Western Regional Laboratory Drug Chemistry Procedure Manual Effective Date: June 19,1997

Name of Procedure:

Thin-Layer Chromatography T₁ Developing Solvent

Suggested Uses:

The T₁ Thin-Layer Chromatography Developing Solvent is used to separate and identify organic compounds. Suggested uses of this TLC solvent include the ergot alkaloids, benzodiazepines, opium alkaloids and other organic compounds.

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 Methanol Concentrated ammonium hydroxide

Formula for Preparing Reagent:

- 1. Measure out 100 milliliters of methanol and 1.5 milliliters of concentrated ammonium hydroxide.
- 2. Pour measured solvents into reagent bottle and mix well.
- 3. Properly label reagent bottle.

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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:

- 1. 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 T₁ 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.
- 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.

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Literature References:

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

Moffat, A.C., <u>Clarke's Isolation and Identification of Drugs</u>, 2nd Ed., The Pharmaceutical Press, 1986.