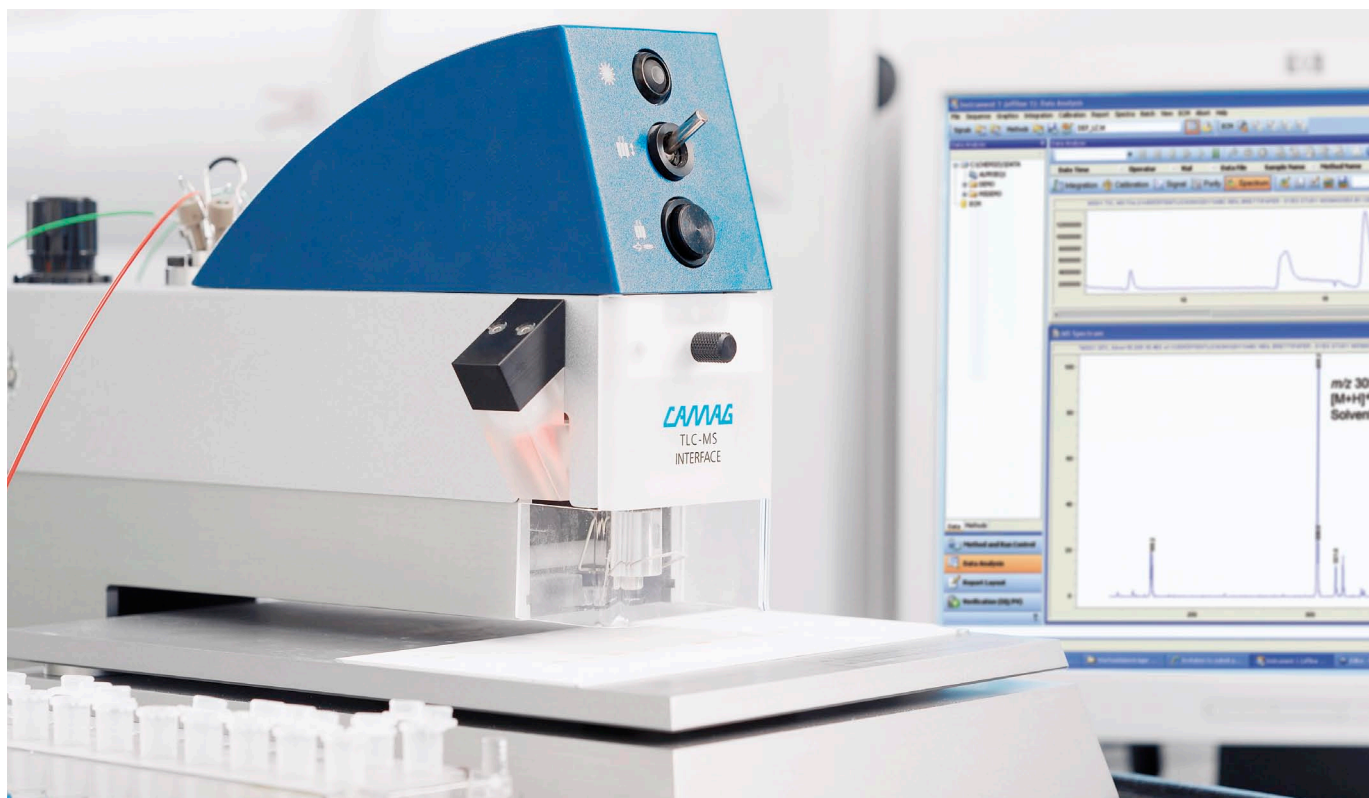


# CBS

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**Die Kopplung der Planar-Chromatographie mit der Massenspektrometrie wird von Analytikern sehr positiv aufgenommen.**

 **INTERNATIONAL SYMPOSIUM FOR  
HIGH-PERFORMANCE THIN-LAYER  
CHROMATOGRAPHY**  
Basel, Switzerland, 06–08 July 2011

**CAMAG**

**106**

Nr. 106, März 2011

CAMAG Literaturdienst  
Planar-Chromatographie  
Herausgegeben von Gerda Morlock  
cbs@camag.com  
Eigenverlag CAMAG Schweiz

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**Direkte Bioautographie zur Bestimmung von Enrofloxacin und Ciprofloxacin in Milch**



Von links: M.Sc. Wioleta Bał, Dr. hab. Irena Choma, M.Sc. Edyta Grzelak, Dr. Karol Pilarz und Dr. hab. Barbara Majer-Dziedzic.

Dr. hab. Irena Choma\*, Wissenschaftlerin und Dozentin am Institut für Chromatographische Methoden der Maria-Curie-Skłodowska-Universität in Lublin, Polen, wendet in der Forschung unterschiedliche chromatographische Methoden an. Für die Entwicklung neuer und optimierter analytischer Verfahren zur Bestimmung von Antibiotika-Rückständen in Lebensmitteln wie Milch oder Fleisch wird vor allem die Planar-Chromatographie eingesetzt und deren Kopplung mit mikrobiologischen Testverfahren (TLC/HPTLC-direkte Bioautographie, TLC/HPTLC-DB). Die vorgestellte TLC-DB-Methode ermöglicht die Bestimmung der antibakteriellen Aktivität von Substanzonen und wurde in Zusammenarbeit mit Dr. hab. Barbara Majer-Dziedzic von der Naturwissenschaftlichen Universität in Lublin entwickelt.

**Einleitung**

Fluorchinolone werden häufig in der Human- und Veterinärmedizin eingesetzt. Ciprofloxacin ist eines der beliebtesten Antibiotika der Humanmedizin und der Hauptmetabolit von Enrofloxacin, das in der Tiermedizin hauptsächlich zur Behandlung von Euterentzündungen bei Kühen angewendet wird. Beide Wirkstoffe sind als Rückstände in der Milch behandelter Kühe zu finden. Geeignete analytische Nachweismethoden müssen so empfindlich und selektiv sein, dass diese Rückstände in der Milch auch bei einem summierten Rückstandshöchstgehalt von 100 µg/kg noch unterschieden werden können.

**Die in der Antibiotika-Analytik üblicherweise eingesetzten HPLC-Methoden können häufig durch HPTLC/TLC in Kombination mit mikrobiologischer Detektion ersetzt werden. Diese ermöglicht neben der Trennung der Antibiotika auch gleichzeitig einen Nachweis auf Basis ihrer antibakteriellen Eigenschaften. Dazu wird die entwickelte Platte direkt in eine Bakteriensuspension getaucht. Nach**

der Inkubation und Sichtbarmachung kann in den antibiotisch wirksamen Chromatogrammm-zonen eine Hemmung des Bakterienwachstums beobachtet werden. Die Methode ist einfach und zugleich höchst empfindlich, genau und präzise. Vorteile im Vergleich zur HPLC sind nicht nur der hohe Probendurchsatz, sondern auch der zusätzliche Hinweis auf die antibakteriellen Eigenschaften der Proben. Von grossem Nutzen ist auch der Nachweis weiterer antibakterieller Substanzen, die unbekannt (z.B. Abbauprodukte) oder im Rahmen der üblichen Untersuchungen nicht im Fokus sind.

## Schicht

DC-Platten Kieselgel 60 F<sub>254</sub> (Merck), 10 × 20 cm

## Standardlösungen

Als Stammlösung wurde eine Mischung von je 1 mg/mL Ciprofloxacin und Enrofloxacin in Methanol hergestellt. Die Standardlösungen wurden durch entsprechendes Verdünnen mit Methanol hergestellt.

## Probenvorbereitung

Für die Milchproben wurde Milch (Fettgehalt 2 %) mit geeigneten Volumina der Stammlösung aufgestockt. Für die Extraktion der Antibiotika-Rückstände aus der Milch wurde eine Modifikation der MSPD-Methode (matrix solid-phase dispersion) eingesetzt. Zum Entfetten wurde die Säule mit Hexan gespült. Enrofloxacin und Ciprofloxacin wurden mit Dichlormethan eluiert, unter einem leichten N<sub>2</sub>-Strom zur Trockne reduziert, und der Rückstand in Methanol aufgenommen.

## Probenauftragung

Die Standardlösung und die Milchextrakte wurden bandförmig mit dem Linomat 5 aufgetragen, Bandlänge 3 oder 5 mm, Auftragevolumina 10–50 µL.

## Chromatographie

In der DS Horizontal-Entwicklungskammer (Chromdes) mit Dichlormethan – Methanol – Isopropanol – 25 % Ammoniaklösung 3:3:5:2.

## Direkte DC-Bioautographie

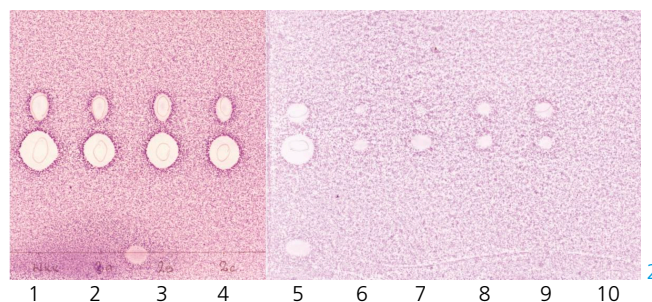
Für den bioautographischen Nachweis mit *Bacillus subtilis* wurde das Chrom Biodip-Testkit für Antibio-

tika (Merck) verwendet [1]. Das Nachweisverfahren mit *Escherichia coli*-Bakterien wurde von uns entwickelt [2]. Nach dem Trocknen wurde die Platte mit der Chromatogramm-Tauchvorrichtung für 5 s in eine mit *E. coli* geimpfte Nährlösung getaucht und danach bei 37 °C während 5 h in einer Feuchtkammer inkubiert. Zur Detektion wurde die Platte mit einer wässrigen Tetrazoliumsalz (MTT)-Lösung (0.2 %) besprüht. Ciprofloxacin und Enrofloxacin konnten als beige-weiße Hemmzonen vor einem rosarot gefärbten Plattenhintergrund nachgewiesen werden. Die Hemmzonen wurden mit einem Planimeter gemessen.

Anmerkung: Die Hemmzonen können auch mit dem TLC Scanner 4 gemessen werden. Da der Hintergrund nicht weiss, sondern gefärbt ist, würde man negative Peaks erhalten. Dies erfordert eine besondere Software-Einstellung für die Absorptionsmessung: Maximale Wellenlänge der Hintergrundfarbe als Messwellenlänge und Fluoreszenzmessung als Messmodus. Dieser unübliche Messmodus ermöglicht die normale Peakdarstellung.

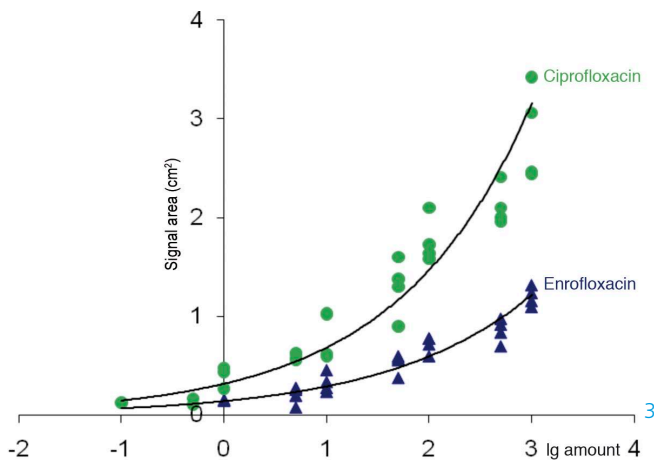
## Ergebnisse und Diskussion

Für den bioautographischen Nachweis mit *Bacillus subtilis* wurden Milchproben mit 50 und 1000 µg/kg aufgestockt, wobei für den Nachweis auf dem tieferen Konzentrationsbereich ein Auftragevolumen von 50 µL Milchextrakt nötig war [3].



TLC-DB (*B. subtilis*-Test) von Milchproben, aufgestockt mit 50 und 1000 µg/kg Enrofloxacin ( $hR_f = 58$ ) und Ciprofloxacin ( $hR_f = 42$ ); Bahn 1: Standardlösung (10 mg/L); Bahnen 2–4: 10 µL Milchextrakt aufgestockt mit 1000 µg/kg; 5–6: Standardlösung (1000 und 500 µg/kg); 7–9: 50 µL Milchextrakt aufgestockt mit 50 µg/kg; 10: Blindprobe

Die Auswertung der direkten Bioautographie erfolgt durch Regressionsanalyse über die Fläche der Hemmzonen. Im Konzentrationsbereich über ein bis zwei Grössenordnungen ist die Korrelation zwischen der Hemmzonenfläche und dem Logarithmus der Antibiotika-Konzentration linear. Für einen weiten Konzentrationsbereich ist die exponentielle Funktion besser geeignet [4].



Korrelation der Fläche der Hemmzonen ( $\text{cm}^2$ ) zum Logarithmus der Antibiotikamenge ( $\text{ng}/\text{Zone}$ ); Daten von 4 Platten

Momentan werden zwei neue Bioautographie-Tests mit *Bacillus subtilis* und *Escherichia coli* [2] entwickelt, die den Nachweis in realen Milchproben von mit Antibiotika behandelten Kühen ermöglichen sollen. Im Vergleich zum Chrom Biodip-Test ist im *E. coli*-Test die Nachweisgrenze für Ciprofloxacin niedriger ( $25 \mu\text{g}/\text{kg}$ ), jedoch für Enrofloxacin etwas höher ( $75 \mu\text{g}/\text{kg}$ ).

Weitere Informationen sind bei den Autoren auf Anfrage erhältlich.

- [1] R. Eymann *et al.* in: Planar Chromatography, Proceedings, Lillafüred, Hungary, 2000, 67.
- [2] E.M. Grzelak *et al.* J. AOAC Int., in press.
- [3] I.M. Choma, J. Planar Chromatogr. 19 (2006) 104.
- [4] I.M. Choma *et al.* J. Liq. Chromatogr. Relat. Technol. 27 (2004) 2071.

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### CAMAG BioLuminizer

Die Trennleistung der Planar-Chromatographie verbunden mit Bioassay-Detektion ermöglicht die Identifizierung von biologisch aktiven Substanzen. Alle Verbindungen, die einen entsprechenden Effekt zeigen, werden in komplexen Gemischen angezeigt, so auch Abbauprodukte, unbekannte Metaboliten oder unerlaubt ein- bzw. zugesetzte Wirkstoffe.

Im Gegensatz zu den in diesem Beitrag eingesetzten Bioassays, die auf einer Farbreaktion nach Inkubation beruhen und mit dem TLC Scanner 4 detektiert werden können, gibt es das Leuchtbakterien-Bioassay als Soforttest (siehe CBS 105). Bioaktive Substanzen sind nach dem Eintauchen in die Bakterien-suspension direkt sichtbar als dunkle oder aufgehellte Zonen auf einem lumineszierenden Hintergrund (Bakterien unbeeinflusst).

Der BioLuminizer ist ein kompaktes, benutzerfreundliches Detektionssystem, das aussagekräftige Bilder von hoher Qualität und Auflösung bei kurzen Belichtungszeiten liefert. Unter der Bezeichnung BioLuminex Standardkit (Nr. 022.9765) liefert CAMAG einen kompletten Satz der benötigten Verbrauchsmaterialien.



## Einsatz der Planar-Chromatographie zur Analyse von Peptiden aus tryptischem Verdau von Proteinen



Michael Schulz und Susanne Minarik



Dr. Gary van Berkel

Seit 1966 bietet Merck eine Vielzahl verschiedener Fertigplatten mit unterschiedlichen Schichtmaterialien und Oberflächenmodifizierungen an. Im Jahr 2006 wurden die ProteoChrom<sup>®</sup> HPTLC-Schichten eingeführt, die sich besonders gut für die Trennung von Peptiden aus dem tryptischen Verdau von Proteinen eignen. Spezielle Merkmale dieser Schichten sind die reduzierte Schichtdicke (100 µm) und ein hinsichtlich Wasserstabilität optimiertes Bindersystem. Damit werden hohe Empfindlichkeiten und grosse Auftragevolumina wässriger Proben ermöglicht. Die eindimensionale Trennung kann auf der ProteoChrom<sup>®</sup> Kieselgelschicht durchgeführt werden, und für die zweidimensionale Trennung ist die ProteoChrom<sup>®</sup> Celluloseschicht besonders gut geeignet. Die anschliessende Identifizierung mit der Massenspektrometrie entstand in einer Zusammenarbeit mit Dr. Gary van Berkel, Oak Ridge National Laboratory, USA.

