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Procedure C-08	Thin Layer Chromatography		
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Name of Procedure:

Thin-Layer Chromatography S₁ Developing Solvent

Suggested Uses:

The S₁ Thin-Layer Chromatography Developing Solvent is used to separate and identify organic compounds. Suggested uses of this TLC solvent include opium alkaloids, synthetic opiates, and other organic compounds and drugs.

Apparatus Needed to Perform Procedure Including Preparation of Reagent:

Thin-Layer Chromatography Plates
Silica gel GF - fluorescent indicator

Thickness: 250 microns

Developing tank

Micro pipets

UV light (long and short wave)

Fume hood

Gloves

Eye protection

Laboratory coat

Graduated cylinders

Reagent bottle

Funnel

Porcelain spot plate

Spatula

Dioxane

Ethyl alcohol

Benzene

Toluene

Ammonium hydroxide

Formula for Preparing Reagent:

1. Measure out 5 milliliters of ethanol, 40 milliliters of dioxane, 50 milliliters of toluene, and 5 milliliters of concentrated ammonium hydroxide.

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- 2. Pour measured solvents into reagent bottle and mix well.
- 3. Properly label reagent bottle.

Note: Toluene can be substituted for benzene in the S₁ formulation.

Quality Control Check:

A quality control check of this reagent will be performed using a known standard of heroin and following the application procedure listed below.

Expiration Date of Chemical Reagent:

The solvent can be used until depletion provided it is stored in an airtight reagent bottle.

Application of Procedure on Evidence:

- A sample of the unknown (approximately 1-2 milligrams) is placed in the well of a porcelain spot plate and several drops of a suitable solvent are added to dissolve the sample.
- 2. A known standard (approximately 1-2 milligrams) is also dissolved in the well of a spot plate with several drops of a suitable solvent. (Known standard is selected after visual observation and/or a series of preliminary tests).
- 3. With a capillary pipet, several microliters of the unknown and standards solutions are placed side by side approximately 5 millimeters from the bottom of the TLC plate.
- 4. Add S₁ solvent to the developing tank to a depth of approximately 2 millimeters and allow several minutes for atmosphere to equilibrate.
- 5. Allow all the spotting solvent to evaporate from the TLC plate.
- 6. Place the TLC plate in the developing tank and close the lid.
- 7. Allow the TLC plate to develop to the top of the plate.
- 8. Remove the TLC plate from the developing tank and allow the solvent to dry.

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9. Visualize with UV light or an appropriate visualizing reagent.

Safety Concerns:

Always wear eye protection, gloves and a laboratory coat when preparing this TLC solvent.

Literature References:

Randerath, Kurt, **Thin-Layer Chromatography**, New York, Academic Press, 1968

Moffat, A.C., <u>Clarkens Isolation and Identification of Drugs</u>, 2nd Ed., The Pharmaceutical Press, 1986, pp. 166-177.

Butler, William P., <u>Methods of Analysis</u>, IRS Publication #341, December 1966, pp. 93-95.