

**Name of Procedure:**

Thin-Layer Chromatography  
Sulfuric Acid Visualizing Reagent

**Suggested Uses:**

A visualizing reagent or detection reagent must be used in Thin-Layer Chromatography if the compound or compounds are not distinguishable by their own color. This visualizing reagent is used to char compounds that do not react or color with other spray reagents such as sugars and non-nitrogen containing compounds.

**Apparatus Needed to Perform Procedure Including Preparation of Reagent:**

Fume hood  
Graduated cylinder  
Eye protection  
Balance  
Laboratory coat  
Gloves  
Spray bottle  
Air compressor  
Funnel  
Spatula  
Bottles  
Tygon or rubber tubing  
Concentrated sulfuric acid  
Ethanol

**Formula for Preparing Reagent:**

1. Gradually add 10 milliliters of concentrated sulfuric acid to 90 milliliters of ethanol.
2. Place in spray reagent bottle.
3. Properly label spray reagent.

**Expiration Date of Chemical Reagent:**

This spray reagent can be used to depletion.

**Application of Procedure on Evidence:**

1. Place well-dried TLC plate in hood.
2. Activate hood.
3. Using the air compressor and spray bottle, apply a fine mist of the visualizing reagent to the TLC plate.
4. Place the TLC plate in a hot oven and allow time for the charring process to take place.
5. Compare the known standard and the compound in question for their size, shape, color and position on the TLC plate.
6. Record the results of your observation.

**Safety Concerns:**

Always wear eye protection, gloves, and a laboratory coat when preparing this reagent for use.

Eye protection and laboratory coat should be worn when visualizing the TLC plate.

**Literature References:**

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

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