### Einleitung

Die Planar-Chromatographie mit HPTLC-Platten ist eine häufig eingesetzte und vielseitige Analysenmethode. Mit dem Einsatz von Präzisionsgeräten für die Auftragung und Auswertung hat sich die Methode auch in der quantitativen Analytik etabliert. In den letzten Jahren ist ein Trend zu Anwendungen zu beobachten, in denen die Planar-Chromatographie mit der Massenspektrometrie (MS) gekoppelt wird; insbesondere die Weiterentwicklung verschiedener Ionisierungstechniken und kommerziell erhältlicher Geräte eröffnen auch in der Bioanalytik neue Möglichkeiten.

In dieser Arbeit wurden Peptide aus dem tryptischen Verdau von Myoglobin, Cytochrom C, β-Casein und bovine serum albumin (BSA) auf HPTLC-Platten aufgetrennt und anschliessend durch Kopplung mit DESI-MS ausgelesen. Die erreichte Sequenzabdeckung wurde mit der HPLC verglichen [1, 2].

### Probenvorbereitung

Die Proteine werden mit dem Enzym Trypsin in einem Verhältnis von 100:1 bei einer Proteinkonzentration von 2 mg/mL in 25 mM Ammoniumcarbonat-Puffer verdaut.

### Schicht

ProteoChrom<sup>®</sup> HPTLC-Platte Kieselgel 60 F<sub>254</sub>S, 20 × 10 cm, Schichtdicke 100 µm (Merck Artikel 1.05650.0001)

ProteoChrom<sup>®</sup> HPTLC-Aluminiumfolie Cellulose, 10 × 10 cm, Schichtdicke 100 µm (Merck Artikel 1.05651.0001)

### Probenauftragung

Bandförmig mit dem DC-Probenautomat 4, Bandlänge 5 mm, Bahnabstand 10 mm, Abstand vom unteren Rand 10 mm, Auftragevolumen 7 µL

### Chromatographie

In der Flachbodenkammer 20 × 10 cm bzw. 10 × 10 cm; eindimensional auf der Kieselgelschicht mit 2-Butanol – Pyridin – Ammoniak – Wasser 39:34:10:26; zweidimensional auf der Celluloseschicht mit 2-Butanol – Pyridin – Essigsäure – Wasser 15:10:3:12 und dem vorherigen Fließmittel für die Kieselgelschicht (Laufstrecke 50 mm)

### Postchromatographische Derivatisierung

Mit dem DC-Sprüngerät wird die Platte mit folgenden Derivatisierungsreagenzien besprüht, evtl. erhitzt und ausgewertet:

- Ninhydrin-Lösung (0,5 % in 2-Propanol) → Platte bei 110 °C für ca. 2 min erhitzen → Weisslicht
- Fluorescamin-Lösung (0,02 % in Aceton) → 10 min bei RT trocknen → Platte mit Triethylamin-Lösung (10 % in Aceton) besprühen → 10 min bei RT trocknen → UV 366 nm

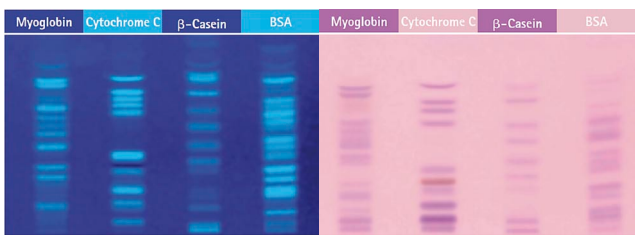
- Merck ProteoChrom® COLOR Peptide Staining Kit: Platte mit ProteoChrom® COLOR Reagenz besprühen → 5 min bei RT trocknen → Platte mit Ninhydrin-Lösung besprühen → Platte bei 110 °C für 1–2 min erhitzen → Weisslicht

## Massenspektrometrie

Die nicht-derivatisierte Platte wurde auf einem x-y-z-Tisch justiert. Für die Ortsbestimmung der Peptide wurde eine Bahn im Fullscan-Modus mit einem selbst modifizierten Desorptions-Elektrospray-Strahl (Einfallswinkel 55°, Wasser als Spray-Lösungsmittel) in einem Abstand von 2–3 mm in einer Geschwindigkeit von 100 µm/s über 55 mm abgefahren (9 min/Bahn). Die desorbierten, ionisierten Moleküle wurden über eine verlängerte Transferkapillare im Abstand von 2 mm über dem Desorptionsbereich ins DESI-MS (LCQ Deca Ionenfallen-Massenspektrometer, ThermoFinnigan) überführt. Für die MS- oder MS/MS-Spektren der lokalisierten Peptide wurden erneut DESI-Massenspektren aufgenommen.

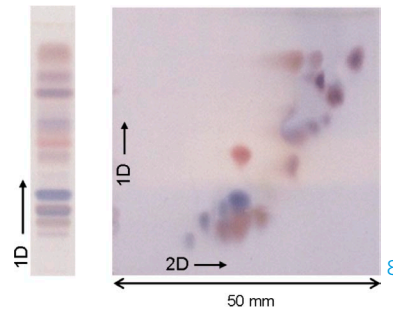
## Ergebnisse und Diskussion

Eindimensional können in der HPTLC bis zu 20 Banden getrennt werden. Das entspricht in etwa der Anzahl von Peptiden, die beim tryptischen Verdau kleiner Proteine entstehen. Zum Beispiel besteht Cytochrom C aus 104 AS, und daraus entstehen 17 Peptide nach dem tryptischen Verdau. Sehr scharfe Banden werden bei eindimensionaler Trennung auf der HPTLC-Kieselgelschicht erhalten.



Eindimensionale Trennung von Peptiden aus dem tryptischen Proteinverdau auf der ProteoChrom® HPTLC-Kieselgelschicht nach Derivatisierung mit Fluorescamin-Reagenz (links) und Ninhydrin-Reagenz (rechts)

Durch eine zweidimensionale Trennung kann die Anzahl aufgetrennter Peptide erhöht werden. Dafür eignet sich besonders gut die HPTLC-Celluloseschicht.



Zweidimensionale Peptid-Trennung nach dem tryptischen Verdau von Cytochrom C auf der ProteoChrom® Celluloseschicht und Derivatisierung mit ProteoChrom® COLOR Staining Kit

In der folgenden Tabelle sind die erreichten Sequenzabdeckungen für Cytochrom C und Myoglobin nach einer HPTLC/DESI-MS-Auswertung gegenübergestellt. Die Sequenzabdeckungen variieren je nach eingesetzter Trennschicht und Trennqualität und sind für die zweidimensionale Trennung auf Celluloseschichten zum Beispiel fast vergleichbar mit der erreichten Sequenzabdeckung durch HPLC/ESI-MS.

		HPTLC/ESI-MS	1D-HPTLC/DESI-MS	2D-HPTLC/DESI-MS
ProteoChrom® HPTLC Silica gel 60 F <sub>254s</sub>	Cytochrome C		58%	
ProteoChrom® HPTLC Cellulose	Cytochrome C		72%	81%
Acclaim PepMap 100 C18	Cytochrome C	92%		
ProteoChrom® HPTLC Silica gel 60 F <sub>254s</sub>	Myoglobin		62%	
ProteoChrom® HPTLC Cellulose	Myoglobin		68%	74%

Damit ist die Methode gut einsetzbar für einfache bis durchschnittlich komplexe Peptidmischungen. Ein grosser Vorteil der HPTLC/MS-Kopplung ist die von der Trennung zeitlich unabhängige Detektion. Die getrennten Zonen sind auf der HPTLC-Platte gespeichert und können nach Bedarf mit der Massenspektrometrie ausgelesen werden. Die generellen Vorteile der HPTLC, viele Proben parallel zu analysieren und die hohe Flexibilität hinsichtlich der Detektion, können auch in der Analytik von Peptiden gut genutzt werden. Eine einfache und schnelle Derivatisierung wird mit dem Ninhydrin-Reagenz erreicht, eine hohe Nachweisempfindlichkeit mit dem Fluorescamin-Reagenz und eine charakteristische Anfärbung der einzelnen Peptide mit dem ProteoChrom® COLOR-Peptide Staining Kit.

Weitere Informationen sind von den Autoren auf Anfrage erhältlich.

[1] S. P. Pasilis *et al.* Anal. Bioanal. Chem. 391 (2008) 317

[2] S. P. Pasilis *et al.* J. Mass Spectrom. 43 (2008) 1627

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## HPLC-MS oder einfach nur HPTLC zur Analyse von Sucralose in Wasser?



Leonard Schuele (links) und Sebastian Grashorn

Am Institut für Lebensmittelchemie der Universität Hohenheim, Stuttgart, werden für Proben mit komplexer Matrix alle relevanten analytischen Methoden berücksichtigt, um die effizienteste dann einzusetzen. Für die Bestimmung von Sucralose in Lebensmitteln wurde die HPTLC gewählt [1, 2, CBS 94]. Diese Methode wurde für Abwasser, Oberflächenwasser und Trinkwasser in den Bachelor-Arbeiten von Leonard Schüle und Sebastian Grashorn unter Betreuung von apl. Prof. Dr. Gerda Morlock optimiert.

### Einleitung

Sucralose, ein persistenter chlorierter Süßstoff, der seit 2005 in der EU zugelassen ist, wurde bereits im Abwasser nachgewiesen. Daher untersuchen einige Länder den Eintrag von Sucralose in die aquatische Umwelt. Als analytische Methoden in diesem Ultraspurenbereich wurden bisher vor allem HPLC-MS bzw. HPLC-MS/MS, HPLC-TOF MS und GC-MS nach Festphasen-Extraktion eingesetzt.

**Eine zu den bisherigen Methoden hinsichtlich Trennprinzip und Detektion unterschiedliche, quantitative HPTLC-Methode erwies sich als hervorragend geeignet zum schnellen Nachweis von Sucralose. 17 Proben wurden innerhalb von 15 min parallel getrennt. Ein weiterer Vorteil ist die post-chromatographische Derivatisierung, die einen selektiven Nachweis von Sucralose in Abwasser und Oberflächenwasser ermöglicht. Der ermittelte Sucralosegehalt in vier durch einen Ringversuch spezifizierten Wasserproben stimmte gut mit den Labor-Mit-**

**telwerten überein, die mit HPLC-TOF MS oder HPLC-MS/MS und überwiegend nach Korrektur mit deuterierten Standards erhalten wurden. Die Genauigkeit, die Kosteneffizienz und der hohe Probendurchsatz belegen HPTLC als Methode der Wahl.**

### Schicht

HPTLC-Platten Kieselgel 60 F<sub>254</sub> (Merck), 20 × 10 cm, vorgewaschen durch Chromatographie mit Methanol und anschliessend für 15 min bei 100 °C getrocknet

### Standardlösung

5 mg Sucralose wurden in 50 mL Methanol gelöst und 1:10 mit Methanol verdünnt (10 ng/μL).

### Probenvorbereitung

Die Wasserprobe (0,5 L, eingestellt auf pH 8) wurde durch Festphasen-Extraktion an einem Styrol-Divinylbenzol-Copolymer-Adsorbens (500 mg/3 mL, Bond Elut PPL, Varian) bei einer Flussrate von 10 mL/min extrahiert. Das Sorbens wurde 30 min mit Luft getrocknet. Die Elution durch Schwerkraft erfolgte mit 1 mL Methanol nach jeweils 5 min Einwirkzeit und wurde 2 mal wiederholt. Das vereinigte Eluat wurde durch eine Aminopropyl-Phase (200 mg/3 mL Bond Elut NH<sub>2</sub>, Varian) gereinigt, die zusätzlich mit 2 mL Methanol eluiert wurde (zusammen mit dem gereinigten Eluat gesammelt), und nach dem schonenden Einengen auf 2 mL in ein Probengefäß überführt.

### Probenauftragung

Flächenförmig (8 × 5 mm) mit ATS 4, 17 Proben, Bahnabstand 10 mm, linker Randabstand 20 mm, unterer Randabstand 8 mm, Auftragevolumina 100–300 μL für Proben und 1, 10, 20 und 30 μL für Standardlösungen, Temperatur der Sprühdüse 60 °C (Erhöhung der Dosiergeschwindigkeit von 150 nL/s auf 1000 nL/s reduziert die Auftragezeit für grosse Volumina um 70 %), 100 bzw. 300 μL-Volumina wurden so in 1,7 bzw. 5 min aufgetragen.

### Chromatographie

In der ADC 2 mit Isopropanol, Methanol und Wasser

Fortsetzung auf Seite 9



## Neue Gesichter im Bereich Vertrieb und Marketing

Diesen Bereich haben wir in den letzten Jahren. ausgebaut. Einige Mitarbeiter sind dazu gekommen, und Verantwortlichkeiten wurden z.T. neu verteilt.



10

**Jürg Leuenberger** (49) ist schon seit 2003 dabei. Nach seiner Ausbildung als Chemielaborant bei Ciba-Geigy arbeitete er in Synthese- und Qualitätskontroll-labors und war dann 6 Jahre für das Rote Kreuz in Afrika, Asien und Südamerika tätig. Bei CAMAG betreute er zunächst unsere Kunden in der Schweiz. Aufgrund seiner Auslandserfahrung und seiner Sprachkenntnisse ernannten wir ihn 2010 zum Gebiets-Manager für Lateinamerika. Als solcher bereist er diese Länder häufig. Vom Mutterhaus aus bearbeitet er Kundenanfragen und wirkt aktiv bei Vertreterkursen mit.



11

**Raphael Vizzini** (36) kam im August 2008 zu CAMAG. Er erhielt seine Ausbildung als eidg. dipl. Laborant in der Pharmaforschung der Roche und war dann in der Verfahrensentwicklung bei Roche, DSM und Synphabase tätig. Bei CAMAG ist er im Vertrieb als Gebietsmanager für Europa, den Mittleren Osten und Afrika zuständig. Dies beinhaltet eine grosse Reisetätigkeit zur Betreuung unserer Vertretungen, Organisation von Seminaren und die Teilnahme an Messen. Bei Vertreterkursen im Hause übernimmt er einen Teil des Trainings.



12

**Tobias Hohler** (30) gehört seit Juli 2010 zu unserem Team. Zuvor war er bei einer Exportfirma beschäftigt. Er ist Diplom-Kaufmann und besitzt das Handelsdiplom. Bei CAMAG arbeitet er in der Administration von Vertrieb & Marketing. Dies beinhaltet u.a. das Offertenwesen im Exportgeschäft, die Organisation von Kunden- und Vertreter-Veranstaltungen in Zusammenarbeit mit unserem Labor. Auch die Auftragsabwicklung für Südamerika gehört zu seinen Aufgaben.



13

**Lukas Frommenwiler** (24) trat im August 2010 bei uns ein. Seine Ausbildung als Chemielaborant erhielt er im Labor Spiez. Anschliessend absolvierte er verschiedene Weiterbildungen, u.a. auf dem Gebiet des Marketing. Bei EMS Chemie war er in einem Produktionslabor tätig, danach bearbeitete er bei LONZA Kundenprojekte und war auch in der Lehrlingsausbildung aktiv. Bei CAMAG betreut er unsere Homepage, hilft bei der Organisation von Kundenveranstaltungen und wird sich in die Erstellung von Verkaufsliteratur und Werbematerial einarbeiten.



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**CBS**

Liebe Freunde

Die Kopplung der Planar-Chromatographie mit der Massenspektrometrie wird von Analytikern sehr positiv aufgenommen. Sie unterstützt ihre Entscheidungen, sichert sie ab oder lässt neue Erkenntnisse gewinnen. Dabei ist die HPTLC-MS-Kopplung nur eine von vielen Möglichkeiten, Informationen aus einem chromatographischen Lauf zu gewinnen. Ein neuer GDCh-Kurs, der auf der letzten gelben Seite detailliert beschrieben ist, zeigt die Vielfältigkeit der aktuellen Kopplungsmöglichkeiten in der HPTLC auf. 2012 wird dieser Kurs in Englisch angeboten.

