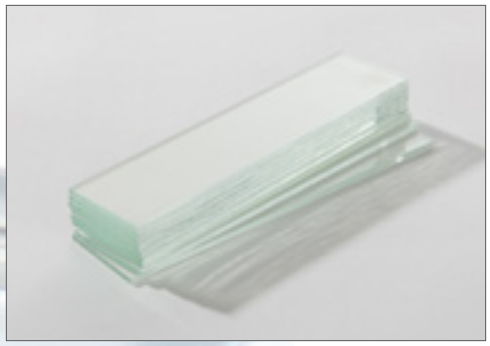
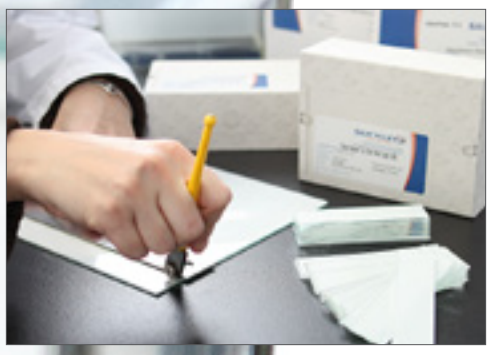




Thin Layer Chromatography

SiliaPlate™ TLC Plates

Thin Layer Chromatography (*TLC*)

SiliCycle is your partner of choice for your purification and chromatography needs

- Optimize your separation conditions by using the same quality silica gel as in your flash columns and cartridges.
- Made with an extra hard layer that ensures the plates don't lose silica upon rubbing and heating.
- The consistent thickness of our Silia*Plate* ensures lot-to-lot reproducibility.

Introduction to Thin Layer Chromatography (TLC)

Thin-layer chromatography (*TLC*) is a quick, simple and inexpensive analytical technique frequently used in various laboratories as it is one of the most verstatile. It is used for:

- Reaction Monitoring
- Screening
- Compound Purity Evaluation

Rapid and cost-efficient selection and optimization of chromatographic conditions prior to flash chromatography purification or HPLC analysis.

Besides speed and low cost, TLC analysis presents other non-negligible advantages like the small quantity of compound required and high sample throughput capability (*up to 20 samples simultaneously*).

Like column chromatography, TLC is a solid-liquid partitioning technique, in which the sample is applied to the plate as a small spot near the base of the plate. The moving liquid phase is then allowed to ascend the plate, causing the sample to partition between moving and stationary phase.

Silia*Plate* Features and Benefits

For more than 20 years, SiliCycle has been offering a wide selection of TLC plates in various sizes (*plate size, thickness, backing*) and chemistries (*10 % Silver Nitrate, CN, C18, NH_2*). Silia*Plate* represents an efficient and economical alternative to other TLC plate manufacturers while demonstrating high separation power, which is due to our narrow particle size distribution silica gel.

The extraordinary silica layer hardness combined to a homogeneous coating and layer thickness allows excellent separation. Each TLC batch is chemically and physically controlled by our Quality Control department to ensure lot-to-lot and layer-to-layer reproducibility.

Types of Plates Available (*TLC / HPTLC / Preparative TLC*)

SiliCycle offers different types of plates for thin-layer chromatography applications: classical TLC, high performance TLC (*also called HPTLC*) and preparative TLC (*PLC*). The plate types are selected based on the type of analysis required and the available budget.

Differences Between Classical TLC, HPTLC and PLC			
Properties	**Classical TLC**	**HPTLC**	**Preparative PLC**
Applications	Quick, inexpensive, flexible and classical separations	Highly sophisticated separation, complex samples	Purification on a TLC plate
Analysis	Qualitative	Qualitative & Quantitative	Quantitative
Detection	UV - Stains	Instrumented analysis (*use of scanners for detection*)	UV
Price	Lower prices than HPTLC	Higher prices than TLC	-
Distribution [*Mean Particle Size*]	5 - 20 µm [*10 - 14 µm*]	4 - 8 µm [*5 - 6 µm*]	5 - 40 µm [*22 - 25 µm*]
Layer Thickness	200 - 250 µm	150 - 200 µm	500 - 2,000 µm
Typical Sample Volume	1 - 5 µL	0.1 - 0.5 µL	5 - 20 µL

TLC Backings

TLC plates are available with different backings (*also called supports*): rigid (*glass-backed*) or flexible sheets (*aluminum & plastic-backed*). Glass-backed plates are the most frequently used due to the ease of handling, transparency (*spot can be seen on both sides*) as well as the chemical resistance and inertness of the support. However, glass plates also present certain disadvantages like fragility and higher weight over flexible backings. On the other hand, aluminum and plastic backings also offer both pros and cons as presented in the table below.

TLC Backings Comparison			
Properties	**Glass**	**Aluminum**	**Plastic**
Advantages	- Rigid - High chemical resistance - High heating stability and charring resistance - Transparent	- Thin - Low weight and consequent shipping costs - High seating stability - Low fragility - Possible to cut with scissors - Can be stored in notebook	- Thin - Low fragility - Possible to cut with scissors - High chemical resistance - Can be stored in notebook
Disadvantages	- Thick - High fragility - Impossible to cut with scissors - Cannot be stored in lab notebook - High weight and consequent shipping costs - Large shelf space	- Low chemical resistance - Opaque	- Medium weight - Opaque - Heating stability up tp 175°C - Possible cracking of matrix due to high flexibility
Approximate Thickness	2.0 - 2.5 mm	1.5 - 2.0 mm	1.5 - 2.0 mm
Total Weight	High	Low	Medium
Heating Stability	High	High	Below 175°C
Fragility	High	Low	Low
Cutting with Scissors	Impossible	Easily	Possible
Chemical Resistance Against			
Mineral Acids	High	Low	High
Bases (*ammoniac*)	High	Low	High

Available Matrices (*or Adsorbents*)

Various adsorbents can be used for TLC coating; silica, aluminum oxide, florisil, etc. However, silica gel is probably the most versatile since it covers almost all types of separation (*if the right solvent system is selected*). More than 80 % of all purifications are performed using silica gel as the adsorbent.

Silica gel

Can be unmodified or functionalized. It is suitable for a very vast array of molecules with various functionalities or polarity, such as aflatoxins, alkaloids, anabolic compounds, barbiturates, carbohydrates, ethers, esters, fatty acids, flavonoids, glycosides, lipids, nucleosides, peptides and proteins, pesticides, sweeteners, vitamins and so on.

Aluminum oxide (*commonly called Alumina*)

Aluminum oxide is the second most commonly used matrix, and it shows similar selectivity to that of silica. Popular applications include the separation for alkaloids, aliphatic compounds, aromatics, steroids, etc. It is manufactured with three different pH ranges: basic, acidic and neutral.

