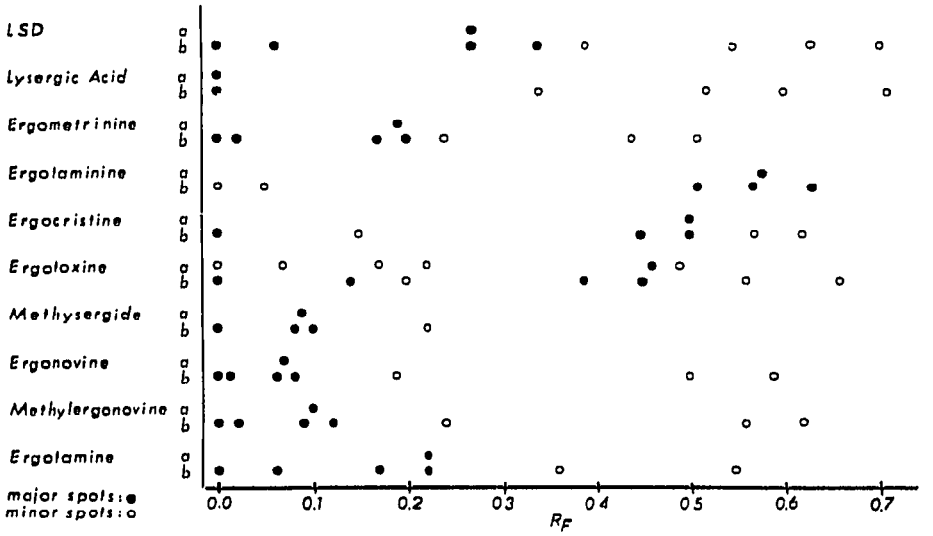

Fig. 1. R_F values of indole alkaloid. (a) Unirradiated. (b) Irradiated. Solid circle is spot of equal or greater intensity than 1 μg unirradiated LSD; open circle is spot of less intensity than 1 μg unirradiated LSD

or greater than 1 μg unirradiated LSD. In Fig. 1 the solid circle respresents these same spots, and the open circle represents the lesser spots. The R_F values given herein represent the average of at least two runs for each. These values have been found to be quite reproducible from day to day with plates and developing solutions made at different times.

The irradiated portion of each of the compounds tested produces a fluorescing spot at the origin, which is one of the major spots in most cases. Many of the other major spots occur in groups of two near the parent spot. None of the degradation products have yet been identified.

These spots vary in intensity depending on degree of decomposition before irradiation, amount spotted and irradiation conditions. The colors are brown, blue, green, white or red. More or fewer minor spots may be seen than those listed in Table I. However, under optimum conditions the principal spots will be observed.

This procedure has the advantages of speed and simplicity combined with the uniqueness of the thin-layer patterns.

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