Das vorläufige Programm für das HPTLC-Symposium in Basel vom 6.–8. Juli 2011 ist fertig. Dieses sowie aktuelle Informationen entnehmen Sie der homepage [www.hptlc.com](http://www.hptlc.com). 179 Abstracts von Wissenschaftlern aus 23 Ländern wurden als Vorträge oder Poster mit aktuellen Themen aus den unterschiedlichsten Anwendungsbereichen angenommen. Die Registrierung ist noch offen (Anmeldeschluss: 30. Mai). Bisher sind schon über 120 Teilnehmer angemeldet. Erfahren Sie in Basel, ob HPTLC für ihre Fragestellungen geeignet ist. Alle Kopplungstechniken und Anwendungsbereiche sind vertreten. Sprechen Sie mit den Experten vor Ort.

Sie sind herzlich willkommen – auch das Rahmenprogramm ist attraktiv. Sehen wir uns dort? Ich würde mich freuen!

Herzlichst

*Gerda Morlock*

Gerda Morlock  
[cbs@camag.com](mailto:cbs@camag.com)

Dear friends

Coupling of planar chromatography with mass spectrometry is obviously appreciated by many analysts. It is suited to support decisions or gain knowledge otherwise. Thereby TLC/HPTLC-MS coupling is just one of many possibilities to gain information from a chromatographic run.

A new course organized by the German Chemical society (see last yellow page) shows the diversity of current coupling techniques. This course will be held in English in 2012.

The tentative program for the International HPTLC Symposium in Basel, 6th–8th July 2011 is ready. This and other current information is available on the homepage [www.hptlc.com](http://www.hptlc.com). 179 abstracts of scientists from 23 countries have been accepted as oral or poster presentations on current topics from a variety of application fields. Registration is still open (deadline 30th May). So far, already more than 120 participants have registered. Join us and experience how far TLC/HPTLC is suited for your tasks. All coupling techniques and application fields are represented. Come and exchange with the experts. The social program is attractive too.

Will I have the privilege to see you in Basel? I would appreciate it!

Sincerely,

*Gerda Morlock*

Gerda Morlock  
[cbs@camag.com](mailto:cbs@camag.com)



**CAMAG**

**MARCH  
2011** **106**

# THE CBS CLASSIFICATION SYSTEM

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- 1. Reviews and books**
  - a) Books on TLC
  - b) Books containing one or several chapters on TLC
  - c) Books containing frequent TLC information spread over several chapters of other information
- 2. Fundamentals, theory and general**
  - a) General b) Thermodynamics and theoretical relationship
  - c) Relationship between structure and chrom. behaviour
  - d) Measurement of physico-chemical and related values
  - e) Optimization of solvent systems
  - f) Validation of methods
- 3. General techniques** (unless they are restricted to the application within one or two classification sections)
  - a) New apparatus/techniques for sample preparation
  - b) Separation material
  - c) New apparatus for sample application/dosage
  - d) New apparatus/techniques for chromatogram development
  - e) New apparatus/techniques for pre- or post-chromatographic derivatization
  - f) New apparatus/techniques for quantitative evaluation
  - g) New apparatus/techniques for other TLC steps (distinguished from section 4)
- 4. Special techniques**
  - a) Automation of sample preparation/application
  - b) Automation of complex chromatogram developing techniques
  - c) Automation, computer application in quantitative chromatogram evaluation
  - d) Combination of TLC with other chromatographic techniques
  - e) Combination of TLC with other (non-chromatographic) techniques...MS, IR...etc.
- 5. Hydrocarbons and halogen derivatives**
  - a) Aliphatic hydrocarbons
  - b) Cyclic hydrocarbons
  - c) Halogen derivatives
  - d) Complex hydrocarbon mixtures
- 6. Alcohols**
- 7. Phenols**
- 8. Substances containing heterocyclic oxygen**
  - a) Flavonoids
  - b) Other compounds with heterocyclic oxygen
- 9. Oxo compounds, ethers and epoxides**
- 10. Carbohydrates**
  - a) Mono- and oligosaccharides, structural studies
  - b) Polysaccharides, mucopolysaccharides, lipopolysaccharides
- 11. Organic acids and lipids**
  - a) Organic acids and simple esters
  - b) Prostaglandins
  - c) Lipids and their constituents
  - d) Lipoproteins and their constituents
  - e) Glycosphingolipids (gangliosides, sulfatides, neutral glycosphingolipids)
- 12. Organic peroxides**
- 13. Steroids**
  - a) Pregnane and androstane derivatives
  - b) Estrogens
  - c) Sterols
  - d) Bile acids and alcohols
  - e) Ecdysones and other insect steroid hormones
- 14. Steroid glycosides, saponins and other terpenoid glycosides**
- 15. Terpenes and other volatile plant ingredients**
  - a) Terpenes
  - b) Essential oils
- 16. Nitro and nitroso compounds**
- 17. Amines, amides and related nitrogen compounds**
  - a) Amines and polyamines
  - b) Catecholamines and their metabolites
  - c) Amino derivatives and amides (excluding peptides)
- 18. Amino acids and peptides, chemical structure of proteins**
  - a) Amino acids and their derivatives
  - b) Peptides and peptidic proteinous hormones
- 19. Proteins**
- 20. Enzymes**
- 21. Purines, pyrimidines, nucleic acids and their constituents**
  - a) Purines, pyrimidines, nucleosides, nucleotides
  - b) Nucleic acids, RNA, DNA
- 22. Alkaloids**
- 23. Other substances containing heterocyclic nitrogen**
  - a) Porphyrins and other pyrroles
  - b) Bile pigments
  - c) Indole derivatives
  - d) Pyridine derivatives
  - e) other N-heterocyclic compounds
- 24. Organic sulfur compounds**
- 25. Organic phosphorus compounds** (other than phospholipids)
- 26. Organometallic and related compounds**
  - a) Organometallic compounds
  - b) Boranes, silanes and related non-metallic compounds
  - c) Coordination compounds
- 27. Vitamins and various growth regulators** (non-peptidic)
- 28. Antibiotics, Mycotoxins**
  - a) Antibiotics
  - b) Aflatoxins and other mycotoxins
- 29. Pesticides and other agrochemicals**
  - a) Chlorinated insecticides
  - b) Phosphorus insecticides
  - c) Carbamates
  - d) Herbicides
  - e) Fungicides
  - f) Other types of pesticides and various agrochemicals
- 30. Synthetic and natural dyes**
  - a) Synthetic dyes
  - b) Chloroplasts and other natural pigments
- 31. Plastics and their intermediates**
- 32. Pharmaceutical and biomedical applications**
  - a) Synthetic drugs
  - b) Pharmacokinetic studies
  - c) Drug monitoring
  - d) Toxicological applications
  - e) Plant extracts, herbal and traditional medicines
  - f) Clinico-chemical applications and profiling body fluids
- 33. Inorganic substances**
  - a) Cations
  - b) Anions
- 34. Radioactive and other isotopic compounds**
- 35. Other technical products and complex mixtures**
  - a) Surfactants
  - b) Antioxidants and preservatives
  - c) Various specific technical products
  - d) Complex mixtures and non-identified compounds
- 36. Thin-layer electrophoresis**
- 37. Environmental analysis**
  - a) General papers
  - b) Air pollution
  - c) Water pollution
  - d) Soil pollution
- 38. Chiral separations**

## 1. Reviews and books

- 106 001 Marta KUCHARSKA\*, J. GRABKA (\*Institute for Engineering of Polymer Materials and Dyes, Department of Dyes and Organic Products in Zgierz, 2/4 Chemików Street, 95-100 Zgierz, Poland): A review of chromatographic methods for determination of synthetic food dyes. *Talanta*, 80 (3), 1045-1051 (2010). Review on chromatographic methods for synthetic food dyes, including the following techniques: TLC, HPTLC, traditional column chromatography, HPLC, ion-pair chromatography, RP HPLC, and high performance ion chromatography, demonstrated by using examples in different conditions for each technique.

quality control, food analysis, HPTLC, review

1, 30

- 106 002 J. SHERMA (Department of Chemistry, Lafayette College, Easton, Pennsylvania, USA, [shermaj@lafayette.edu](mailto:shermaj@lafayette.edu)): Pesticides: TLC analysis. *Encyclopedia of Chromatography Third Edition* 1, 1749-1756 (2009). This review describes the current materials and techniques most widely used for the analysis of pesticides by TLC. In detail, information regarding sample preparation, stationary and mobile phases, detection and quantification is included. Specific examples of pesticides analysis in water and soil are also described.

environmental, HPTLC, densitometry, quantitative analysis, review

1b

## 2. Fundamentals, theory and general

- 106 010 V.G. BEREZKIN\*, Svetlana KHREBTOVA (\*Topchiev Inst., Petrochem. Syth., Russian Acad. Sci., Moscow 119991, Russia): Investigation of TLC in an N-Chamber. *Chromatographia*, 72(11-12), 1169-1176 (2010). Experimental and theoretical study of the adsorption kinetics of the gas phase of solvents most often used in TLC (methanol, acetic acid, ethyl acetate, toluene, chloroform, and acetone) in the N-chamber (the chamber most often used for analytical TLC, in 95 % of publications). To describe the kinetics an equation is proposed, which relates the solvent vapor adsorption to the time, the vapor pressure in saturated chambers and the diffusion coefficients of the solvents. It was found that as the characteristics of the adsorbent layer substantially improved by the manufacturers, the weight of mobile phase adsorbed by plates has increased several fold.

HPTLC

2c

- 106 011 M. KAMINSKA, Irena CHOMA\* (\*Department of Chromatographic Methods, University of M. Curie-Skłodowska, M. Skłodowska Sq. 3, 20-031 Lublin, Poland; [irena.choma@poczta.umcs.lublin.pl](mailto:irena.choma@poczta.umcs.lublin.pl)): The influence of perchlorate ion concentration on the retention of fluoroquinolones in RP-TLC. *J. Liq. Chromatogr. Relat. Technol.* 33, 894-902 (2010). Five amphoteric piperazynyl fluoroquinolones and flumequine were analyzed in an RP system on C8 plates with acetonitrile/aqueous acidic mobile phases containing various concentrations of potassium perchlorate. Perchlorate as a so-called chaotropic ion caused the increase in the retention of basic fluoroquinolones. TLC of sarafloxacin, difloxacin, norfloxacin, enrofloxacin, ciprofloxacin, and flumequine on RP-8 with acetonitrile - water containing constant concentration of citric acid but various concentrations of potassium perchlorate. After air-drying, fluoroquinolone zones were detected at 366 nm and 254 nm and flumequine only at 254 nm. The retention increased with the increasing concentrations of perchlorate ion in the mobile phase and achieved a plateau for the mobile phases containing 10-20 mM of chaotropic perchlorate.

qualitative identification

2d, 28a

- 106 012 C. SARBU\*, R. D. BRICIU (\*Babes-Bolyai University, Faculty of Chemistry and Chemical Engineering, Arany Janos Str. No. 11, 400028, Cluj Napoca, Romania; [csarbu@chem.ubbcluj.ro](mailto:csarbu@chem.ubbcluj.ro)):

Lipophilicity of natural sweeteners estimated on various oils and fats impregnated thin-layer chromatography plates. *J. Liq. Chromatogr. Relat. Technol.* 33, 903-921 (2010). A variety of TLC-plates impregnated with oils (paraffin, olive, sunflower, corn, castor, cod liver) and fats (margarine butter, pig, sheep, pullet, human) were indirectly evaluated regarding lipophilicity by employing a series of experimental parameters estimated for a representative group of natural sweeteners from retention data. TLC of 13 sugars on oil- and fat-impregnated silica gel layers using five organic solvents as organic modifiers. Best results were obtained with mixtures of acetonitrile and water with chamber saturation for 10 min. Detection of the sugars by treatment with silver nitrate and sodium hydroxide followed by heating at 105 °C for 5 min.

food analysis, qualitative identification

2d, 11

106 013 J. TRIFKOVIC, F. ANDRIC, P. RISTIVOJEVIC, D. ANDRIC, Z. TESIC, Dusanka MILOJKOVIC\* (\*Faculty Chemistry, University of Belgrade, 11158 Belgrade, Serbia, dusankam@chem.bg.ac.rs): Structure-retention relationship study of arylpiperazines by linear multivariate modeling. *J. Sep. Sci.* 33, 2619-2628 (2010). HPTLC of 33 newly synthesized arylpiperazines on RP-18 with 1) methanol - water (with increasing concentrations of methanol from 80-100 %), 2) dioxane - water (with increasing concentrations of dioxane from 60-80 %), and 3) dimethyl sulfoxide - water (with increasing concentrations of dimethyl sulfoxide from 80-100 %). In all cases the organic modifier was increased in steps of 5 %. Quantitative determination by absorbance measurement at 254 nm. A quantitative structure-retention relationship study allowed to understand the chromatographic behavior of similar compounds.

pharmaceutical research, HPTLC, quantitative analysis

2c

### 3. General techniques

106 014 Virginia COMAN\*, S. KREIBIK, M. VLASSA, M. FILIP (\*Babes-Bolyai University, Raluca Ripan Institute for Research in Chemistry, 40 Fântânele Street, 400294 Cluj-Napoca, Romania; coman\_virginia@yahoo.com, mvcoman@chem.ubbcluj.ro): Study of electric field geometry using the vertical planar dielectrochromatographic chamber. *J. Planar Chromatogr.* 23, 434-439 (2010). TLC of indophenol blue, Sudan red G, and 4-dimethylaminoazobenzene on aluminium oxide with toluene at 20 +/- 2°C with chamber saturation for 20 min. Experiments were done with a vertical PDEC chamber changing the geometry of the electric field to obtain the most favorable chromatographic results. The tests showed that different electric field geometries which do not cross the glass support of the TLC plates can be used. This results in enhancement of the intensity of the electric field in the chromatographic layer, especially for the spherical and conical armatures. With these two armatures the best resolution of the solutes were achieved.

planar electrochromatography

3d

106 015 Y.Q. CUI (Cui Yongquan), Z.H. GE (Ge Zhaohui), D.Y. WANG\* (Wang Dongyuan), B. YUAN (Yuan Bo) (\*Department of Analytical Chemistry, Shenyang Pharmaceutical University, Shenyang, 110016 CHINA; wdyxysy@hotmail.com): The behavior of an improved RP-18 sintered plate in planar electrochromatography. *J. Planar Chromatogr.* 23, 426-433 (2010). Mobile phases without addition of buffer salts could be successfully used in planar electrochromatography with a new type of RP-18 sintered plate, prepared from a 1:4 mixture of silica gel for TLC and powdered glass which was suspended in a solution of 0.3 % CMC-Na. The sintering step was the same as already described in *J. Planar Chromatogr.* 19, 313-318 (2006) except for the temperature which was 651 °C. A suitable degree of bonding of C18 was from 0.03 to 0.06, and good end-capping was achieved by placing the bonded sintered plates in a solution of dimethyldichlorosilane in toluene (ratio 1:100 to 1:1000) at room temperature for 20 min.

planar electrochromatography

3b



- 106 016 Ágnes M. MÓRICZ\*, H. KALÁSZ (\*Plant Protection Institute, Hungarian Academy of Sciences, Herman O. Str. 15, 1022 Budapest, Hungary; moricz\_am@nki.hu): Centrifugal layer chromatography - rotation planar chromatography. *J. Planar Chromatogr.* 23, 415-419 (2010). The paper summarizes the classification and applicability of centrifugal layer chromatography and rotation planar chromatography (RPC). The combination of TLC with centrifugal force resulted in the introduction of centrifugal layer chromatography which was renamed rotation planar chromatography because the abbreviation CLC had been used for column liquid chromatography. In analytical RPC three development modes are possible - circular, anticircular, and linear. On-line and off-line sample application, separation, and detection can be combined. RPC is suitable for analytical, micro-preparative, and preparative separations. The term RPC covers five basic techniques: normal chamber, micro-chamber, ultra-micro chamber, column and sequential RPC. Most RPC methods are applicable to preparative separation of a single sample. Analytical RPC is rarely used.