Before use, the plates need to be activated by heating between 90° and 120°C for 10 minutes (*since water molecules are easily adsorbed and can greatly influence separation*).

Cellulose

Cellulose can be unmodified or positively charged at acidic and neutral pH. This adsorbent is hence frequently used for the partition of hydrophilic molecules and is also useful for challenging separations of sensitive biomolecules or molecules carrying ion exchange groups. The ratio of charged cellulose / unmodified cellulose can be varied, in order to provide more or less retention of negatively charged molecules.

Available Sorbents

Classical Silica Gel: for daily, fast, reliable analysis of the largest spectra of molecules

The particle size distribution used for the silica is related to the nature of the plate. For standard TLC, silica gel with a mean particle size of 10 - 14 µm is used compared to HPTLC where a smaller particle size is required. In both cases, pore diameter is always 60 Å.

Reversed & Special Phases

The two most popular modes of separation employed in TLC are normal and reversed phases. In normal phase separation, the mobile phase is less polar than the stationary phase. Inversely, in reversed mode, the mobile phase (*usually a mixture of water and organic solvent*) is more polar than the stationary phase (*C18*).

Functionalized silica gels can also be used as TLC adsorbents for particular needs when satisfactory separations cannot be achieved by unmodified silica. They are mostly used as pilot methods for ulterior HPLC analysis. Here are some typical issues that can affect separation and can be solved using functionalized phases:

- Aqueous solvent systems
- Ambient humidity
- Direct HPLC correlation
- Degradation of sensitive molecules (*oxidation, hydrolysis, etc.*)

Reversed-phases TLC plates include C2, C8 and C18 phases where functionalization of silica is performed using organosilanes of various chain lengths. Retention of molecules and the ability to tolerate water in the moving phase are directly dependent on the chain length: the shorter the chain, the more water tolerant it is and hence the shorter the migration time will be.

Special phases such as Diol and Nitrile (*CN*) are moderately polar. They can thus be suitable for both normal and reversed phase chromatography, depending on your application. Amino phases (NH_2) have specifically been designed for charged compounds, as they show weak anion exchange characteristics.

Layer Thicknesses

The layer thickness is related to the nature of the analysis (*analytical or preparative*) as well as the performance of the plate (*TLC or HPLTC*). The most common layer thicknesses are:

- 150 - 200 µm (*HPTLC plates*)
- 200 - 250 µm (*analytical TLC plates*)
- 500 - 2,000 µm (*preparative TLC plates*)

Binder & UV Indicator

All standard Silia*Plate* products are made with a Gypsum binder and have an UV indicator (*F254*). Contact us for custom products.

Plate Sizes

Silia*Plate* TLC plates are available in the following standard sizes depending on the coating used:

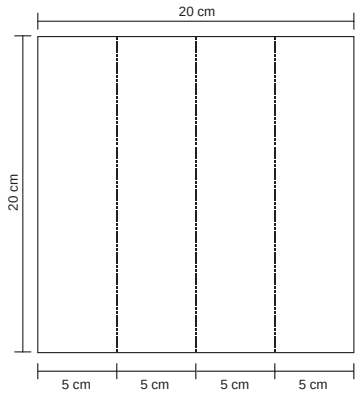
- 20 x 20 cm
- 10 x 20 cm
- 5 x 20 cm
- 5 x 10 cm
- 10 x 10 cm

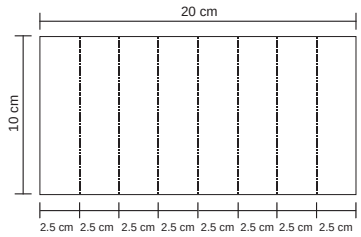
Also for your convenience, SiliCycle provides ready to use micro TLC plates in the following formats:

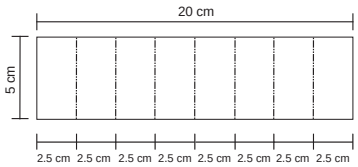
- 2.5 x 10 cm
- 2.5 x 7.5 cm
- 2.5 x 5 cm

An interesting compromise between standard and micro plate sizes is our Scored Silia*Plate* (*glass backing*). Three different formats are available and possible cut combinations are shown in the image below.

- 20 x 20 cm plates scored to four 5 x 20 cm plates (*or multiple of 5 cm width*)
- 10 x 20 cm plates scored to eight 2.5 x 10 cm plates (*or multiple of 2.5 cm width*)
- 5 x 20 cm plates scored to eight 2.5 x 5 cm plates *(or multiple of 2.5 cm width)*







« Many products have been successfully purified with the silica gel. We have had problems with other companies' TLC plates not running the same as their silica gel, but everything was fixed when we switched over to all SiliCycle products. »

William Nguyen from Stanford University, Stanford, CA, USA

Silia*Plate* TLC Plates Portfolio

SiliCycle offers the possibility to analyze reactions on thin layer chromatography support and rapidly develop optimized purification conditions for efficient transfer to flash columns. Maximize the benefits by using our *UltraPure* Silia*Plate* TLC plates with an extra hard layer of silica. For your convenience, SiliCycle offers different sizes, choice of backings, reversed-phase & specialty plates. Contact us for more information.

Various combinations are possible with Silia*Plate* TLC plates and are summarized in the table below.

Silia*Plate* TLC Plates Portfolio			
Properties	**Analytical**	**HPTLC**	**Preparative**
Available Backings			
Glass	Yes	Yes	Yes
Aluminum	Yes	No	No
Plastic	Yes	No	No
Available Adsorbents			
Bare silica	Yes	Yes	Yes
Functionalized Silica	No	Yes	Yes
Silica Specifications			
Mean Particle Size	10 - 14 µm	5 - 6 µm	22 - 25 µm
Mean Pore Diameter	60 Å	60 Å	60 Å
Type of Plate Available			
Scored Plate	Yes	No	Yes
Channeled Plate	Yes	No	No
Layer Thickness	Glass: 250 µm Flexible: 200 µm	Glass: 150 - 200 µm	Glass: 500 µm & 1,000 µm Flexible: 1,500 µm & 2,000 µm
Plate Size*	2.5 x 5 cm; 2.5 x 7.5 cm; 2.5 x 10 cm; 5 x 10 cm; 5 x 20 cm; 10 x 20; 20 x 20 cm	2.5 x 5 cm; 2.5 x 7.5 cm; 2.5 x 10 cm; 5 x 10 cm; 5 x 20 cm; 10 x 20; 20 x 20 cm	20 x 20 cm

*For the glass-backed TLC plates.

