

TLC Stains

Iodine

The plate can be stained with iodine. This can be achieved rapidly, by shaking the plate in a bottle containing silica and a few crystals of iodine. The iodine will stain any compound that reacts with it and so is especially good for visualizing unsaturated compounds. Most spots show up within a few seconds, but the stain is not usually permanent.

UV light

The plate can be viewed under a ultraviolet lamp to show any uv-active spots.

Dipping Solutions:

The plate can be treated with one of the reagents listed below and then heated to stain the spots. The reagent can be sprayed onto the plates, but this technique is quite hazardous and it is more effective for them to be dipped in the reagent. To do this, first let the tlc solvent evaporate, then holding the edge of the plate with tweezers, immerse the plate as completely as possible in the stain and remove it quickly. Rest the edge of the plate on a paper towel to absorb the excess stain before heating carefully on a hot plate or with a heat gun, until the spots show. This method is always permanent and should be done last. When glass plates are used the spots can sometimes be seen more clearly from the glass side of the plate.

p-Anisaldehyde

Preparation: anisaldehyde (15 g) in ethanol (250 ml) + conc. sulfuric acid (2.5 ml).

[Good general reagent, gives a range of colors. Carbohydrate](#)

Bromocresol green: carboxylic acids

Preparation: 0.3% in 1:4 water-methanol

add 8 drops of 30% NaOH/100 mL

[carboxylic acids stain yellow-green on a blue background.](#)

Ceric Ammonium Sulfate Spray

Preparation: 1% cerium (IV) ammonium sulfate (CAS) in 50% phosphoric acid.

[Vinca alkaloids \(Aspidospermas\).](#)

CAM

Preparation: 400ml 10% H₂SO₄ (aq.) , 10g ammonium molybdate , 4g ceric ammonium sulfate

[peptides](#)

Ceric Sulfate (Ce(SO₄)₂)

Preparation: 15% aqueous sulfuric acid saturated with ceric sulfate.

[Fairly general, gives a range of colors.](#)

Distilled Water Spray

Spots turn translucent or opaque while background of plates turns clear.

2,4-DNP

Preparation: 2,4-dinitrophenylhydrazine (12 g) + conc. sulfuric acid (60 ml) + water (80 ml) + ethanol (200 ml).

[Mainly for aldehydes and ketones, gives orange spots.](#)

Dragendorff Reagent

Preparation: Solution A: 1.7 g basic bismuth nitrate in 100 ml water/acetic acid (4:1). Solution B: 40 g potassium iodide in 100 ml of water. Mix reagents together as follows: 5 ml A + 5 ml B + 20 ml acetic acid + 70 ml water. Spray plates, orange spots develop. Spots intensify if sprayed later with HCl, or 50% water-phosphoric acid.

[Good for phenols.](#)

Ferric Chloride Spray

Preparation: 1% Ferric (III) Chloride in Methanol/water (1:1).

[Good for phenols.](#)

Iodoplatinate

Preparation: 10ml 5% PtCl₂ 20ml H₂SO₄ 50ml H₂O

[alkaloids](#)

Ninhydrin

Preparation:

200 mg ninhydrin 95 ml butanol 5 ml 10% AcOH Comments:

[Good for amines.](#)

PDAB-NaNO₂ Reagent - Ehrlichs, Van Urks

Preparation: Solution A: 0.1%p-dimethylaminobenzaldehyde (PDAB) in conc. HCl.

Solution B: 0.1% NaNO₂ (nitrite) in water. Spray plates with PDAB (solution A) first - warm -heat gun, then spray with NaNO₂.

Indoles give blue colors if the 2-position of the indole ring is unsubstituted.

Permanganate (KMnO₄)

Preparation: potassium permanganate (3 g) + potassium carbonate (20 g) + 5% aqueous, NaOH (5 ml) + water (300 ml).

Mainly for unsaturated compounds and alcohols, gives yellow spots.

PMA

Preparation: phosphomolybdic acid (12 g)(in ethanol (250 ml).

Good general reagent, gives blue-green spots.

Sulfanilic Acid Reagent (Diazotized), Pauly's Reagent

Preparation:

Solution A: 0.5% sulfanilic acid in 2% HCl.

Solution B: 0.5% NaNO₂ (nitrite) in water

Solution C: 0.5% NaOH in 50% ethanol

Mix equal volumes of A and B and spray TLC plates. Warm sprayed plate with a heat gun if necessary. Spray plates with solution C.

phenolic compounds turn orange or yellow with this reagent.

Sulfuric Acid

Preparation: 5% sulfuric acid in methanol.

This reagent is usually sprayed on the TLC.

Vanillin

Preparation: vanillin (15 g) in ethanol (250 ml) + conc. sulfuric acid (2.5 ml).

Good general reagent, gives a range of colors.

Others

Ceric Ammonium Nitrate	Ammonia
Vanadium Oxinate	Ferric chloride
Silver-Iodine	Benedict
Formaldehyde-Phosphoric Acid	Cobal chloride
Alizarine	Lead tetracetate
Ferric chloride-Potassium Ferrocyanide	N,N-dimethyl-p-phenyldiamine
Vanillin-KOH	Ferrous sulfocyanate
p-Nitrophenyl diazonium fluoroborate	Diphenylamine-Zinc chloride
Morine	3,5-Dinitrobenzoic Acid
p-aminobenzoic Acid	Kedde
2,3,5-Triphenyltetrazolium	Magnesium acetate
p-Anisidine-Phthalic acid	Ferric Hydroxamate
Antrone	Antimonium Chloride
Naphthoresorcinol	Liebermann-Burchard
Cupric Acetate-Potassium Ferrocyanide	Sodium Nitroprussiate-Sodium Hydroxide