comparison of methods

3d

- 106 017 E. TYIHÁK\*, E. MINCSOVICS (\*Plant Protection Institute, Hungarian Academy of Sciences, Herman O. Str. 15, P. O. Box 102, 1525 Budapest, Hungary; etyih@nki.hu): Forced-flow planar liquid chromatographic techniques (after twenty-two years). *J. Planar Chromatogr.* 23, 382-395 (2010). The paper summarizes progress in the main forced-flow planar liquid chromatographic (FFPLC) techniques taking into account one group of FFPLC used in practice, OPLC and rotation planar chromatography and another group like e. g. electrochromatographic techniques and shear-driven chromatography which show interesting results at an experimental level. Progress in FFPLC deals with the diversity of further instrumental developments and its basis, the instrument developments, determination of the role of the adsorbent layer in OPLC, challenges in the OPLC instrument development, and analytical and preparative applications of OPLC. Centrifugal layer or rotation planar chromatography is mentioned briefly. Two electrochromatographic techniques have been developed to accelerate the mobile phase flow, planar electrochromatography and planar dielectrochromatography using direct and alternating currents, respectively. One of the newest techniques among FFPLC is shear-driven chromatography in which the mobile phase is between two plates and is forced to flow above and inside the very thin adsorbent layer with shear-driven force generated by a moving plate. For the study of the biological activity of natural and synthetic compounds the planar adsorbent layer is advantageous, namely for in-vitro and in-vivo studies.

review

3d

#### 4. Special techniques

- 106 018 L. BLUMBERG\*, M.S. KLEE (\*Fast GC Consulting, P.O. Box 1243, Wilmington, DE 19801, USA): A critical look at the definition of multidimensional separations. *J. Chromatogr. A* 1217(1), 99-103 (2010). Discussion of the definition of multidimensional (MD) separations, especially some potentially powerful separation techniques such as comprehensive 2D LC (LC × LC), and comprehensive 2D GC (GC × GC). The definitions of MD separations have been extended by some researches beyond their intended scope. This disqualifies comprehensive 2D techniques as LC × LC, GC × GC and 2D TLC from being considered as 2D techniques. In other instances, extended treatment of the definition is used as a basis to justify design-parameters of comprehensive 2D separations despite the fact that these parameters lead to sub-optimal implementations. The review draws attention to the shortcomings in the definition, discusses the weaknesses in the currently used definitions, and proposes to define n-dimensional analysis as one that generates n-dimensional displacement information.

doping, quantitative analysis, qualitative identification, review

4

106 019 Gertrud MORLOCK\*, W. SCHWACK (\*University of Hohenheim, Institute of Food Chemistry, Garbenstrasse 28, 70599 Stuttgart, Germany): Hyphenations in planar chromatography. J. Chromatogr. A 1217 (43), 6600-6609 (2010). A review on hyphenations of planar chromatography and its most important subcategory HPTLC. Examples from the field of natural product search, food, and lipid analysis point out the hyphenation with effect-directed analysis and mass spectrometry and illustrate the efficiency gain. Depending on the task at hand, hyphenations can readily be selected, for example with MS, bioassays etc. as required to reach the relevant information about the sample. At the same time, information is obtained for many samples in parallel. The flexibility and the unrivalled features through the planar format valuably assist separation scientists.

doping, HPTLC, qualitative identification, review

4e

106 020 O. OVCHINNIKOVA, G. VAN BERKEL\* (\*Organic and Biological Mass Spectrometry Group, Chemical Sciences Division, Oak Ridge National Laboratory, Oak Ridge, TN 37831-6131, USA, vanberkelgj@ornl.gov): Thin-layer chromatography and mass spectrometry coupled using proximal probe thermal desorption with electrospray or atmospheric pressure chemical ionization. Rapid Commun. Mass Spectrom. 24, 1721-1729 (2010). Ambient proximal probe thermal desorption (TD) sampling of substances from a HPTLC plate and coupled with secondary ionization by atmospheric pressure chemical ionization (APCI) or electrospray ionization (ESI). The method does not require a specialized ionization source. Different analytical parameters and performance metrics are reported and the method covers a wide range of analyte types including explosives, dyestuffs, herbicides and pharmaceuticals.

pharmaceutical research, environmental, herbal, agricultural, HPTLC, quantitative analysis, comparison of methods

4e

## 5. Hydrocarbons and halogen derivatives

106 021 T. BOROLE\*, R. MEHENDRE, M. DAMLE, K. BOTHARA (\*Dept. of Pharmaceutical Chemistry, AISSMS College of Pharmacy, Pune, Maharashtra, India): Development and validation of stability indicating HPTLC method for determination of prasugrel. Journal of Chemical and Pharmaceutical Research 2(4), 907-913 (2010). TLC on silica gel with dichloromethane - methanol 99:1. The  $hR_f$  value of prasugrel was  $58 \pm 3$ . Prasugrel was subjected to stress test conditions like acid, alkali, neutral hydrolysis, oxidation, dry heat, and photo degradation. The zones corresponding to degradation products were well resolved from the main drug. Densitometric evaluation in absorbance mode at 254 nm. Linearity was in the range of 300-1500 ng/band.

pharmaceutical research, densitometry

quantitative analysis, HPTLC

5c

106 022 R. PATEL\*, Mrunali PATEL, K. BHATT, B. PATEL (\*A. R. College of Pharmacy and G. H. Patel Institute of Pharmacy, Sardar Patel University, Vallabh Vidyanagar 388120, India, rbp.arcp@gmail.com): HPTLC method development and validation quantification of paliperidone in formulations and in vitro release study. Analytical Methods 2, 521-531 (2010). An HPTLC method for determination for paliperidone in formulation as well as for in vitro release studies has been developed. HPTLC on silica gel with methanol - ethyl acetate 4:1. The  $hR_f$  value was 54. Quantification was performed by densitometric evaluation at 254 nm. The method was linear in the range of 100-600 ng/band. The method was suitable for estimation of the drug in mucoadhesive microemulsion formulations, as well as for solubility and diffusion studies.

pharmaceutical research, quality control, densitometry, quantitative, analysis, HPTLC

5c

106 023 W. SOLOMON\*, M. MANU, R. SIVAKUMAR, P. ANAND, R. VENKATANARAYANAN (\*Dept. of Pharmaceutical Analysis, RVS College of Pharmaceutical Sciences, Suler, Coimba-

tore, T.N., India, samwd\_2000@yahoo.com): Application of TLC-densitometry method for estimation of acebrophylline in pharmaceutical dosage forms. *Journal of Pharmacy Research* 3(11) (2010). TLC on silica gel (plates pre-washed with methanol) with chloroform - isopropanol - toluene 8:1:1. The  $hR_f$  value was 22. Densitometric evaluation at 254 nm. The method was linear in the range of 100-5000 ng/band. The recovery was in the range of 99.8-101.5 %.

pharmaceutical research, quality control, densitometry, quantitative, analysis 5c

## 6. Alcohols

106 024 S. ARIYANATHAN\*, A. SARASWATHY, G. RAJAMANICKAM (\*Centre for Advanced Research in Indian system of Medicine (CARISM) SASTRA University, Thanjavur 613402, India, saraswathy20042000@yahoo.co.in): Quality control standards for the roots of three plumbago species. *Ind. J. Pharma. Sci.* 72(1), 86-91 (2010). Three species of Plumbago (Plumbaginaceae), i.e. *P. zeylanica*, *P. carpensis*, and *P. rosea* were studied for different physico-chemical parameters in addition to the estimation of microbial contamination, aflatoxins and pesticide residues and heavy metal content. All three species are used as herbs. The fingerprint profile of each species was compared using plumbagin as marker. Chloroform extracts of each plant were subjected to chromatography on silica gel with toluene - ethyl acetate 4:1 in a saturated twin trough chamber. Detection under UV 254 nm and 366 nm. The  $hR_f$  value of plumbagin was 70. The identity of plumbagin in the samples was shown by overlay of the UV spectra. Linearity was between 200 and 1000 ng/zone. The amount of plumbagin in the three species was between 0.01 and 0.17 %.

quality control, herbal, traditional medicine, densitometry, quantitative analysis 6

## 7. Phenols

106 025 V. DIGHE\*, G. CHAREGAONKAR (\*Dept. of Chemistry, Ramanarain Ruia College, Mantunga (E), Mumbai 400019, India): HPTLC quantification of sinigrin from seed powder of *Brassica juncea* (L.) czern and *Brassica nigra* (L.). *Asian J. Chem.* 22(7), 5349-5354 (2010). An HPTLC method is reported for estimation of sinigrin from seeds of *Brassica juncea* and *Brassica nigra* (Cruciferae). HPTLC of methanolic seed extracts on silica gel with ethyl acetate - methanol - water 6:2:1. Densitometric evaluation at 230 nm. The method was linear in the range of 400-1200 ng/band. Amounts of 0.43 and 1.55 % sinigrin were found in *Brassica juncea* and *B. nigra*, respectively. The method is suitable for routine quality control of herbal raw material.

traditional medicine, quality control, herbal, densitometry, quantitative analysis, HPTLC 7

106 026 Vidya DIGHE\*, G. CHAREGAONKAR (\*S. P. Mandal's Ramnarain Ruia College, Mantunga, Mumbai 400019, India): HPTLC quantitation of eugenol from leaf and berry powder of *Pimenta dioica* (L) merr. *Analytical Chemistry - An Indian Journal* 8(1), 29-33 (2009). A sensitive and accurate HPTLC method has been developed for the determination of eugenol from the leaf and berry powder of *Pimenta dioica* (L) Merr. The leaf and berry powders were extracted with methanol. HPTLC on silica gel with ethyl acetate - methanol - water 6:2:1. Detection and quantification by densitometry at 280 nm. Linear response to eugenol was found to be in the concentration range of 200-600 ng/band. The developed method can be used for routine quantitative monitoring of eugenol from the dried leaf and berry powder of *Pimenta dioica* (L) Merr.

quality control, herbal, densitometry, quantitative analysis, HPTLC 7

106 027 M. FAIYAZUDDIN\*, S. AHMAD, Zeenat IQBAL, Sushma TALEGAONKAR, F. AHMAD, Aseem BHATNAGAR, R. KHAR (\*Dept. of Pharmaceutical Faculty of Pharmacy, Hamdard University (Jamia Hamdard), Hamdard Nagar, New Delhi 110062, India): Stability indicating TLC method for determination of terbutaline sulphate in bulk and from submicronized dry pow-

der inhalers. *Analytical Sciences* 26 (4), 467-472 (2010). TLC of terbutaline sulphate on silica with chloroform - methanol 9:1. The  $hR_f$  value was 34. Densitometric evaluation at 366 nm. The method was linear in the range of 100–1000 ng/band. The recovery was being 99.6 %. When subjected to different stress conditions (acid, base, oxidative, and UV) the drug was not stable under acidic condition but stable under all other conditions. All degradation products are well resolved from the main compound. The proposed method is stability-indicating and suitable for quality control.

pharmaceutical research, quality control, densitometry, quantitative analysis

7

## 8. Substances containing heterocyclic oxygen

106 028 V. BORHADE\*, Hema NAIR, D. HEGDE, C. BARHADE (\*Dept. of Pharmacognosy & Phytochemistry, Bombay College of Pharmacy, University of Mumbai, Kalina, Santacruz (East), Mumbai, India): Development and validation of HPTLC method for estimation of tacrolimus in formulations. *Drug Development and Industrial Pharmacy* 35, 440-448 (2009). TLC of tacrolimus on silica gel with toluene - acetonitrile - glacial acetic acid 60:40:1. For quantitative evaluation the plate was derivatized with anisaldehyde sulfuric acid reagent and scanned at 675 nm. Using this method a compact spot was obtained at an  $hR_f$  value of 40. The linearity range was 100-800 ng/band. The method is suitable for routine analysis of formulations.

pharmaceutical research, quality control, densitometry, postchromatographic derivatization, quantitative analysis, HPTLC

8b

106 029 G. CHAKRABORTHY (SVKM'S, NMIMS University, School of Pharmacy and Technology Management, Shirpur Campus Shirpur, Maharashtra 425405, India): Quantitative estimation of ascorbic acid by HPTLC in different varieties of Amla. *J Young Pharm* 1(5), 82-85 (2010). HPTLC of ascorbic acid in different varieties of amla fruit (collected from different geographical regions) on silica gel with ethanol - acetic acid 19:1. The  $hR_f$  value of ascorbic acid was 76. Densitometric evaluation at 254 nm. The method was linear in the range of 1-5 µg/band. Bigger fruits were found to contain higher concentrations of ascorbic acid. The reported method was applied for estimation of ascorbic acid in crude drug as well as in herbal and pharmaceutical dosage form with reproducible results.

pharmaceutical research, herbal, food analysis, HPTLC

8b

106 030 P. HAMRAPURKAR\*, M. PHALE, N. SHAH (\*Dept. of Pharmaceutical Analysis, Prin. K. N. Kundanani College of Pharmacy, Jote Joy Bldg., R. S. Rd., Cuffe Parade, Coloba, Mumbai 400005, India): Quantitative estimation of efavirenz by high-performance thin-layer chromatography. *J Young Pharm* 1(4), 359-363 (2010). TLC of efavirenz on silica gel with acetonitrile - water - glacial acetic acid 12:8:1 in a twin-trough chamber saturated for 10 min. Densitometric evaluation at 247 nm. The method was linear in the range of 10-400 ng/band. The average recovery was 99.7 %.

pharmaceutical research, quality control, densitometry, quantitative analysis

8b

106 031 P. KHUSHBOO\*, V. JADHAV, V. KADAM (\*Dept. of Q. A., Bharati Vidyapeeth's College of Pharmacy, Sector 8, CBD Belapur, Navi-Mumbai 400614, India, drvmjadhav\_bvcop@rediffmail.com): Development and validation of a HPTLC method for determination of psoralen in *Psoralea corylifolia* (Bavachi). *International Journal of ChemTech Research* 1(4), 1122-1128 (2009). TLC on silica gel with toluene - ethyl acetate 3:1. The  $hR_f$  value of the furanocoumarin psoralen was 47. Densitometric evaluation at 299 nm. The method was linear in the range of 10-100 ng/band. The average recovery was 99.7 %.

herbal, densitometry, quantitative analysis

8b



- 106 032 U. PAWAR\*, A. SULEBHAVIKAR, A. NAIK, S. PINGALE, K. Mangaonkar (\*Dept. of Chemistry, Mithibai College of Arts, Chauhan Institute of Science & Amrutben Jivanlal College of Commerce & Economics, Vile Parle, Mumbai 400056, India): Simultaneous determination of rofecoxib and tizanidine by HPTLC. E-Journal of Chemistry 6(1), 295-302 (2009). HPTLC of rofecoxib and tizanidine on silica gel (plates pre-washed with methanol and dried at 110 °C) with toluene - ethyl acetate - methanol - triethyl amine 60:30:5:1 with chamber saturation for 15 min. The  $hR_f$  value of tizanidine was 49 and of rofecoxib 68. Rosiglitazone was used as internal standard. Densitometric evaluation at 235 nm. The method was linear in the range of 3.75-11.35 µg/band for rofecoxib and 0.30-0.70 µg/band for tizanidine. The recovery was 99.6-101.0 %.
- pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 8b,17a
- 106 033 S. RAKESH\*, P. PATIL, V. SALUNKHE, P. DHABALE, K. BURADE (\*Govt. College of Pharmacy, Vidyannagar, Tal. Karad 415124, Dist. Satara, M.S., India, sachinrakesh@rediffmail.com): HPTLC method for quantitative determination of quercetin in hydroalcoholic extract of dried flower of *Nymphaea stellata* willd. International Journal of ChemTech Research 1(4), 931-936, (2009). TLC of quercetin in hydroalcoholic extracts of dried flower of *Nymphaea stellata* (Nymphaeaceae). Separation on silica gel with toluene - ethyl acetate - formic acid 25:20:1. The  $hR_f$  value was 26. Densitometric evaluation at 380 nm. The method was linear in the range of 20-200 ng/band. The average recovery was 99.3 %.
- quality control, herbal, densitometry, quantitative analysis 8b
- 106 034 A. RAO\*, D. VARMA (\*Dept of Pharmaceutical Ana, Shri vishnu college of Pharmacy, Vishnupur, Bhimavaram 534 202 (A.P) India): Development and validation of HPTLC method for the simultaneous estimation of rofecoxib and tizanidine hydrochloride in tablet dosage form. Int. J. Chem Sci 7(2), 986-992 (2009). A validated HPTLC method is reported for estimation of rofecoxib and tizanidine hydrochloride in combined dosage form. HPTLC on silica gel with methanol - ethyl acetate 1:1. The  $hR_f$  value of tizanidine was 45 and of rofecoxib 67. Densitometric evaluation at 254 nm. The method was linear in the range of 180-260 ng/band for tizanidine and 2000-3000 ng/band for rofecoxib. Recovery was in the range of 99.5-102.5 % for both compounds. The method was suitable for routine quality control of combine dosage form.
- pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis 8b
- 106 035 R. THORAT\*, V. JADHAV, V. KADAM, S. KAMBLE & K. SALASKAR (\*Dept. of Q. A., Bharati Vidyapeeth's College of Pharmacy, Sector 8, CBD Belapur, Navi-Mumbai 400614, India, drvmjadhav\_bvcop@rediffmail.com): Development of HPTLC method for estimation of wedelolactone, quercetin and jatamansone in polyherbal formulation. International Journal of Chem-Tech Research 1(4), 1079-1086 (2009). TLC of wedelolactone, quercetin, and jatamansone in a polyherbal formulation. The formulation (oil) was extracted with methanol, the supernatant was concentrated under vacuum and used for chromatographic analysis. TLC on silica gel with toluene - acetone - formic acid 11:6:1 for wedelolactone, toluene - ethyl acetate - methanol 44:50:6 for quercetin and petroleum ether - acetone 3:1 for jatamansone. The  $hR_f$  value of jatamansone was 34, of wedelolactone 56, and of quercetin 47. Densitometric evaluation at 254 nm, 285 nm and 366 nm for quercetin, jatamansone, and wedelolactone respectively. The method was linear in the range of 500-2500 ng/band for wedelolactone, 3000-8000 ng/band for quercetin and 2000-6000 ng/band for jatamansone. The polyherbal oil formulation contained 1.62 %, 0.24 % and 0.11 % of wedelolactone, quercetin, and jatamansone respectively.
- herbal, clinical routine analysis, densitometry, quantitative analysis, HPTLC 8b