Two Types of Glass-Backed, 20 x 20 cm, TLC Plates

SiliCycle offers two types of Silia*Plate* glass-backed 20 x 20 cm TLC Plates, with different sensitivities and areas of applications.

The difference between the two plates is in the binder chemistry:

- **TLG-R10014B-323** 's layer is polymeric: it has been added a small percentage of inorganic, hardening agent for a uniform and hard surface, smooth and dense, that will not crack, blister nor swell up. They were designed for maximum robustness of the binder: they are very easy to handle and to write on, as well as completely wettable.
  They are compatible with all solvents, yet, they might oxidize a bit faster when dipped into $KMnO_4$ (*fading in a few minutes from flashy purple to yellow ocher*). Also, spots are a bit less definite when using CAM as a revelatory.
  Such binder also contains a higher percentage of fluorescent indicator for greater brilliance of spots and less background noise from the silica layer.
- **TLG-R10014BK-323** 's layer is gypsum (*calcium sulfate*), and do not contain the polymeric additive that provides the former plates a harder surface and ruggedness. This means that the layer is softer, so spots can be easily scrapped off from the glass support, and are particularly recommended for aggressive visualization methods (*strong charring, CAM staining solution*) or, if dipped into $KMnO_4$, ought to remain bright-purple a longer period of time.

Here is a chart which can hopefully help you quiclky choose the right plate for your specific application.

General View of Specificities & Characteristics to be Considered		
TLC Plate PN	**TLG-R10014B-323**	**TLG-R10014BK-323**
UV Fluorescence (F_{254})	• Higher brightness • Less background noise from layer	• Yes
Binder Sensitivity	Stable in almost all organic solvents	
	• Increased separation efficiency	• Resistant to aggressive visualization methods
Surface Layer	• Robust and rugged	• Easily scratched off
Water Tolerance	• Up to 80 %	• Up to 40 %
Specific Surface (*BET*)	≈ 500 m^2/g	
Mean Pore Size	60 Å	
Mean Pore Volume	0.75 mL/g	
Distribution (*Mean Particle Size*)	5 - 20 µm [10 - 14 µm]	
Layer Thickness	≈ 250 µm	
Stain Compatibility		
$KMnO_4$	Compatible	**Highly compatible**
CAM	Compatible	
p-Anisaldehyde	Compatible	**Highly compatible**
Ninhydrin	**Highly compatible**	
Vanilin	**Highly compatible**	

Silia*Plate* Ordering Information

All our plates bear an F_{254} UV indicator for direct visualization of results or derivatization, but all can be available with no UV indicator. A long-wavelength (F_{366}) UV indicator is also available upon request.

Please note that this is an overview of plates that SiliCycle offers. Different sizes are available, as well as more exotic layers for special separations (chiral layers, layers for surfactant separations, for PAH analysis, layers for basic or acidic ion exchange, cellulose layers, etc.). Contact us.

CLASSICAL TLC Plates Portfolio

GLASS Silia*Plate* TLC				
SiliCycle PN	**Product Name**	**Plate Size (*cm*)**	**Thickness (*µm*)**	**Qty / Box**
Analytical Silia*Plate* Glass				
TLG-R10014B-417	Micro Silia*Plate* Glass	2.5 x 5	250	200
TLG-R10014B-124	Micro Silia*Plate* Glass	2.5 x 7.5	250	100
TLG-R10011B-624	Micro Silia*Plate* Glass	2.5 x 10	250	100
TLG-R10011B-527	Silia*Plate* Glass	5 x 10	250	200
TLG-R10011B-424	Silia*Plate* Glass	5 x 20	250	100
TLG-R10011B-723	Silia*Plate* Glass	10 x 20	250	25
TLG-R10014B-323	Silia*Plate* Glass, Extra Hard Layer, Increased UV Content	20 x 20	250	25
TLG-R00014BK-323	Silia*Plate* Glass, Optimized Layer for $KMnO_4$ Revelation	20 x 20	250	25
Scored Analytical Silia*Plate* Glass				
TLGSR10011B-723	Silia*Plate* Glass (*scored to 2.5 x 10*)	10 x 20	250	25
TLGSR10011B-423	Silia*Plate* Glass (*scored to 2.5 x 5*)	5 x 20	250	25
TLGSR10011B-424	Silia*Plate* Glass (*scored to 2.5 x 5*)	5 x 20	250	100
TLGSR10011B-323	Silia*Plate* Glass (*scored to 5 x 20*)	20 x 20	250	25
Channeled Analytical Silia*Plate* Glass *(with Preadsorbent Zone)*				
TLGCZ-R10011B-323	Channeled Silia*Plate* Glass (*w/ PreAd.*)	20 x 20	250	25
TLGCZ-R10011B-723	Channeled Silia*Plate* Glass (*w/ PreAd.*)	10 x 20	250	25
TLGCZ-R10011B-423	Channeled Silia*Plate* Glass (*w/ PreAd.*)	5 x 20	250	25

ALUMINUM Silia*Plate* TLC Plates				
SiliCycle PN	**Product Name**	**Plate Size (*cm*)**	**Thickness (*µm*)**	**Qty / Box**
Silia*Plate* Al *(Aluminum)*				
TLA-R10011B-124	Micro Silia*Plate* Aluminum	2.5 x 7.5	200	200
TLA-R10011B-515	Silia*Plate* Aluminum	5 x 10	200	50
TLA-R10011B-415	Silia*Plate* Aluminum	5 x 20	200	50
TLA-R10011B-712	Silia*Plate* Aluminum	10 x 20	200	20
TLA-R10011B-323	Silia*Plate* Aluminum	20 x 20	200	25
TLA-R10011B-323N	Silia*Plate* Aluminum (*no UV*)	20 x 20	200	25
Silia*Plate* Al C18 *(Aluminum)*				
TLA-R30411B-303	Silia*Plate* Aluminum C18	20 x 20	150	25

PLASTIC Silia*Plate* TLC Plates				
SiliCycle PN	**Product Name**	**Plate Size (*cm*)**	**Thickness (*µm*)**	**Qty / Box**
Silia*Plate* Pl (*Plastic*)				
TLP-R10011B-2575	Micro Silia*Plate* Plastic	2.5 x 7.5	200	200
TLP-R10011B-323	Silia*Plate* Plastic	20 x 20	200	25

HPTLC TLC Plates Portfolio

BARE Silia*Plate* HPTLC Plates with Glass Backing (*Thickness: 150 microns, 25 plates / Box*)			
SiliCycle PN	**Plate Size (*cm*)**	**SiliCycle PN**	**Plate Size (*cm*)**
Silia*Plate* Silica HPTLC			
HPTLG-R10011B-1010	10 x 10	HPTLG-R10011B-2020	20 x 20
HPTLGSR10011B-1010	10 x 10 (*scored to 5 x 5 cm*)	HPTLGSR10011B-1020	10 x 20 (*scored to 2.5 x 10 cm*)

FUNCTIONALIZED Silia*Plate* HPTLC with Glass Backing (*25 plates / Box*)				
SiliCycle PN	**Plate Size (*cm*)**	**SiliCycle PN**	**Plate Size (*cm*)**	**Thickness (*µm*)**
Silia*Plate* REVERSED-PHASE MODIFIED HPTLC				
Silia*Plate* C18 HPTLC				
TLG-R30414BK-213	10 x 10	TLG-R30414BK-313	20 x 20	200
Silia*Plate* C8 HPTLC				
TLG-R31014BK-203	10 x 10	TLG-R31014BK-303	20 x 20	200
Silia*Plate* C2 HPTLC				
TLG-R32614BK-713	10 x 20	TLG-R32614BK-313	20 x 20	200
Silia*Plate* NORMAL-PHASE MODIFIED HPTLC				
Silia*Plate* NH_2 *(Amine)* HPTLC				
TLG-R52014BK-213	10 x 20	TLG-R52014BK-313	20 x 20	200
Silia*Plate* CN *(Cyano)* HPTLC				
TLG-R38014BK-213	10 x 10	TLG-R38014B-313	20 x 20	200
Silia*Plate* Diol HPTLC				
TLG-R35014BK-213	10 x 10	TLG-R35014BK-313	20 x 20	200

SPECIALITY SORBENTS SiliaPlate TLC Plates with Glass Backing (25 plates / Box) *				
SiliCycle PN	Plate Size (cm)	SiliCycle PN	Plate Size (cm)	Thickness (µm)
SiliaPlate Ag (*Silver Nitrate 10 % impregnated*) TLC				
TLG-R23511B-423	5 x 20	TLG-R23511B-303	20 x 20	250
TLG-R23511B-433	5 x 20	TLG-R23511B-333	20 x 20	500
SiliaPlate Ag (*Silver Nitrate 15 % impregnated*) TLC				
TLG-R23611B-423	5 x 20	TLG-R23611B-323	20 x 20	250
TLG-R23611B-433	5 x 20	TLG-R23611B-333	20 x 20	500
SiliaPlate Ag (*Silver Nitrate 20 % impregnated*) TLC				
TLG-R23711B-423	5 x 20	TLG-R23711B-323	20 x 20	250
TLG-R23711B-433	5 x 20	TLG-R23711B-333	20 x 20	500
SiliaPlate Aluminum Oxide (*Neutral*) TLC				
TLG-AUT0337-423N	5 x 20	TLG-AUT0337-323N	20 x 20	250
TLG-AUT0337-433N	10 x 20	TLG-AUT0337-333N	20 x 20	500
SiliaPlate Cellulose TLC (*Contact us for a specific ratio of charged cellulose / unmodified cellulose to suit your application*)				
TLG- AUT0307-423	5 x 20	TLG- AUT0307-323	20 x 20	250

* Scored plates are also available, please contact us for dimension inquiry.

PREPARATIVE TLC Plates Portfolio

PREPARATIVE TLC Plates Portfolio			
SiliCycle PN	Plate Size (cm)	Thickness (µm)	Qty / Box
Preparative SiliaPlate Prep (*Glass Preparative*)			
TLG-R10011B-333	20 x 20	500	25
TLG-R10011B-341	20 x 20	1,000	25
TLG-R10011B-363	20 x 20	1,500	25
TLG-R10011B-353	20 x 20	2,000	25
Scored SiliaPlate Prep (*Glass Preparative*)			
TLGSR10011B-333	20 x 20 (*scored to four 5 x 20*)	500	25
TLGSR10011B-341	20 x 20 (*scored to four 5 x 20*)	1,000	25
TLGSR10011B-363	20 x 20 (*scored to four 5 x 20*)	1,500	25
TLGSR10011B-353	20 x 20 (*scored to four 5 x 20*)	2,000	25
SiliaPlate Prep C18 (*Glass Preparative*)			
TLG-R30411B-341	20 x 20	1,000	25

TRIAL PACKAGES

Trial Package of Functionalized SiliaPlate TLC Plates with Glass Backing (5 plates of each / Box) *			
SiliCycle PN	Plate Size (cm)	Composition	Thickness (µm)
TLGSR1234511B-723	10 x 20 (*scored to 2.5 x 10 cm*)	C18, C8, C2, NH_2 & CN	250

* Other scored plates are also available, please contact us for dimension inquirie.

Silia*Plate* TLC Accessories

Silia*Plate* TLC Developing Chamber

The most commonly used accessory to develop a TLC plate.

AUT-0161	Silia*Plate* Rectangular TLC Developing Chamber

Other Silia*Plate* TLC Accessories

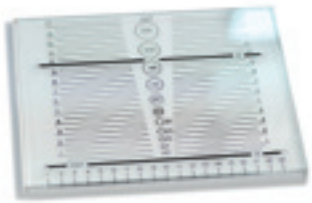
AUT-0162	Silia*Plate* TLC Scraper
AUT-0163	Silia*Plate* TLC Spotting Capillary Tubes
AUT-0164	Silia*Plate* TLC Spotting Guide
AUT-0182	TLC Cutter for Silia*Plate* (*up to 20 x 20 cm*)
AUT-1182	Pencil Glass Cutter for Silia*Plate*
AUT-0183	Replacement Scriber for Silia*Plate* TLC Cutter

PN: AUT-1182

PN: AUT-0162

PN: AUT-0161

PN: AUT-0182



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An Ideal Partnership Between SiliCycle and AR2i

SiliCycle has entered into an exclusive strategic specialty worldwide distribution partnership with the company AR2i specialized in the conception and the manufacturing of innovative devices in the field of Thin-Layer Chromatography.

Chromimage® Documentation

- Perform qualitative analysis on TLC plates in a few minutes.
- Detect and numerize your TLC plates under UV254 nm and visible mode.
- Classify and archive your TLC analyses under several storage formats (*jpg, eps, pdf, etc.*).
- Suitable for reading 10 x 10 cm, 10 x 20 cm and 20 x 20 cm plates.

Derivapress® System

It's as simple as opening and closing a book: the Derivapress immersion derivatization system provides a cost-effective, efficient and safe alternative to perfect this essential stage of TLC and to move towards densitometric measurements like quantitative and semi-quantitative TLC.

Furthermore Derivapress complies with the GLP requirements and can be used in 21 CFR Part 11 work environments.