- 106 036 K. WANG (Wang Keqin)\*, J. LUO (Luo Junwu), J. CHEN (Chen Jingping), L. CHEN (Chen Liang) (\*Key Lab. of Tea Science, Ministry of Education, Hunan Agr. Univ., Changsha 410128, China): (Quantitative analysis of apigenin in celery by high-performance thin-layer chromatography (HPTLC) and high-performance liquid chromatography (HPLC)) (Chinese). Chinese J. Food Sci. 29 (4), 291-295 (2008). HPTLC of celery extracts on silica gel with chloroform - methanol - water 360:46:7. Detection and identification of the flavonoids by the characteristic chemical reaction colors and the UV spectrum of the prepared samples. Quantitative determination of apigenin by absorbance measurement at 345 nm. The linearity was between 330 and 990 ng/zone. Precision (% RSD) was 1.33 % ( $n = 5$ ) and the recovery was 97.8 % ( $n = 5$ ,  $RSD = 2.3$  %).

food analysis, HPTLC, densitometry, quantitative analysis, qualitative identification, comparison of methods

8a

## 10. Carbohydrates

- 106 037 J. ANTOSCH, N. HADZIFEJZOVIC\*, M. HUBBERT, L.-N. PRENNER, B. DONNER, J. SCHRAMM (\*Rottapharm/Madaus, Madaus GmbH, Cologne, Germany; hadnih@hotmail.de): Establishment of xylose in *Plantago ovata* forssk. as a leading compound for quantification in raw material and finished product. J. Liq. Chromatogr. Relat. Technol. 33, 996-1004 (2010). TLC of xylose with acetonitrile - water 9:1 with chamber saturation at ambient temperature. Detection by dipping into 4-aminobenzoic acid reagent for 1 to 2 s followed by heating at 110 °C for 10 min. The  $hR_f$  of xylose was 56. Quantitative determination by densitometric evaluation at 366 nm. The linearity determination coefficient was  $r^2 = 0.9999$ . The recovery of xylose was between 102 and 106 %. The precision of a six-time preparation was 1.6 %.

herbal, quality control, densitometry, quantitative analysis

10a

- 106 038 S. ARORA\*, P. SINGH, V. SHARMA, B. WADHWA, V. GEORGE, A. SINGH, G. SHARMA (\*Dairy Chemistry Div. National Dairy Research Institute, Karnal 132001, India, sumitak123@yahoo.com): Analysis of sucralose and its storage stability in burfi. J Food Sci Technol 46(2), 114-117 (2009). A method for isolation and analysis of the artificial sweetener sucralose from the Indian sweet Burfi has been developed. For the isolation samples were suspended in water, treated with  $K_3Fe(CN)_6$  solution and zinc sulfate, and filtrated. Semi-quantitative HPTLC analysis on amino layer with acetonitrile - water 4:1. The developed plate was heated at 190 °C for 20 min and spots were detected under UV 365 nm. Treating the plate with 5 % methanol CTMA increased the sensitivity (25 ng/zone). Quantitative HPTLC on silica gel with dichloromethane - methanol 4:1. Detection by spraying with 15 % methanolic sulfuric acid and heating at 100 °C for 10 min. The spot density was evaluated with Bio-rad fluorescence imaging software and confirmed by comparison with the standard. The method was linear in the range of 0.5-1.25  $\mu$ g/band.

food analysis, HPTLC, quantitative analysis, qualitative identification

10a

- 106 039 M. VLASSA, Virginia COMAN\*, M. FILIP, F. ONITA (\*Babes-Bolyai University - Raluca Ripan Institute for Research in Chemistry, 30 Fântânele Street, 400294 Cluj-Napoca, Romania; coman\_virginia@yahoo.com): OPLC of carbohydrate content for quality control of commercial Romanian wines. J. Planar Chromatogr. 23, 400-405 (2010). OPLC of fructose, glucose, and sucrose in 31 different types of wine (white and red; dry, semi-dry, semi-sweet, and sweet) on silica gel with acetonitrile - water 13:2 (as the best of 6 mobile phases tested) under isocratic conditions in overrunning operation mode. Detection by spraying with a mixture of aniline and diphenylamine. Quantitative determination by absorbance measurement at 420 nm.

food analysis, quality control, quantitative analysis, densitometry

10a

## 11. Organic acids and lipids

- 106 040 S. AHMAD\*, Y. KAMAL, M. SINGH, R. PARVEEN, M. MUSTHABA (\*Dept. of Pharmacognosy & Phytochem., Faculty of Pharmacy, Jamia Hamdard, New Delhi 110062, India, sahmajh@yahoo.co.in): HPTLC determination of gallic acid in crude drugs and herbal formulations. *Asian Journal of Chemistry* 23(1), 469-470 (2011). Several herbal formulations were analyzed for gallic acid contents. Tablets were powdered, subjected to hydrolysis by refluxing with 10 % HCl, filtered and extracted with chloroform. Acidic aqueous extracts were concentrated and the residue was taken up in methanol. TLC on silica gel with ethyl acetate - formic acid 85:11. Gallic acid was observed at an  $hR_f$  value of 89. Densitometric quantification of gallic acid at 272 nm. The method was linear in the range of 100-3000 ng/band. The method was suitable for analysis of formulations without interference from excipients. Gallic acid contents of different tablet samples varied from 0.06-0.15 % w/w.
- traditional medicine, herbal, densitometry, quantitative analysis 11a
- 106 041 S. AZHLWAR\*, T. RAVI (\*Dept. of Pharmaceutical Analysis, College of Pharmacy, Sri Ramkrishna Institute of Paramedical Science, Coimbatore, India): Simultaneous densitometric analysis of Drotaverine and aceclofenac by HPTLC method. *Scholars Research Library* 2(2), 328-332 (2010). A validated densitometric method has been developed for simultaneous estimation of drotaverine and aceclofenac in combined dosage form. TLC on silica gel with methanol - ethyl acetate - glacial acetic acid 100:90:1. The  $hR_f$  value of drotaverine was 18 and of aceclofenac 51. Densitometric evaluation at 300 nm. The method was linear in the range of 80-360 ng/band for drotaverine and 100-700 ng/band for aceclofenac. The average recovery was 99.8-100.2 %.
- pharmaceutical research, quality control, densitometry, quantitative analysis 11a
- 106 042 Girija BHAVAR, V. CHATPALLIWAR\*, D.PATIL, S. SURANA (\*Dept. of Pharmaceutical Chemistry, R. C. Patel College of Pharmacy Karvad Naka, Shirpur, Dist. Dhule 425405, vchatpalliwar@yahoo.co.in): Validated HPTLC method for simultaneous determination of quinapril hydrochloride and hydrochlorothiazide in a tablet dosage form. *Indian J. Pharma. Sci.* 70(4), 529-531 (2008). HPTLC on silica gel with ethyl acetate - acetone - acetic acid 13:6:1 with chamber saturation for 15 min. Densitometric evaluation at 208 nm. The  $hR_f$  value was 51 for quinapril hydrochloride and 76 for hydrochlorothiazide. The method was linear in the concentration range of 400-2800 ng/zone for quinapril and 500-3500 ng/zone for hydrochlorothiazide. The recovery was in the range of 98.3-100.8 %.
- pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 11a
- 106 043 J. BOLSTRIDGE, B. FRIED\*, J. SHERMA (\*Department of Biology, Lafayette College, Easton, PA 18042, USA, fried@lafayette.edu): Effects of temperature on the neutral lipid content of *Biomphalaria glabrata* as determined by high-performance thin-layer chromatography-densitometry and observations on snail fecundity. *J. Liq. Chromatogr. Relat. Technol.* 33, 1005-1012 (2010). HPTLC of lipids (free sterols, free fatty acids, triacylglycerols, methyl esters, and steryl esters) on silica gel (plates with concentration zone) with petroleum ether - diethylether - glacial acetic acid 80:20:1. Detection by spraying with 5 % ethanolic phosphomolybdic acid reagent and heating for 10 min at 110 °C. Quantitative densitometric analysis was performed at 610 nm.
- HPTLC, densitometry, quantitative analysis 11c
- 106 044 S. CHITALANGE\*, N. KUMAR, S. WANKHEDE (\*Dept. of Pharmaceutical Chem., Dr. D. Y. Patil Institute of Pharmaceutical Sciences & Research, Sant Tukaram Nagar, Pimpri 411018, India, sohanchitlange@rediffmail.com): Stability-indicating HPTLC method for estimation

of dexibuprofen in pharmaceutical dosage form. *Journal Pharmacy Research* 2(9), 1542-1546 (2009) TLC on silica gel with *n*-hexane - ethyl acetate - glacial acetic acid 75:25:2. The  $hR_f$  value was 38. Densitometric evaluation at 225 nm. The method was linear over a concentration range of 100-350 ng/band. The average recovery was 100.2 %. The samples were subjected to different stress conditions (acid, base, oxidative, heat) and all the degradation products were well resolved from the main compound. The method is suitable for stability studies and routine quality control.

pharmaceutical research, quality control, densitometry, quantitative analysis 11a

- 106 045 S. CHITLANGE\*, G. PAWBAKE, A. MULLA, S. WANKHEDE (\*Padm. DR. D. Y. Patil Institute of Pharmaceutical Sciences & Research, Pimpri, Pune 411018, M.S., India, sohanchitlange@rediffmail.com): Stability-indicating HPTLC method for estimation of diacerein in pharmaceutical dosage form. *Research Journal of Pharmaceutical, Biological and Chemical Sciences* 1(2), 226-234 (2010). TLC on silica gel with toluene - isopropanol - 25 % ammonia 23:23:4. The  $hR_f$  value was 30. Densitometric evaluation at 258 nm. The linearity range was 100-350 ng/band. For stability studies diacerein was subjected to different stress conditions (acid, alcohol, oxidative, thermal, photolytic). Degradation products were well resolved from the main zone of diacerein with significantly different  $hR_f$  values.

pharmaceutical research, quality control, quantitative analysis, densitometry 11a

- 106 046 Fatma HELMY\*, S. WHITE, S. AMIRI, R. AMIRI, A. SALIU (\*Department of Biological Sciences, Delaware State University, 1200 North Dupont Highway, Dover, DE 19901, USA; fhelmy@desu.edu): On the different lipolytic capability of diverse organs from young adult guinea pigs. A chromatographic study. *J. Planar Chromatogr.* 23, 277-281 (2010). TLC of cardiolipin, monolysocardiolipin, and phosphatidylethanolamin plasmalogen on silica gel and alumina pre-washed sequentially with chloroform - methanol 2:1 and acetone. For one-dimensional chromatography the mobile phase was 1-propanol - chloroform - ethyl acetate - methanol - water 50:50:50:21:18. Detection of phospholipids with thionine reagent. Two-dimensional chromatography in the first direction with the mobile phase mentioned before and in the second dimension with hexane - diethyl ether 4:1 after hydrolysis with 1 % hydrochloric acid to reveal alkenyl phospholipids. Detection by spraying with leucofuchsin reagent. TLC of choline lipids, phosphatidylcholine, and sphingomyelin on alumina with chloroform - methanol - water 65:30:4. Detection by treatment with Biebrich scarlet. Densitometric evaluation at 600 nm for thionine reagent and at 560 nm for leucofuchsin reagent.

densitometry, quantitative analysis, biochemical research 11

- 106 047 K. HUSSAIN\*, Z. ISMAIL, A. SADIKUN (\*Dept. of Pharmaceutical Chem. School of Pharmaceutical Science, University Sains Malaysia, Pulau Pinang 11800, Malaysia, hussain\_761@yahoo.com): High-performance thin-layer chromatographic method for quantification of betulinic acid in extracts of leaves of *Orthosiphon stamineus* benth. *Asian Journal of Chemistry* 23(2), 977-979 (2011). Part of the aqueous extract of *Orthosiphon stamineus* (Lamiaceae) was chromatographed on a silica gel column and eluted with *n*-hexane - ethyl acetate 3:2. The eluate was concentrated in order to obtain betulinic acid for use as standard solution. TLC on silica gel with *n*-hexane - ethyl acetate - formic acid 150:100:1. For derivatization the plate was sprayed with anisaldehyde reagent followed by heating at 100 °C for 10 min. Quantification of betulinic acid by densitometric measurement in fluorescence mode at 366 nm. The method was linear in the range of 0.2-500 µg/band. The recovery was 96.1-98.4 %.

traditional medicine, herbal, densitometry, quantitative analysis, preparative TLC 11a



- 106 048 Snehal INGALE\*, Dipali TAJANE, V. MODAK, S. GITE, V. CHOUDHARI, B. KUCHEKAR (\*Maharashtra Institute of Pharmacy, MIT Campus, Paud Raod, Kothrud, Pune, MS, India): Development and validation of a HPTLC method for simultaneous estimation of drotaverine hydrochloride and diclofenac potassium in combined dosage form. *Der Pharma Chemica* 2(5), 126-132 (2010). HPTLC of drotaverine hydrochloride (DRO) and diclofenac potassium (DFK) in bulk and pharmaceutical dosage form on silica gel with toluene - ethyl acetate - methanol 1:4:1. Densitometric evaluation at 298 nm. The  $hR_f$ -value was  $28 \pm 5$  for DRO and  $51 \pm 5$  for DFK. The linearity was in the range of 160-1280 ng/band and 100-800 ng/band. The recovery was in the range of 99.8-101.2 % for DRO and 98.3-101.4 % for DFK.
- pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis 11a
- 106 049 Varsha JADHAV\*, U. KEDAR, S. GHOLVE, V. KADAM (\*Dept. of Q. A., Bharati Vidyapeeth's College of Pharmacy, Sector 8, CBD Belapur, Navi-Mumbai 400614, India, drvmjadhav\_bv-cop@rediffmail.com): Development and validation of HPTLC method for determination of glycyrrhizin in herbal extract and in herbal gel. *International Journal of ChemTech Research* 1(4), 826-831 (2009). A herbal extract and herbal gel were extracted with 70 % ethanol, filtered and used for application. TLC on silica gel (plates pre-washed with methanol) with ethyl acetate - methanol - water - formic acid 15:2:1:1. The  $hR_f$ -value was 34. Densitometric evaluation at 252 nm. Glycyrrhizin monoammonium salt was used as standard. The method was linear in the range of 200-1000 ng/band, the recovery was 95.9-98.2 % . The glycyrrhizin contents in the extract and gel were in the range of 17.9-18.5 %.
- quality control, herbal, densitometry, quantitative analysis 11a
- 106 050 R. JESWANI\*, P. SINHA, K. TOPAGI, M. DAMLE (\* Dept. of Pharmaceutical Chem., A.I.S.S.M.S. College of Pharmacy, Pune 411001, India, mcdamle@rediffmail.com): A validated stability indicating HPTLC method for determination of cephalexin in bulk and Pharmaceutical formulation. *International Journal of PharmaTech Research* 03, 527-538 (2009). A stability indicating HPTLC method has been developed for analysis of cephalexin in bulk and dosage formulation. HPTLC on silica gel with ethyl acetate - methanol - 25 % ammonia 6:4:1. The  $hR_f$  value was 56. Densitometric quantification at 260 nm. The method was linear in range of 500-1500 ng/band. Cephalexin was subjected to forced degradation (acid, alkali, oxidation, thermal, photolytic). All degradation products were well separated from the drug, indicating specificity of the method.
- pharmaceutical research, densitometry, quantitative analysis, HPTLC 11a
- 106 051 R. KUMAR\*, N. SRISUTHERSON, P. NALLASIVAN, P. ARULRAJ, R. VENKATNARAYANAM (\*Dept. of Pharmaceutical Analysis, RVS College of Pharmaceutical Sciences, Suler, Coimbatore 641402, Tamil Nadu, India, andrilan@rediffmail.com): HPTLC method for the simultaneous estimation of aceclofenac and diacerein in tablets dosage forms. *Research J. Pharm. and Tech.* 3(3), 825-827 (2010). TLC on silica gel (plates pre-washed with methanol) with chloroform - methanol 4:1. The  $hR_f$  value of aceclofenac was 38 and of diacerein 66. Densitometric evaluation at 256 nm. The linearity range was 10-50  $\mu\text{g}/\text{band}$  for aceclofenac and 5-25  $\mu\text{g}/\text{band}$  for diacerein. The recovery was in the range of 99.3-101.8 % for both compounds. The method was suitable for routine quality control of combined dosage forms without any interference from the excipients.
- pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 11a
- 106 052 S. MANIMARAN\*, M.V.N.L. CHAITANYA, S. DHANABAL, B. SURESH, R. CHANDRASEKAR, M. NANJAN (\*Dept. for Phytopharm. & Phytomedicine TIFAC CORE in Herbal Drugs