PN: AUT-0165

PN: AUT-0166

Thin Layer Chromatography Practical Guide

Select a Stationary Phase

As almost 80 % of all separations can be performed using **silica gel plates**, it is suggested to try using this coating first. However, for acid sensitive compounds, alumina is probably a better choice (*useful for amine purification*). If you are working with highly polar compounds, reversed-phase mode is more suitable.

Select a Mobile Phase (*Solvent Systems*)

The selection of the mobile phase (*also called solvent system or eluent*) is perhaps the most important parameter to achieve efficient thin-layer chromatography separation. It is based on the compound's solubility with the solvent and the difference in the affinity for the mobile phase versus the stationary adsorbent (*silica, alumina or cellulose*).

In normal phase chromatography, where non-polar solvents such as hexane or pentane are used, non-polar compounds will move up the plate while most polar compounds will stay on the baseline. Inversely, polar solvents will allow polar compounds to move off the origin. The most suitable solvent system is the one that moves all components off the baseline with Rf values between 0.15 and 0.85 (*ideally, close to 0.2 - 0.4*).

For most applications, a common solvent system to start with is **EtOAc / Hexane (*1:1*)**. Varying the ratio can have a pronounced effect on the Rf. If it is not working, then try: MeOH / DCM (*2:8 - 10:90*); or toluene with acetone, EtOAc, or DCM.

Remember: in normal phases, to increase the compound's Rf, increase the polarity of the mobile phase; increase the ratio of the polar solvent or choose another solvent. Inversely, to decrease Rf, decrease the polarity of the eluent.

Rules of Thumb

- Standard compounds (*most popular solvent system*): 10 - 50 % EtOAc / Hexane
- Polar compounds: 100 % EtOAc or 5 - 10 % MeOH / DCM
- Non-polar compounds: 5 % EtOAc (*or ether*) / Hexane or 100 % Hexane
- For basic compounds: (*amine or nitrogen containing*), it could be useful or required to add a small quantity of triethylamine (Et_3N) to the solvent mixture (*0.1 - 2.0 % but typical quantity is 0.1 %*) or 1 - 10 % ammonia (NH_3) in MeOH / DCM.
- For acidic compounds: it could be useful to add acetic (*AcOH*) or formic acid (*FA*) to the solvent mixture (*0.1 - 2.0 %*).

Reversed-phase mode

In reversed-phase chromatography, the typical solvent systems are:

- Mixtures of water or aqueous buffers and water miscible organic solvents such as acetonitrile (*ACN*), methanol and tetrahydrofuran (*THF*). Other solvents can be used such as ethanol (*EtOH*) & isopropanol (*IPA*).
- MeOH, to improve peak shape in flash chromatography, 0.1 % of acetic, formic or trifluoroacetic acid (*TFA*) can be added to the solvent system.

« Have given your products to other folks within organisation and used it myself with great success (both the Prep SPE, HPLC columns, TLC plates and silica gel). »

Kerry M. Keertikar, Merck Research Labs, Kenilworth, NJ, USA

TLC Plate Preparation

Using a pencil, lightly draw a straight-line parallel to the width of the plate at about 1 cm from the base end of the plate. Sample application will be done on this line called baseline or origin.

Note: never use a pen because ink can move with some solvents used as eluent.

Sample preparation

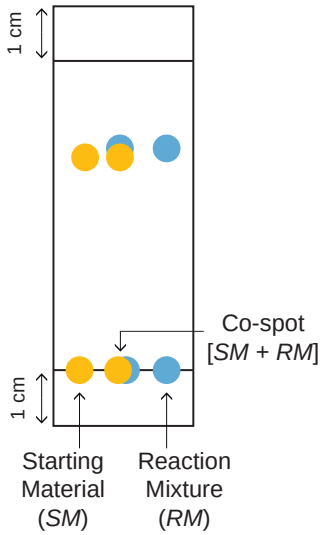
Thorough sample preparation is a prerequisite for an optimal and efficient TLC separation. Typical sample preparation processes could consist in a sample crushing, filtration, extraction or concentration of the product of interest.

Sample Application

Sample preparation will differ depending on the nature of the plate (*analytical or preparative*). For analytical plates, because thin layer chromatography is extremely sensitive, it is really important to apply a small quantity using a glass capillary (*or a micro pipette*) to get optimal resolution. For preparative plates, apply a series of small adjacent spots to form a band or a streak using a glass capillary (*or a microliter syringe*). In both cases, a spotting guide can be used to facilitate sample application.

Co-spotting

For analytical chromatography, co-spotting is frequently used for similar polarity products. This consists to apply on the same spot, the starting material and reaction mixture as shown by the image below.



TLC Plate Development

The most commonly used method to perform thin layer chromatography separation is to place vertically the TLC plate inside a sealed developing chamber to ensure solvent saturation. Place approximately 0.5 cm of the suitable solvent system inside the chamber. Slowly place the TLC inside the chamber and allow the eluent to travel up the plate until it gets to 1 cm from the top of the plate. Immediately remove the plate and draw a line along the solvent front.

***Note**: for optimal solvent saturation, a filter paper can be added inside the TLC chamber. This also prevents eluent evaporation. The solvent level needs to be below the baseline; otherwise the spots will be dissolved.*

TLC Plate Visualization

If components of the reaction are colored, no visualization method is required (*spots can be seen directly on the silica layer*). However, most of the time it is not the case, therefore one of the methods described below should be used to reveal the spots.

Non-destructive methods

As a general visualization procedure, before treating the TLC plate with any destructive methods, UV-active compounds can be viewed under an ultraviolet lamp (*usually for polyconjugated compounds like benzophenones and anthracenes*). Furthermore, an iodine chamber can be useful for thiols, phosphines and alkenes but it works in about 50% of cases for alkanes. It is recommended to circle the spots with a pencil on the TLC plate prior to visualization by destructive methods.

Destructive methods

For compounds that are not UV-active, there are several varieties of stains that can be used depending on the nature of the compound of interest. To use a stain, simply dip the TLC plate into the staining solution as quickly as possible, and then immediately absorb the excess stain with paper and heat carefully with a heat gun or on a hot plate at 110°C until spots are revealed. See next pages.

Chromatogram Interpretation

Retention factor (*Rf*) definition

Retention factor analysis is used to evaluate if the solvent system is adequate. Rf is defined as the distance traveled by the compound divided by the distance traveled by the solvent front. This means: the larger the Rf value of a compound, the larger is the distance traveled by the compound. In other words, when comparing Rf values of various compounds under identical chromatography conditions, the compound with the larger Rf is less polar because it interacts less strongly with the polar adsorbent on the plate.

Remember, a good solvent system is one that moves all components off the baseline with Rf values between 0.15 and 0.85 (*ideal Rf is 0.2 - 0.4*). Otherwise, when possible, it is preferable to chose another solvent system.

$$\text{Retention factor } (Rf) = \frac{\text{distance traveled by the compound}}{\text{distance traveled by the solvent front}}$$

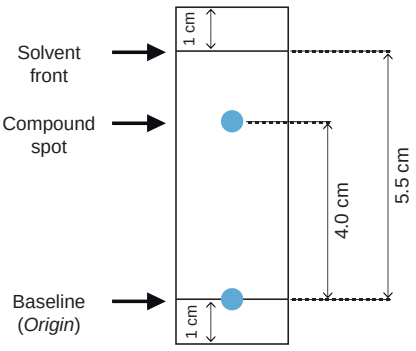
Rf calculation based on the example shown here:
Rf = 4.0 cm / 5.5 cm = 0.73

Prediction of Column Volumes (*CV*)

TLC data can be used to predict column elution based on the relationship between the retention factor and the column volume. CV is the number of column volumes required to elute the component from the column regardless of column dimensions [*(bed volume) - (volume of packing)*].

$CV = 1 / Rf$ & $\Delta CV = 1 / Rf_1 - 1 / Rf_2$

The greater the ΔCV, the greater will be the separation and resolution between the spots (easier separation).
A bigger ΔCV will therefore allow more sample to be loaded onto the column.



TLC Tool App

NEW

TLC has become as easy as taking a picture!

This mobile application is the perfect tool to help you save time for real chemistry issues: it will automatically calculate Rfs, store the TLC picture in your phone before the spots fade away (*so you can eventually print it or share it with other lab members*), it can even notify you if the mobile phase travels up to far and reaches the top of the plate !

Please find more information visiting us at:
http://www.silicycle.com/tlcapp

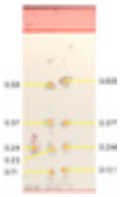
Features

AUTO CROP

- Crops TLC from photo automatically
- Removes things you don't need

AUTO DETECTION
AUTO CALCULATE RF

- Detects spots automatically
- Calculates Rf without need for ruler nor calculator



SMART STORAGE

- TLC is hard to store or to find when needed
- Plus, spots eventually fade away or the layer oxidizes, making pencil marks hard to detect
- Your TLC plates are now just a click away in your phone!

SMART TIMER

- Automatically detects if colors in the specific area change over time

Plus, you can have access to all SiliCycle's offering of TLC Plates, from the most standard ones to most exotic layers.

Described below are the most frequently used TLC visualization methods (*also called stains*) in alphabetical order.

Stains for Thin Layer Chromatography

Name	Visualization of...	Stain Recipe	Comments
p-Anisaldehyde #1	**Universal stain** *Good for nucleophiles and oxygenated compounds*	**Prepare stain as follows** • 2 mL of glacial acetic acid • 5 mL of p-anisaldehyde • 7 mL of conc. sulfuric acid • 185 mL of 95 % ethanol ***Tip*: *Add dropwise the acid at the end and stir vigorously.***	**Visualization Colors** • Spots: Various colors • BG: Orange to pink **Appropriate Storage** • Aluminum wrapped at 0°C
Note: Tends to be insensitive to alkenes, alkynes and aromatic compounds unless other functional groups are present.			
p-Anisaldehyde #2	*Acronycine* *Cineoles* *Terpenes*	**Prepare stain as follows [*1:10:20:80*]** • p-anisaldehyde • perchloric acid • acetone • water	**Visualization Colors** • Spots: Various colors • BG: Orange to pink **Appropriate Storage** • Aluminum wrapped at 0°C
Bromocresol Green	*Acidic groups (pK_a < 5)* *Carboxylic acids*	**Prepare stain as follows** • 0.04 g of bromocresol green • 100 mL of 95 % ethanol • 0.1 M solution of sodium hydroxide ***Tip*: *Add the base slowly at the end until the solution turns pale blue.***	**Visualization Colors** • Spots: Yellow to green • BG: Blue **Appropriate Storage** • Aluminum wrapped at 0°C ***Heating NOT required***
Cerium Molybdate (*CAM or Hanessian's Stain*)	**Universal stain** *Good for peptides*	**Prepare stain as follows** • 12 g of ammonium molybdate • 0.5 g of ceric ammonium molybdate • 15 mL of conc. sulfuric acid • 235 mL of water	**Visualization Colors** • Spots: Blue • BG: White **Appropriate Storage** • Aluminum wrapped
Note: Highly sensitive stain; very low concentration of product may appear as a significant impurity.			
Cerium Sulfate ($Ce(SO_4)_2$)	**Difficultly stainable compounds**	**Prepare stain as follows** • 15 % aqueous sulfuric acid saturated with ceric sulfate	**Visualization Colors** • Spots: Black • BG: Yellow to white
Chromic Acid	**Difficultly stainable compounds**	**Prepare stain as follows** • 2.5 g of potassium chromate • 100 mL of 20 % sulfuric acid in water	
Cobalt Chloride ($CoCl_2$)	**Universal stain** *Used in conjunction with PMA when this one is not effective enough*	**Prepare stain as follows** • 2 g of cobalt chloride • 100 mL of water • 10 mL of conc. sulfuric acid ***Tip*: *Simply dip PMA treated plate in $CoCl_2$ solution.***	**Visualization Colors** • Spots: Various colors • BG: Pink ***Heating NOT required***
p-Dimethylamino-benzaldehyde (*PDAB or Ehrlich's Reagent*)	*Amines* *Indoles*	**Prepare stain as follows** • 0.5 g of p-dimethylamino-benzaldehyde • 10 mL of conc. hydrochloric acid • 40 mL of acetone *(or 95 % ethanol)*	**Visualization Colors** • Spots: Blue • BG: White

N.B. Shaded lines refer to "Universal stains"

Stains for Thin Layer Chromatography (*Con't*)