J.S.S. College of Pharmacy, Ootacamund, Tamilnadu, India): Estimation of *n*-valeric acid content in *Valeriana officinalis* by HPTLC technique. *Indian Drugs* 47(3), 55-56 (2010). An HPTLC method is described for estimation of *n*-valeric acid in *Valeriana officinalis* (Valerianaceae). Methanolic extracts of the plant were subjected to chromatographic analysis on silica gel with toluene - ethyl acetate 3:1. Densitometric evaluation at 254 nm. The method was linear in the range of 500-1500 ng/band with a recovery between 98.4-104.1 %. The plant extract was found to contain 1.13 % of *n*-valeric acid.

herbal, densitometry, quantitative analysis, HPTLC

11a

- 106 053 I. MAREKOV\*, S. PANAYOTOVA, R. TARANDJIISKA (\*Institute of Organic Chemistry with Centre of Phytochemistry, Bulgarian Academy of Sciences, Acad. Georgi Bonchev Str., blok 9, 1113 Sofia, Bulgaria; ilko@orgchm.bas.bg): Silver ion TLC of minor triacylglycerol components for unambiguous detection of adulteration of olive oil with vegetable oil. *J. Liq. Chromatogr. Relat. Technol.* 33, 1013-1027 (2010). Preparative TLC of triacylglycerol (TAG) fractions on silica gel with hexane - acetone 25:4. Analytical TLC of TAG classes from sunflower, corn, soybean, cotton, and olive oil (differing in saturation) on silica gel, impregnated by dipping into a 5 % methanolic solution of silver nitrate, with petroleum ether - acetone - ethyl acetate 100:3:2 and 50:3:2. Detection by exposure to bromine and sulfuryl chloride vapor and heating at 180-200 °C. Quantitative determination by absorbance measurement at 450 nm.

food analysis, quality control, quantitative analysis, densitometry

11c

- 106 054 A. MOHAMMAD\*, S. SHARMA, S. BHAWANI (\*Analytical Research Lab., Dept. of Applied Chemistry, Faculty of Engineering & Technology, Aligarh Muslim Univ., Aligarh 202002, India, alimohammad08@gmail.com): Identification of ketoprofen in drug formulation and spiked urine samples by micellar thin-layer chromatography and its quantitative estimation by high performance liquid chromatography. *International Journal of ChemTech Research* 2(1), 89-96 (2010). A micellar thin layer chromatographic method has been reported for identification of ketoprofen in drug formulation and spiked urine samples, followed by quantification of the drug by HPLC both in formulation and spiked urine samples. TLC on amino layer with a micellar mobile phase of 0.5 % aqueous solution of sodium dodecyl sulphate -Triton X100 - acetone 16:10:3. The drug was extracted from spiked urine samples with ethylene dichloride containing 10 % each of isomyl alcohol and diethyl ether, then the organic phase was evaporated and the residue was taken up in acetonitrile. Derivatization by exposure to iodine vapor and by spraying with Dragendorffs reagent. The  $hR_f$ -value of ketoprofen was 50.

pharmaceutical research, comparison of methods, qualitative identification, postchromatographic derivatization

11a

- 106 055 Preeti NAYAK\*, S. UPADHYAYA, Anubha UPADHYAYA (\*Dept. of Crop & Herbal Physiology, College of Agriculture, JNKV, Jabalpur, M.P., India): A HPTLC densitometer determination of sinapic acid in *Chandrasur* (*Lepidium sativum*). *J. Sci. Res.* 1(1), 121-127 (2009). An HPTLC method is described for qualitative and quantitative estimation of sinapic acid in *Lepidium sativum* (Brassicaceae). Methanolic extracts of the plant material were subjected to chromatographic separation on HPTLC silica gel with *n*-butanol - acetic acid - water 4:1:5. Derivatization with anisaldehyde sulfuric acid reagent, followed by heating at 110 °C for 10 min. Densitometric quantification at 326 nm. Identification was confirmed by comparison of  $hR_f$ -values of sample and standard.

quality control, herbal, HPTLC, densitometry, qualitative identification, quantitative analysis, postchromatographic derivatization

11a

- 106 056 K. PAGI\*, S. LAHIRI, G. YADAV, Mamta SHAH (\*Dept. of Phytochem. and Pharmacognosy, L.M. College of Pharmacy, Navarangpura, Ahmedabad, Gujarat 380009, India, mbshah2007@rediffmail.com): Development and validation of HPTLC method for determination of betulinic acid in *Helicteres isora* root extract. International Journal of ChemTech Research 2(2), 851-855 (2010). Defatted powdered roots of *Helicteres isora* (Sterculiaceae) were extracted with ethyl acetate. TLC on silica gel with toluene - acetone - formic acid 25:5:2. The  $hR_f$  value was 43. Densitometric evaluation at 540 nm. The method was linear in the range of 100-500 ng/band. Recovery was 98.4-99.9 %.

pharmaceutical research, quality control, herbal, densitometry, quantitative analysis,  
HPTLC

11a

- 106 057 Marta PALUSINSKA-SZYSZ\*, M. JANCZAREK, R. KALITYNSKI, A. DAWIDOWICZ, R. RUSSA (\*Department of Genetics and Microbiology, Institute of Biotechnology and Microbiology, Maria Curie-Sklodowska University, Akademicka St. 19, Lublin, Poland, marta.szysz@poczta.umcs.lublin.pl): *Legionella bozemanii* synthesizes phosphatidylcholine from exogenous choline. Microbiol. Res. 166, 87-98 (2011). Two-dimensional TLC of phospholipids from *Legionella bozemanii* on chloroform - methanol - water 14:6:1 in the first dimension and chloroform - methanol - glacial acetic acid 13:5:2 in the second dimension. Lipids were detected by spraying with concentrated sulfuric acid or by exposure to iodine vapor. The following phospholipids were identified: phosphatidylcholine, phosphatidyl-N,N-dimethylethanolamine, phosphatidylethanolamine, phosphatidyl-N-monomethylethanolamine, phosphatidylglycerol and diphosphatidylglycerol.

pharmaceutical research, HPTLC, quantitative analysis, densitometry

11c

- 106 058 A. PAPRIKAR, K. PATIL, S. RANAFE, V. PANE, S. RAO, M. DAMLE\* (\*Dept. of Pharmaceutical Chemistry, A.I.S.S.M.S. College of Pharmacy, Kennedy Rd., Near R.T.O. Pune 411001, mcdamle@rediffmail.com): Stress degradation monitoring of ofloxacin by HPTLC and bioautography. Research J. Pharm. and Tech. 3(4), 1275-1278 (2010). A stability indicating HPTLC method has been developed for ofloxacin. The results were confirmed by bioautography. HPTLC on silica gel with *n*-butanol - ethanol - 25 % ammonia 5:5:4. The  $hR_f$  value was 53. Densitometric evaluation at 299 nm. The method was linear in the range of 500-2500 ng/band. The sample was subjected to forced degradation (acid, base, oxidation, thermal, photolytic) and both degraded and un-degraded sample were analyzed by HPTLC and bioautography. The result obtained by bioautography confirmed that the antimicrobial activity of ofloxacin was directly related to its degree of degradation under different stress conditions. Further no inhibition zone was observed at another  $hR_f$  than that of ofloxacin, which indicates that degradation products do not exhibit antibacterial activity. Bioautography studies were carried out in petri plates (10 cm diameter) using as media Mueller Hinton agar and as organism *Escherichia coli* NCIM 2066.

pharmaceutical research, quality control, densitometry, comparison of methods,  
quantitative analysis, HPTLC

11a

- 106 059 N. PATEL\*, V. JAIN, A. BHARGAV, D. SHAH (\*C. K. Pithawala Institute of Pharmaceutical Science & Research Surat 395007, Gujarat, India): High-performance thin-layer chromatographic method for quantification of gallic acid in bark powder of *Terminalia crenulata* Roth. Indian Drugs 47(6), 59-61 (2010). HPTLC of gallic acid in bark powder of *Terminalia crenulata* (Combretaceae) on silica gel with toluene - ethyl acetate - formic acid - methanol 60:60:16:4 with chamber saturation for 30 min. Densitometric evaluation at 254 nm. The method was linear in the range of 200-1200 ng/band. Samples extracted with different solvents were compared. Both alcoholic and water extracts were found to contain gallic acid where as it was absent in the chloroform extract.

pharmaceutical research, quality control, herbal, HPTLC, quantitative analysis,  
densitometry 11a

- 106 060 L. POTALE, Mrinalini DAMLE\*, A. KHODKE, K. BOTHARA (\*AISSIMS College of Pharmacy, Kennedy Rd., Near RTO Pune 411001, M.S., India, mcdamle@rediffmail.com): A validated stability indicating HPTLC method for simultaneous estimation of ramipril and telmisartan. International Journal of Pharmaceutical Sciences Review and Research 2(2), 35-39 (2010). TLC on silica gel with methanol - chloroform 1:6. The  $hR_f$  value was 38 and 68 for ramipril and telmisartan respectively. Densitometric evaluation at 210 nm. The method was linear in the range of 300-3000 ng/band and 500-4000 ng/band for ramipril and telmisartan respectively. The recovery was 98-102 %. The sample was subjected to different stress conditions (acid, base, oxidative, heat, photolytic) and all the degradation products were well separated from drug.

pharmaceutical research, quality control, densitometry, quantitative analysis 11a

- 106 061 Manisha PURANIK\*, D. BHAWSAR, Prachi RATHI, P. YEOLE (\*Institute of Pharmaceutical Education and Research, P. G. Dept. of Q. A., Borgaon (Meghe), Wardha 442001, India, manis-ha68\_12@yahoo.com): Simultaneous determination of ofloxacin and ornidazole in solid dosage form by RP-HPLC and HPTLC techniques. Ind. J. Pharma. Sc. 72(4), 513-517 (2010). TLC on silica gel with dichloromethane - methanol - 25 % ammonia 95:10:3. Ofloxacin and ornidazole were well separated. Linearity was in the range of 20-100 ng/band for ofloxacin and 50-250 ng/band for ornidazole. Recovery was in the range of 99.3-100.5 %.

pharmaceutical research, quality control, comparison of methods, quantitative analysis,  
densitometry 11a

- 106 012 C. SARBU et al., see section 2

- 106 062 A. RAO\* D. SUNEETHA (\*Dept. of Pharmaceutical Analysis, Shri Vishnu College of Pharmacy, Vishnupuri, Bhimavaram 5342012, India): High-performance thin-layer chromatographic estimation of simvastatin in bulk and tablet dosage form. Asian J. Chemistry 22(1), 27-30 (2010). HPTLC of simvastatin (in bulk and tablet dosage form) on silica gel with toluene - ethyl acetate - formic acid 10:3:1. The  $hR_f$  value of simvastatin was 26. Densitometric evaluation at 242 nm. The method was linear in the range of 200-1000 ng/band.

pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis 11a

- 106 063 A. RAO\*, N. PRUDHVI, K. BASAVARAJ, Y. MANOHARA (\*Dept. of Pharmaceutical Analysis, Shri Vishnu College of Pharmacy, Vishnupur, Bhimavaram 534202, AP, India): Development and validation of HPTLC method for the estimation of cefetamet. Rasayan J. Chem. 2(3), 720-723 (2009). HPTLC of cefetamet on silica gel with toluene - chloroform - methanol 3:6:1. The  $hR_f$  value of cefetamet was 35. Densitometric evaluation at 236 nm. The method was linear in the range of 1-5  $\mu\text{g}/\text{band}$ . The recovery was 99.3-101.5 %. The method was found suitable for routine quality control.

pharmaceutical research, quality control, densitometry, HPTLC, quantitative analysis 11a

- 106 064 P. RATHEE\*, Sushila RATHEE, Deepti AHUJA (no address given): Simultaneous quantification of glycyrrhetic acid and apigenin using HPTLC from Glycyrrhiza glabra linn. Eurasian J. Anal. Chem 5(1), 95-103 (2010). An HPTLC method is reported for quantification of glycyrrhetic acid and apigenin in Glycyrrhiza glabra. Methanolic and acidic-methanolic extracts are subjected to chromatographic separation on silica gel with ethyl acetate - ethanol - water - 25 % ammonia



63:20:4:1. The method was linear in the range of 160-960 ng/band for glycyrrhetic acid and 32-96 ng/band for apigenin. Glycyrrhiza glabra was found to contain 0.65 % of glycyrrhetic acid and 0.0004 % of apigenin.

quality control, herbal, densitometry, quantitative analysis, HPTLC 11a

106 065 R. RELE\*, S. SAWANT (\*Dept. for Chemistry, D.G. Ruparel College, Mahim, Mumbai 400016, India): Simultaneous determination of paracetamol and Ibuprofen from combined dosage formulation by HPTLC method. Analytical Chemistry - An Indian Journal 9(1) (2009). An HPTLC method has been developed for simultaneous estimation of paracetamol and ibuprofen. HPTLC on silica gel with ethyl acetate - acetone - *n*-butanol - 25 % ammonia 3:4:3:1. Densitometric quantification at 254 nm. The  $hR_f$  value of ibuprofen was 32 and of paracetamol 86. The method was linear in the range of 120-600 ng/band for paracetamol and 130-650 ng/band for ibuprofen.

pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 11a

106 066 C. ROOSEWELT\*, N. HARIHRISHNAN, V. GUNASEKARAN, S CHANDRASEKARAN, V. HARIBASKAR, B. PRATHAP (\*Dept. of Pharmaceutical Analysis, School of Pharmaceutical Sciences, Vel's University, Pallavaram, Chennai 600117, India): Simultaneous estimation and validation of tramadol and paracetamol by HPTLC in pure and pharmaceutical dosage form. Asian Journal of Chemistry 22(2), 850-854 (2010). HPTLC of tramadol and paracetamol in pharmaceutical dosage form on silica gel with chloroform - methanol - glacial acetic acid 90:20:1. The  $hR_f$  value of tramadol was 48 and of paracetamol 68. Densitometric quantification at 270 nm. The method was linear in the range of 500-2000 ng/band for both compounds. Recovery was 98.9-99.7 %.

pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis 11a

106 067 P. ROVINA, C. GRAF, F. BORNANCIN\* (\*Novartis Institutes for Biomedical Research, Brunnerstrasse 59, Vienna, Austria, frederic.bornancin@novartis.com): Modulation of ceramide metabolism in mouse primary macrophages. Biochem. Biophys. Res. Commun. 399, 150-154 (2010). HPTLC of the lipids from mouse primary macrophages on silica gel with butanol - acetic acid - water 3:1:1. Detection by imaging using Fujifilm intelligent dark box in SYBR green fluorescent light. Quantitative determination with a TLC plate imaging software.

pharmaceutical research, HPTLC, quantitative analysis, densitometry 11c

106 068 M. SAJEWICZ, D. KRONENBACH, M. GONTARSKA, Teresa KOWALSKA\* (\*Institute of Chemistry, University of Silesia, 9 Szkolna Street, 40-006 Katowice, Poland, teresa-kowalska@us.edu.pl): TLC and polarimetric investigation of the oscillatory in vitro chiral conversion of R-beta-hydroxybutyric acid. J. Liq. Chromatogr. Relat. Technol. 33, 1047-1057 (2010). TLC of R-beta-hydroxybutyric acid on silica gel pre-washed with methanol - water 9:1 and impregnated by dipping for 2 s in a 0.05 mol/L water - methanol solution of copper acetate. TLC was carried out at 22 +/- 1 °C with dioxane - water 9:1. Detection by spraying with 5 % sulfuric acid in ethanol and heating at 100-110 °C for 10 min. Quantitative determination by absorbance measurement at 326 nm. It was experimentally established that R-beta-hydroxybutyric acid can undergo oscillatory chiral conversion.

quantitative analysis, densitometry 11a

106 069 Smita SHARMA\*, M. SHARMA, D. KOHLI, A. SHARMA (\*Dept. of Pharmaceutical Sciences, Dr. Hari Singh Gour University, Sagar (M.P.), India): Development and validation of TLC-densitometry method for simultaneous quantification of montelukast sodium and levocetirizine

dihydrochloride pharmaceutical solid dosage form. *Scholars Research Library* 2(1), 489-494 (2010). HPTLC of montelukast sodium and levocetirizine dihydrochloride on silica gel with toluene - chloroform - methanol - glacial acetic acid 6:20:10:1 in a twin trough chamber with chamber saturation for 25 min. The  $hR_f$  value of levocetirizine was 64 and of montelukast 89. Densitometric evaluation at 302 nm. The method was linear in the range of 200-3200 ng/band for montelukast and 400-1200 ng/band for levocetirizine. The recovery was 99.9-100.0 %.

pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 11a

- 106 070 M. SHARMILA, S. SHARMA\* (\*Devi Ahilya Vishwavidyalaya Takshshila Campus, Khandwa Rd., Indore 452001, M.P., India, mukeshcsharma@yahoo.com): Development and validation of an HPTLC method for determination of oseltamivir phosphate in pharmaceutical dosage form. *Indian Drugs* 47(11), 68-72 (2010). TLC on silica gel with ethyl acetate - acetic acid - water 15:3:2. The  $hR_f$  value was 57. Densitometric evaluation at 265 nm. The method was linear in the range of 200-2400 ng/band. The method was suitable for routine quality control of the drug in formulation as there is no interference from excipients.

pharmaceutical research, quality control, densitometry, quantitative analysis 11a

- 106 071 P. SINHA\*, Munalini DAMLE, K. BOTHARA (\*AISSMS College of Pharmacy, Kennedy Rd., Pune 411001, India): A validated stability indicating HPTLC method for determination of aspirin and clopidogrel bisulphate in combined dosage form. *Eurasian J. Anal Chem* 4(2), 152-160 (2009). A stability indicating HPTLC method has been reported for estimation of aspirin and clopidogrel bisulphate in combined dosage form. HPTLC on silica gel with carbon tetrachloride - acetone 5:2. The  $hR_f$  value of aspirin was 13 and of clopidogrel 78. Densitometric evaluation at 220 nm. Linearity was in the range of 200-600 ng/spot for aspirin and 300-600 ng/spot for clopidogrel. Degradation products (acid, base oxidative, dry heat, photodegradation) did not interfere with the analysis.

pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 11a

- 106 072 Malgorzata STAREK\*, J. KRZEK (\*Department of Inorganic and Analytical Chemistry, Jagiellonian University, Collegium Medium, 9 Medyczna Str., 30-688 Cracow, Poland): TLC chromatographic-densitometric assay of ibuprofen and its impurities. *J. Chromatogr. Sci.* 48(10), 825-829 (2010). TLC of ibuprofen and its impurities in pharmaceutical preparations on silica gel with toluene - ethyl acetate - glacial acetic acid 17:13:1. Quantification by densitometry. The limit of detection and quantification ranges from 0.13-0.27  $\mu$  g/zone. The recovery is 96.8-99.0 % for the individual constituents. The method is suitable for routine quality-control analysis of pharmaceuticals containing ibuprofen.

pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC, qualitative identification 11a

- 106 073 M. TARES\*, L. BEBAWY, S. MOHAMMED (\*Analytical Dept. Faculty of Pharmacy, Cairo University, Egypt): Determination of lacidipine by thin-layer chromatographic densitometric, spectrophotometric method and modified vierordt's method in presence of its photodegradates. *Analytical Chemistry - An Indian Journal* 8(3) (2009) (without page number). A TLC method has been developed for determination of lacidipine in presence of its photodegradates. TLC on silica gel with toluene - acetone - methanol - 25 % ammonia 30:10:3:1. Densitometric evaluation at 284 nm. The chromatographic method was compared with a derivative spectrophotometric method and vierordt's method and was found suitable for analysis of lacidipine in bulk drug as well as in dosage form.

pharmaceutical research, quality control, comparison of methods,  
quantitative analysis, densitometry 11a

- 106 074 T. TOGAWA, I. KAWASHIMA, T. KODAMA, T. TSUKIMURA, T. SUZUKI, T. FUKUSHIGE, T. KANENURA, H. SAKURABA\* (\*Department of Analytical Biochemistry, Meiji Pharmaceutical University, Tokyo, Japan, sakuraba@my-pharm.ac.jp): Tissue and plasma globotriaosylsphingosine could be a biomarker for assessing enzyme replacement therapy for Fabry disease. *Biochem. Biophys. Res. Commun.* 399, 716-720 (2010). HPTLC of globotriaosylceramide (Gb3) in mouse tissues on silica gel with chloroform - methanol - water 65:25:4. Detection by HPTLC-immunostaining with an anti-Gb3 monoclonal antibody. Quantification by densitometry with a luminescent image analyzer. The limit of detection of Gb3 was 50 ng/zone.

pharmaceutical research, HPTLC, quantitative analysis, densitometry 11e

- 106 075 N. VEKARIYA\*, G. PATEL, H. BHATT, M. PATEL, R. DHOLAKIYA, G. RAMANI (\*Shree Dhanvantary Pharmacy College, Kim, Dist Surat, Gujarat, India, Smt B.N.B. Swaminarayan Pharmacy College, Salvav Dist. Valsad, Gujarat, India): Application of TLC-densitometry method for simultaneous estimation of telmisartan and amlodipine besylate in pharmaceutical dosage form. *International Journal of PharmTech Research* 1(4), 1644-1649 (2009). TLC of telmisartan and amlodipine besylate on silica gel with tetrahydrofuran - dichloromethane - methanol - 25 % ammonia 30:10:5:1. The  $hR_f$  value for telmisartan was 22 and for amlodipine 45. Densitometric evaluation in absorbance mode at 326 nm. The method was linear in the range of 1200-7200 ng/band for telmisartan and 400-1400 ng/band for amlodipine. The average recovery was 100.7-101.5 %.

pharmaceutical research, quality control, densitometry, quantitative analysis 11a

- 106 076 N. VEKARIYA\*, M. PATEL, G. PATEL, R. DHOLAKIYA (\*Shree Dhanvantary Pharmacy College, KIM, Smt. B.N.B. Swaminarayan Pharmacy College, Salvav, India, nitin.vekariya@gmail.com): Development and validation of TLC-densitometry method for simultaneous determination of telmisartan and amlodipine besylate in bulk and tablets. *J Young Pharm* 1(3), 259-263 (2009). A validated HPTLC method is described for simultaneous estimation of amlodipine besylate and telmisartan in dosage form. HPTLC on silica gel with tetrahydrofuran - dichloroethane - methanol - 25 % ammonia solution 60:20:10:4. The  $hR_f$  value of amlodipine besylate was 45 and of telmisartan 22. Densitometric evaluation at 326 nm. Linearity was in the range of 1200-7200 ng/band for telmisartan and 400-1400 ng/band for amlodipine besylate. The limit of detection was 149 ng/zone and 53 ng/zone for telmisartan and amlodipine besylate, respectively. The limit of quantification was 453 ng/zone for telmisartan and 161 ng/zone for amlodipine besylate. The recovery was between 100.4 and 100.8 %.

pharmaceutical research, quality control, HPTLC densitometry,  
quantitative analysis 11a, 23e

### 13. Steroids

- 106 077 C. ONISOR, M. POSA, S. KEVRESAN, K. KUHAJDA, C. SARBU\* (\*Babes-Bolyai University, Faculty of Chemistry and Chemical Engineering, Arany Janos 11, 400028 Cluj-Napoca, Romania, csarbu@chem.ubbcluj.ro): Estimation of chromatographic lipophilicity of bile acids and their derivatives by reversed-phase thin layer chromatography. *J. Sep. Sci.* 33, 3110-3118 (2010). HPTLC of bile acids and their derivatives on 1) RP-18, 2) RP-18W and 3) cyano phase with methanol - water mixtures. The volume fraction of organic solvent in the mobile phase ranged from 70-90 % for (1), 50-70 % for (2) and 45-65 % for (3). Detection by spraying with a solution of manganese chloride in sulfuric acid, followed by heating at 100-120 °C for 10 min. Quantifica-

tion by absorbance measurement at 254 nm and 365 nm.

pharmaceutical research, HPTLC, quantitative analysis, densitometry

13d

### 15. Terpenes and other volatile plant ingredients

106 078 Rani BHAGAT, D. KULKARNI\* (\*Botany Group, Agharkar Research Institute, Pune 411004, India, dilipkkulkarni@gmail.com): Quantification of beta-sitosterol from three *Jatropha* species by high-performance thin-layer chromatography. *Asian Journal of Chemistry* 22(10), 8117-8120 (2010). HPTLC of beta-sitosterol in leaves, roots and seed oil of 3 species of *Jatropha*. Chromatographic separation on silica gel with toluene - methanol 9:1. The  $hR_f$  value of beta-sitosterol was 54. Derivatization with anisaldehyde-sulfuric acid reagent, followed by heating at 120 °C. After derivatization densitometric evaluation at 525 nm. The method was linear in the range of 100-500 ng/band. The proposed method was used for the estimation of beta-sitosterol in extracts of different species of plants. The presence of beta-sitosterol in samples was confirmed by multi-wave length scanning.

traditional medicine, quality control, herbal, HPTLC, densitometry, quantitative analysis 15a

106 079 Ágnes M. MÓRICZ\*, G. HORVÁTH, P. MOLNÁR, B. KOCSIS, A. BÖSZÖRMÉNYI, É. LEMBERKOVICS, P. G. OTT (\*Plant Protection Institute, Hungarian Academy of Sciences, Herman O. Str. 15, 1022 Budapest, Hungary; moricz\_am@nki.hu): Investigation of thyme (*Thymus vulgaris* L.) essential oil by use of the BioArena system. *J. Planar Chromatogr.* 23, 406-410 (2010). TLC and OPLC of essential oil of thyme (thymol, carvacrol, and linalool as standards) on silica gel with chloroform (previously extracted with a 0.1 % aqueous solution of sodium hydrogen carbonate and dried on sodium sulfate to eliminate the stabilizer amylene, free hydrochloric acid, and chlorine) in an unsaturated chamber. Detection at 254 nm, by spraying with vanillin - sulfuric acid reagent (50 mg vanillin with 12 mL ethanol and 200 mL 98 % sulfuric acid) and heating to 70 °C for 10 min, and by use of the BioArena system. Quantitative determination by densitometry at 275 and 600 nm (dual-wavelength measurement).

herbal, traditional medicine, densitometry, quantitative analysis,  
qualitative identification

15b

106 080 F.A. MEHTA\*, B.G. PATEL\*, S.S. PANDYA, K.B. AHIR (\*Indukaka Ipcowala College of Pharmacy, P. O. Box 53, PO Vithal Udyognagar, Beyond GIDC Phase IV, New Vallabh Vidyanagar, Gujarat 388 121, India; fm999@ymail.com): Densitometric HPTLC method for analysis of oleonic acid in *Achyranthes aspera* L. *J. Planar Chromatogr.* 23, 289-292 (2010). HPTLC of oleonic acid on silica gel pre-washed with methanol and acetone using toluene - ethyl acetate - formic acid 9:1:2 in a twin-trough chamber saturated for 30 min. Detection by spraying with 10 % sulfuric acid in ethanol, followed by heating at 130 °C for 3 min. Quantitative determination by densitometry at 490 nm. Linearity was between 600 and 1000 ng/band; the correlation coefficient was 0.994. The limit of detection and quantification was 27 and 82 ng/band. The repeatability (%RSD,  $n = 6$ ) was 0.61 %, and the recovery was between 97.8 and 99.2 %. The inter-day and intra-day precision (RSD,  $n = 3$ ) was 1.25 and 0.83 % (600 ng/band), 1.00 and 1.03 % (800 ng/band), and 1.11 and 0.81 % (1000 ng/band).

herbal, traditional medicine, densitometry, quantitative analysis,  
qualitative identification

15a

106 081 S. MUSTHABA\*, M. ATHAR, S. BABOOTA & S. AHMAD (\*Dept. of Pharmaceutics, Faculty of Pharmacy, Jamia Hamdard, New Delhi 110062, India, sahmajh@yahoo.co.in): HPTLC analysis of neem oil in herbal dosage form. *Asian Journal of Chemistry* 23(1), 385-387 (2011). HPTLC on silica gel with chloroform - ethyl acetate containing 1 % acetic acid. Two well resol-

ved zones with  $hR_f$  values of 33 and 55 were obtained by illumination at 254 nm. The zones were labelled as substance I and II and used for standardization of the oil. Densitometric evaluation at 265 nm. The linearity range for both substances was 4-100  $\mu\text{g}/\text{band}$ . The recovery was in the range of 97.4-98.7 %. Several commercially available tablets and capsules were analyzed for the content of Neem oil using substances I and II as marker (in the absence of chemical markers).

herbal, traditional medicine, densitometry, quantitative analysis, qualitative identification, radioscanning 15b

- 106 082 A. PAREKH, V. JADHAV\* (\*Dept. of Pharmacology Bharati Vidyapeeth's College of Pharmacy, Navi Mumbai, India, drvmjadhav\_bvcop@rediffmail.com): Development of validated HPTLC method for quantification of jatamansone in jatamansi oil. Journal of Pharmacy Research 2(5), 975-977 (2009). Quantitative analysis of jatamansone in jatamansi oil obtained by steam distillation of rhizome extracts of Nardostachys jatamansi (Valerianaceae). HPTLC on silica gel with petroleum ether - acetone 3:1 in a saturated twin trough chamber. The  $hR_f$  value of jatamansone was 43. Densitometric evaluation at 285 nm. The method was linear in the range of 250-1500  $\text{ng}/\text{band}$ . The recovery was 98.8-102.2 %. The oil was found to contain 20.3 % of jatamansone.

herbal, HPTLC, quantitative analysis, densitometry 15b

- 106 083 K.C. PATRA\*, K.J. KUMAR (\*Institute of Pharmaceutical Sciences, Guru Ghasidas Viswavidyalaya (Central University), Bilaspur 495009, India: herbalkartik@gmail.com): A validated HPTLC method for simultaneous analysis of eugenol and piperine in a Siddha formulation. J. Planar Chromatogr. 23, 293-297 (2010). TLC of eugenol and piperine on silica gel with toluene - ethyl acetate 9:3 with chamber saturation for 15 min at room temperature. The  $hR_f$  value of eugenol was 73 and of piperine 28. Linearity was between 1 and 12  $\mu\text{g}/\text{band}$  for both eugenol and piperine. A good linear relationship with  $r^2 = 0.9979$  and  $0.9980$  for eugenol and piperine was obtained. The limit of detection was 0.1  $\text{ng}/\text{band}$  for eugenol and 10  $\text{ng}/\text{band}$  for piperine. Recovery of eugenol and piperine was above 97 %. The recovery (measured at different concentration levels) of eugenol was 95.9- 97.5 % and of piperine 91.8-96.6 %. For eugenol the robustness of the method (%RSD,  $n = 3$ ) was 0.74 % (12  $\text{ng}/\text{band}$ ) and 0.69 % (25  $\text{ng}/\text{band}$ ) for toluene - ethyl acetate 9:3 and 0.79 % (12  $\text{ng}/\text{band}$ ) and 0.88 % (25  $\text{ng}/\text{band}$ ) for toluene - ethyl acetate 9:2. For piperine the robustness of the method (%RSD,  $n = 3$ ) was 0.74 % (5  $\text{ng}/\text{band}$ ) and 0.89 % (10  $\text{ng}/\text{band}$ ) for toluene - ethyl acetate 9:3 and 0.72 % (5  $\text{ng}/\text{band}$ ) and 0.73 % (10  $\text{ng}/\text{band}$ ) for toluene - ethyl acetate 9:2.