Name	Visualization of...	Stain Recipe	Comments
2,4-Dinitrophenyl-hydrazine (*DNP*)	*Aldehydes* *Ketones*	**Prepare stain as follows** • 12 g of 2,4-dinitrophenylhydrazine • 60 mL of conc. sulfuric acid • 80 mL of water • 200 mL of 95 % ethanol	**Visualization Colors** • Spots: Yellow to red • BG: Light orange ***DO NOT HEAT dipped plate***
Dragendorff Reagent	**Nitrogenous Compounds** *Alkaloids, amines, organics bases, etc.* **Phenols**	**Prepare stain as follows** **Solution A** • 1.7 g of bismuth nitrate • 80 mL of water • 20 mL of acetic acid **Solution B** • 40 g of potassium iodide • 100 mL of water ***Tip:*** *mix 5 mL of each solution A and B to a solution of 20 mL of acetic acid in 70 mL of water.*	**Visualization Colors** • Spots: Orange to red • BG: Yellow **Appropriate Storage** • Aluminum wrapped **Stain Shelf-Life** • One or two weeks • Solutions A and B are long term storable ***DO NOT HEAT dipped plate***
Ferric Chloride ($FeCl_3$)	*Phenols*	**Prepare stain as follows** • 2 g of ferric chloride • 102 mL of 0.5 N hydrochloric acid	**Visualization Colors** • Spots: Red • BG: Yellow
Iodine	*Unsaturated & Aromatic compounds*	**Prepare stain as follows** • Iodine crystals in an amber bottle	**Visualization Colors** • Spots: Dark brown • BG: Light brown
Note: iodine stain can be removed by heating.			
Morin Hydrate (*Hydroxy Flavone*)	**Universal stain** *Fluorescently active*	**Prepare stain as follows** • 0.1 % of morin hydrate in methanol	**Visualization Colors** • Spots: Various colors • BG: White
Ninhydrin (*Indanetrione Hydrate*)	*Amino acids* *Amino sugars* *Amines*	**Prepare stain as follows** • 1.5 g of ninhydrin • 3 mL acetic acid • 100 mL of n-butanol	**Visualization Colors** • Spots: Various colors • BG: White
Phosphomolybdic Acid (*PMA*)	**Universal stain** *Very effective against diluted sample*	**Prepare stain as follows** • 10 % of PMA solution in ethanol • or 10 g of PMA in 100 mL of ethanol	**Visualization Colors** • Spots: Dark green to black • BG: Light green
Potassium Permanganate ($KMnO_4$)	*Olefins* *Readily oxidized groups* *Alcohols, aldehydes, alkenes, alkynes, etc.*	**Prepare stain as follows** • 1.5 g of potassium permanganate • 10 g of potassium carbonate • 1.25 mL of 10 % sodium hydroxide • 200 mL of water	**Visualization Colors** • Spots: Yellow to light brown • BG: Purple to pink **Stain Shelf-Life** • Three months
Note: Can be used for detection of alcohols, amines, sulfides and mercaptans groups when gently heated.			
Vanillin	**Universal stain** *Very effective for same polarity products (Rf)*	**Prepare stain as follows** • 15 g of vanillin • 250 mL of 95 % ethanol • 2.5 mL of conc. sulfuric acid	**Visualization Colors** • Spots: Various colors • BG: Light tan

Note: Occasionally, spots can be seen more clearly from glass side with glass backed TLC plate. Otherwise mentioned, stains are long-term stable when stored in a tightly-closed container to prevent solvent evaporation.
"BG" stands for "background".
N.B. Shaded lines refer to "Universal stains"

Silia*Plate* TLC Troubleshooting

Problem: Streaking or elongated spot rather than a defined spot?

Possible Solutions:

- Sample was overloaded: run the TLC again using a more diluted solution of your sample.
- In presence of a base sensitive compound: try to add acetic or formic acid to the eluent (*0.1 - 2.0 %*).
- In presence of an acid sensitive compound: try to add triethylamine to the eluent (*0.1 - 2.0 %*) or 1 - 10 % ammonia in MeOH / DCM. If it is not working use Alumina as TLC coating.
- In presence of too highly polar compounds: try using a specialized silica TLC plate like reversed-phase (*C18 for example*).

Problem: Unable to see any spots on the TLC?

Possible Solutions:

- If you have not been able to visualize any spots on your TLC using UV light, try another method; maybe your compound is not UV-active.
- Maybe your sample is too diluted. Try to apply several times your sample on the same spot (*do not forget to dry solvent between each application for optimal results*) or to concentrate your solution.
- Make sure the solvent level inside the tank is lower than the spotting line to avoid sample dissolution by the eluent.

Problem: How to monitor a reaction in presence of similar Rfs for both starting materials and product of interest?

Possible Solutions:

- Try the co-spotting method (*see page 272*).
- Try to visualize the plate using anisaldehyde or molybdene. Spot color or brightness differ for two compounds when using these stains.
- If none of the two previous solutions work, change solvent systems (*use another class of solvent*).

Tips: in chromatography, there are three classes of solvent systems providing significantly different results:

1: Mixture of polar / hydrocarbon solvents (*i.e.: EtOAc / Hexane; Ether / Petroleum ether*).

2: Mixture of polar / dichloromethane solvents (*examples of polar solvent: Ether, EtOAc, MeOH*).

3: Mixture of polar / benzene (*or toluene*) solvents (*examples of polar solvent: Ether, EtOAc, MeOH*).

Problem: Compounds stay too close to the baseline or solvent front.

Possible Solutions:

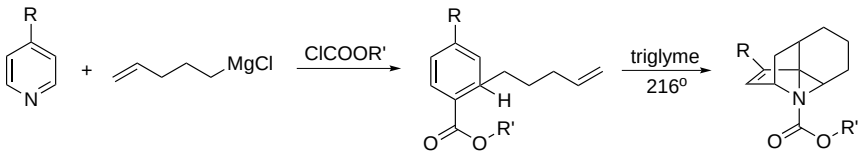
- Too close to the baseline: your eluent is not polar enough; increase the proportion of polar solvent in the same solvent system or chose a more polar solvent.
- Too close to the solvent front: inversely, your eluent is too polar; decrease the proportion of polar solvent in the same solvent system or chose a less polar solvent.

Silia*Plate* TLC Case Studies

Diels-Alder Cyclization of a Dihydropyridine

Structural studies of the *N*-(2,4-dinitrophenyl) derivative of a Diels-Alder-cyclized 1,2-dihydropyridine both unequivocally established the polycyclic framework and revealed interesting distortions of aromatic structure and unique dimeric clustering of the aromatic entities in the solid state.

SiliCycle Silia*Plate* TLC were used to monitor the conversion of a 1,2-dihydropyridine to the intramolecular Diels-Alder corresponding adduct, that was shown to be almost quantitative.



9a	^{t}Bu	Et
9b	Me	Me
9c	^{t}Bu	^{t}Bu

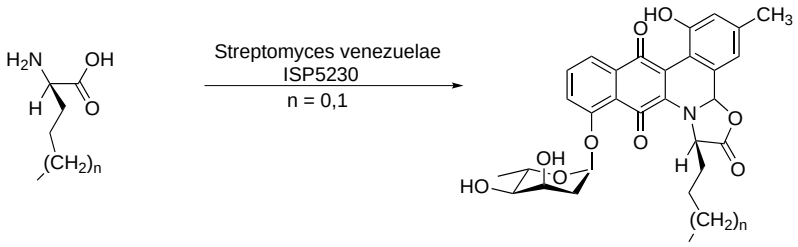
10a	^{t}Bu	Et
10b	Me	Me

Related Publication:
Helvetica Chimia Acta, **2014**, *97*, 1365-1382

Jadomycins Derived from the Assimilation and Incorporation of Norvaline and Norleucine

Streptomyces venezuelae ISP5230 is recognized for the production of chloramphenicol and the jadomycin family of natural products. The jadomycins are angucycline natural products containing a unique oxazolone ring incorporating an amino acid present in the minimal culture media. Substitution of different amino acids results in products of varying biological activity. Analysis of cultures of S. venezuelae ISP5230 incubated with L and D-norvaline and L and D-norleucine indicated that only the D-configured amino acids were incorporated into the natural products. Subsequently, jadomycin DNV and jadomycin DNL were isolated and characterized (*titers 4 and 9 mg L-1, respectively*). The compounds were evaluated in the National Cancer Institute cell line cancer growth inhibition and cytotoxicity screens, for antimicrobial activity against selected Gram-positive and Gram-negative bacteria, and as DNA-cleavage agents in vitro.

Glass-backed preparative TLC Silia*Plate* (*extra hard layer, 60 Å, 1,000 µm, UV indicator F_{254}*, PN: ***TLG-R10011B-341***) were used throughout the study for reactions monitoring.



Related Publication:
J. Nat. Prod., **2011**, *74*, 2420-2424

« We had tried working with TLC plates of another brand and realized that the SiliCycle brand was the most durable and long-lasting as well as clear when visualizing with UV light so we switched back. »

Jessica Kisunzu from UC Berkeley, Berkeley, CA, USA

Enantioselective Synthesis of an Ophiobolin Sesterterpene

Terpenes are a large and highly diverse class of natural products, produced by plants and mostly conifers.

They derive biosynthetically from isoprene, which cannot undergo by itself linking in a head-to-tail fashion, followed by a rearrangement to form rings. Cyclase enzyzmes are often used in the synthetic pathway, but are difficult to emulate under abiotic conditions.

Maimone et al. report in Science a impressive strategy to complex terpenes whereby simple prenyl-derived chains are cyclized using radical, rather than cationic, reaction pathways. This approche lead to the synthesis of 5-8-5 fused ring systems found in numerous complex natural product classes and also enabled a nine-step total synthesis of (–)-6-epi-ophiobolin N.

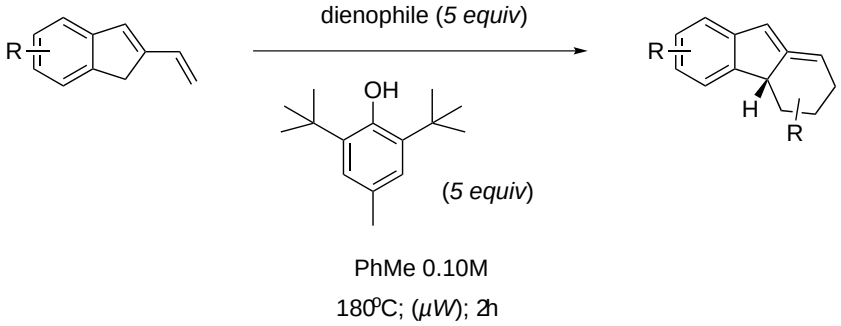
Reactions were followed via *Glass-Backed* TLC Silia*Plate* (*Special Layer for KMnO4, 250 µm, 20x20 cm UV indicator* F_{254}, PN: ***TLG-R10014BK-323***)

Related Publication:
Science, **2016**, 352, 1072-1082

Diels-Alder Reactivity of 2-vinylindenes in the Synthesis of Functionalized Tetrahydrofluorenes

Functionalized terahydrofluorenes are well-known starting block in the synthesis of various natural products, including kinamycins, taiwaniaquinoids, pharmaceutical compounds (for instance a selective estrogen receptor β-antagonist.

Sarpong et al. studied the synthesis of functionalized tetrahydrofluorenes using a normal electron-demand Diels-Alder cycloaddition reaction between 2-vinylindenes and various dienophiles. Electron rich 2-vinylindenes bearing methoxy groups at the 4- and 7- positions were accessed through their corresponding 2-indenylpivalates obtained using a Pt-catalyzed cycloisomerization reaction.



All reactions were assessed using Glass-backed TLC Silia*Plate* (*Glass-Backed, Hard Layer, 250 µm, 20x20 cm, UV indicator* F_{254}, PN: ***TLG-R10014B-323***).

Related Publication:
Tetrahedron, **2016**, *72*, 3635-3640

One-Step Synthesis of Methanesulfonyloxymethyl Ketones via Gold-Catalyzed Oxidation of Terminal Alkenes: A Combination of Ligand and Counter-Anion Enables High Efficiency and a One-Pot Synthesis of 2,4-Disubstituted Thiazoles

By using Mor-DalPhos as the P,N-bidentate ligand and mesylate as the counter-ion, the resulting gold(I) complex catalyzes efficient oxidative transformations of various terminal alkynes into synthetically versatile methanesulfonyloxymethyl ketones. The mild reaction conditions and high efficiency permit the one-pot synthesis of a range of valuable 2,4-disubstituted thiazoles by subjecting the resulting reaction mixture to a further condensation with thioamides under mild conditions.

All reactions were monitored by thin layer chromatography using SiliCycle analytical Silia*Plate*.

Related Publication:
Adv. Synth. Catal., **2014**, *356*, 1229-1234

Utilizing Mor-DalPhos/Palladium-Catalyzed Monoarylation in the Multicomponent One-Pot Synthesis of Indoles

The application of a Mor-DalPhos/palladium catalyst system in the one-pot, multicomponent assembly of substituted indoles from ortho-chlorohaloarenes, alkyl ketones (*including acetone*) and primary amines is reported. The described protocols offer improved substrate scope in all three reaction components, under more mild conditions and without the need for an additional drying agent. Also reported are the first examples of such multicomponent reactions where all reactants are combined at the start of the reaction, without the need for inert atmosphere reaction conditions.

Preparatory thin layer chromatography to monitor monoarylations was carried out using SiliCycle glass-backed TLC Silia*Plate* (*extra hard layer, 60 Å, 1,000 µm, UV indicator F_{254}, PN:* ***TLG-R10011B-341***).

Related Publication:
Adv. Synth. Catal., **2015**, *357*, 100-106