herbal, traditional medicine, quality control, HPTLC, densitometry, quantitative analysis 15

- 106 084 M. SAJEWICZ, L. WOJTAL, D. STASZEK, M. HAJNOS, Monika WAKSMUNDZKA-HAJNOS, Teresa KOWALSKA\* (\*Institute of Chemistry, University of Silesia, 9 Szkolna Street, 40-006 Katowice, Poland; teresa.kowalska@us.edu.pl): Low temperature planar chromatography - densitometry and gas chromatography of essential oils from different sage (Salvia) species. J. Liq. Chromatogr. Relat. Technol. 33, 936-947 (2010). TLC of essential oils (multicomponent mixtures of mono-, di, tri-, and sesquiterpenes) from Salvia species at  $22 \pm 1$  °C and at  $-10 \pm 0.5$  °C on silica gel with toluene - ethyl acetate 19:1 in sandwich chambers saturated with mobile phase for 15 min. Quantitative absorbance measurement at 340 nm. Also preparative TLC after bandwise application at  $-10 \pm 2$  °C in saturated sandwich chambers. After extraction of the separated bands GC with mass spectrometric detection was performed. Low temperature TLC densitometry can successfully be used for fingerprinting of essential oils contained in the different sage species.

pharmaceutical research, herbal, traditional medicine, preparative TLC, densitometry, qualitative identification, quantitative analysis 15b

- 106 085 M. SAJEWICZ, L. WOJTAL, M. HAJNOS, Monika WAKSMUNDZKA-HAJNOS, Teresa KOWALSKA\* (\*Institute of Chemistry, Silesian University, 9 Szkolna Street, 40-006 Katowice, Poland; teresa.kowalska@us.edu.pl): Low-temperature TLC-MS of essential oils from five different sage (*Salvia*) species. *J. Planar Chromatogr.* 23, 270-276 (2010). TLC of essential oils (obtained from different sage species by vapor distillation) on silica gel at  $-10 \pm 0.5$  °C with toluene - ethyl acetate 19:1 in sandwich chamber saturated with mobile phase for 15 min. Quantitative absorbance measurement at 340 nm and 254 nm. After densitometry mass spectrometric analysis was performed using a TLC-MS Interface for direct elution of a given band from the plate.  
herbal, qualitative identification, densitometry 15b
- 106 086 S. SAPANA, V. JADHAV\*, V. KADAM (\*Dept. of Q. A., Bharati Vidyapeeth's College of Pharmacy, Sector 8, CBD Belapur, Navi-Mumbai 400614, India, drvmjadhav\_bvcop@rediffmail.com): Development and validation of HPTLC method for determination of 3-hydroxy androstane [16,17-C] (6-methyl, 2'-1-hydroxy-isopropene-1-yl) 4,5,6 H pyran in jambul seed (*Syzygium cumini*). *International Journal of ChemTech Research* 1(4), 1129-1135 (2009) The marker compound was first isolated by column chromatography over silica gel by elution with toluene - ethyl acetate 17:3. TLC of ethanolic extracts of *Syzygium cumini* seed on silica gel with toluene - ethyl acetate 17:3. The  $hR_f$  value of the marker compound was 50. Densitometric evaluation in fluorescence mode at 366 nm. The method was linear in the range of 1-5 µg/band. The extract of the powdered sample contained 7.4 % of the marker compound.  
quality control, herbal, environmental, densitometry, quantitative analysis 15
- 106 087 S. SARFARE\*, S. MENON, S. SHAILAJAN (\*Herbal Research Lab, Ramnarain Ruia College, Mumbai 400019, India): Cornsilk as a bioavailable source of beta-sitosterol: a pharmacokinetic study using HPTLC. *Asian Journal of Plant Sciences* 9(1), 44-50 (2010). Beta-sitosterol is one of the major phytoconstituents in cornsilk (style and stigma of *Zea mays* Linn.). An HPTLC method for the standardization of cornsilk as a bioavailable source of beta-sitosterol is reported. HPTLC of methanolic extracts of dried cornsilk and plasma of rabbits (1 h after injection of plant slurry) on silica gel with toluene - ethyl acetate - methanol - glacial acetic acid 80:10:5:3 with chamber saturation for 30 min. Detection by treatment with Liebermann-Burchard reagent. Beta-sitosterol was well resolved with an  $hR_f$  value of 48. Quantitative determination by fluorescence measurement at 366 nm. The method was suitable for the pharmacokinetic study (absorption - elimination). The study confirmed the bioavailability of beta-sitosterol from cornsilk, which is therefore a potential natural source of beta-sitosterol.  
herbal, clinical routine analysis, clinical chemistry research, HPTLC 15a
- 106 088 Willy SHAH\*, S. PEDNEKAR, Sunita SHAILAJAN, V. VAIDYA (\*Ramnarain Ruia College, Matunga, Mumbai, India): A rapid densitometric method for simultaneous quantification of two biologically active compounds in *Vernonia cinerea* whole plant powder using HPTLC. *Analytical Chemistry - An Indian Journal* 8(4), 608-612 (2009). An HPTLC method is reported for estimation of lupeol and beta-sitosterol from the whole plant of *Vernonia cinerea* (Acanthaceae). Methanolic extracts of the plant were subjected to chromatographic analysis on silica gel with toluene - methanol 220:3. Derivatization with Liebermann-Burchard reagent. Densitometric evaluation at 366 nm. The plant was found to contain 0.49 mg/g and 1.4 mg/g of lupeol and beta-sitosterol respectively. The method was suitable for quality control of herbal raw material.  
quality control, herbal, densitometry, HPTLC, postchromatographic derivatization, quantitative analysis 15a
- 106 089 Smruti SHAH\*, Sunita SHAILAJAN (\*Herbal Research Lab, Ramnarain Ruia College, Matunga,



Mumbai, India): Quantitation of beta-sitosterol from *Woodfordia fruticosa* (Linn.) Kurz and a polyherbal formulation used for treating female reproductive disorders. *Analytical Chemistry - An Indian Journal* 8(1), 82-86 (2009). An HPTLC method is reported for estimation of beta-sitosterol in flowers of *Woodfordia fruticosa* (Lythraceae) and its polyherbal formulation. HPTLC on silica gel with toluene - ethyl acetate - methanol - glacial acetic acid 80:10:5:3. Methanolic extracts of the sample were used for chromatographic separation. Derivatization with Liebermann-Burchard reagent, densitometric quantification at 366 nm. The linearity range was 100-700 ng/band. The flowers of the plant contained 0.18 µg/mg beta-sitosterol and the tablet herbal formulation contained 0.15 µg/mg.

traditional medicine, quality control, herbal, densitometry, quantitative analysis, postchromatographic derivatization, HPTLC

15a

106 090 S. SHAIKH\*, K. MANGAONKAR (\*Analytical Chemistry lab., Mithibai College of Arts, Chauhan Institute of Science & Amrutben Jivanlal College of Commerce & Economics, Vile Parle (W) Mumbai 400056, India): High-performance thin-layer chromatographic method for quantification of beta-sitosterol from *Tridax procumbens*. *Analytical Chemistry - An Indian Journal* 9(2) (2010) (without page number). An HPTLC method has been reported for estimation of beta-sitosterol in whole plant powder of *Tridax procumbens*. HPTLC on silica gel with toluene - ethyl acetate - glacial acetic acid 40:10:1. Methanolic extracts of the plant material were used. For detection the plate was sprayed with anisaldehyde - sulfuric acid reagent. Densitometric quantification at 550 nm. The method was linear in the range of 10.0-70.0 µg/band. The plant material was found to contain 0.04 % of beta-sitosterol. The proposed method was suitable for routine quality control of plant as well as quantification of beta-sitosterol .

quality control, herbal, densitometry, quantitative analysis, HPTLC

15a

106 091 Sunita SHAILAJAN\*, S. MENON (\*Herbal Research Lab, Ramnarain Ruia College, Matunga, Mumbai, India): Simultaneous quantitation of lupeol and beta-sitosterol from the whole plant powder of *Asteracantha longifolia* Nees. *Analytical Chemistry - An Indian Journal* 8(1), 77-81 (2009). An HPTLC method is reported for estimation of lupeol and beta-sitosterol in the whole plant powder of *Asteracantha longifolia* (Acanthaceae). The proposed method was also employed for estimation of beta-sitosterol in herbal formulation containing *Asteracantha longifolia*. Methanolic extracts of the sample were used for chromatography. HPTLC on silica gel with toluene - ethyl acetate - methanol 75:15:7. Derivatization with Liebermann-Burchard reagent. Densitometric quantification at 366 nm. The concentration of lupeol and beta-sitosterol was 0.162 mg/g and 0.045 mg/g, respectively in the whole plant, while the concentration of beta-sitosterol was 0.039-0.048 mg/g in the formulation. The method was suitable for the quality control of the herbal formulation.

herbal, HPTLC, densitometry, postchromatographic derivatization, quantitative analysis 15a

106 092 Maria-Loredana SORAN\*, I. LUNG (\*National Institute of Research and Development for Isotope and Molecular Technology, 65-103 Donath Street, 400293 Cluj-Napoca, Romania; loredana\_soran@yahoo.com): HPTLC analysis of thymol in extracts of *Satureja hortensis* L. obtained by different techniques. *J. Planar Chromatogr.* 23, 320-322 (2010). HPTLC of thymol in *Satureja hortensis* L. extracts on silica gel with toluene - ethyl acetate 93:7. Detection by spraying with anisaldehyde reagent. Quantitative determination by absorbance measurement at 513 nm. The quantity of thymol extracted depends on the composition of the extraction solvent and the extraction technique. The best results were obtained by maceration with ethanol.

traditional medicine, herbal, quality control, HPTLC, densitometry, quantitative analysis 15

**17. Amines, amides and related nitrogen compounds**

- 106 093 Anindita BEHERA\*, D. SANKAR, S. CHANDRA (\*Dept. of Pharmaceutical Science Andhra University Waltair, Visakapatnam 530003, AP, India): Development and validation of an HPTLC - densitometric method for determination of levodopa in seeds of *Mucuna pruriens* and its dosage form. *Eurasian J. Anal. Chem.* 5(1), 25-27 (2010). An HPTLC method is reported for estimation of levodopa in seeds of *Mucuna pruriens* and its herbal formulation. HPTLC on silica gel with *n*-butanol - glacial acetic acid - water 5:1:4. Densitometric evaluation at 280 nm. The  $hR_f$  value of levodopa was 39. The method was linear in the range of 100-1000 ng/band. The method is suitable for estimation of levodopa in herbal formulation.
- quality control, herbal, densitometry, quantitative analysis, HPTLC 17c
- 106 094 R. CHAKRABORTY\*, K. PAL, M. SHAIBA, N. SANGEPU, P. SRIDEVI (\*CM College of Pharmacy, Maisammaguda, Hakimpet Post, Komapally, Secunderabad 500014, A.P., India): High-performance thin-layer chromatographic estimation of ranolazine. *Research Journal of Pharmaceutical, Biological and Chemical Sciences* 1(4), 152-157 (2010). TLC on silica gel with methanol - 10 mM ammonium acetate 3:2. The  $hR_f$  value of ranolazine was 49. Densitometric evaluation at 271 nm. The method was linear in the range of 1-6 µg/band with a mean recovery of 100.0 %.
- pharmaceutical research, quality control, densitometry, quantitative analysis 17
- 106 095 P. DESHPANDE, S. GANDHI\*, Vandana BHAVNANI, R. BANDEWAR, A. DHIWARE, Vrushali DIWALE (\*Dept. of Q.A. AISSMS College of Pharmacy, Kennedy Rd., near R.T.O. Pune 411001, M.S., India, santoshgandhi@rediffmail.com): High-performance thin-layer chromatographic determination of famotidine and domperidone in combined tablet dosage form. *Research Journal of Pharmaceutical, Biological and Chemical Sciences* 1(4), 354-359 (2010). TLC on silica gel with toluene - methanol - triethylamine 12:6:1 with chamber saturation for 15 min. Densitometric evaluation at 290 nm. The  $hR_f$  value was 23 and 67 for famotidine and domperidone, respectively. The method was found to be linear in the range of 100-500 ng/band. The recovery was 98.8 %.
- pharmaceutical research, densitometry, quantitative analysis 17c
- 106 096 M. FAISAL\*, N. ZRINE, S. FAIYAZ, A. SAYEED, K. KOHLI, R. KHAR (\*Dept. of Pharmaceutics, Faculty of Pharmacy, Jamia Hamdard Nagar, New Delhi 110062, India): A new TLC densitometric method for stability assessment of modafinil. *Chem. Anal. (Warsaw)* 54, 1-12 (2009). An HPTLC method has been developed for estimation of modafinil in tablet formulation and stability assessment. HPTLC on silica gel with toluene - chloroform - methanol 2:2:1. The  $hR_f$  value of modafinil was 46. Densitometric evaluation at 220 nm. The method was linear in the range of 100-500 ng/band. The sample compound was well separated from all degradation products. The method was suitable for analysis and also for stability studies.
- pharmaceutical research, quality control, densitometry, HPTLC, quantitative analysis 17c
- 106 097 N. JAIN,\* G. JAIN, Zeenat IQBAL, Sushma TALEGAONKAR, F. AHMAD, R. KHAR (\*Dept. of Pharmaceutical, Faculty of Pharmacy, Jamia Hamdard, New Delhi 110062, India): Retraction: an HPTLC method for the determination of minocycline in human plasma, saliva and gingival fluid after single step liquid extraction. *Analytical Science* 25(1), 57-62 (2009). HPTLC of minocycline on silica gel plates previously treated with 10 % EDTA solution of pH 9.0 with methanol - acetonitrile - isopropyl alcohol - water 10:8:1:1. The  $hR_f$  value was 32. Densitometric evaluation at 345 nm. The method was linear in the range of 100-1200 ng/band for all biological samples. The average recovery was 95.1 %. The method was found suitable for estimation of the drug in

different biological fluids (human plasma, saliva, gingival fluid) and can be used for pharmacokinetic studies.

clinical routine analysis, densitometry, quantitative analysis, HPTLC 17c

- 106 098 D. PATEL\*, N. PATEL (\*Dept. of Pharmaceutical Chemistry, S. K. Patel College of Pharmaceutical Education & Research, Ganpat University, Kherva, Mehsana 382711, Gujarat, India, diptibpatel\_24980@yahoo.co.in): Validated stability indicating HPTLC method for the determination of tamsulosin hydrochloride in pharmaceutical dosage forms. International Journal of ChemTech Research 2(1), 646-652 (2010). TLC on silica gel with toluene - methanol - triethylamine 9:3:1 with chamber saturation for 30 min. The  $hR_f$ -value was 71. Densitometric evaluation at 282 nm. The method was linear in the range of 100-2000 ng/band with  $r^2 = 0.9973$ . The repeatability of sample application and measurement of peak area (%RSD,  $n = 6$ ) was 0.461 % and 0.363 %. The %RSD for intra-day and inter-day variation was 0.752-0.961 % and 0.848-1.082 % respectively. The LOD and LOQ was 10 and 50 ng/zone, respectively. The average recovery was 99.5 %. The samples were subjected to stress conditions (acid, base, oxidative, thermal) and all degradation products were well resolved from the main compound. The method was found suitable for stability studies and for routine analysis of the drug in biological fluids.

pharmaceutical research, quality control, densitometry, quantitative analysis 17c

- 106 099 A. PATIL\*, Darshana PATIL, A. SHARMA, N. CHANDRA (\*Dept. of Botany-Herbal Sciences, Birla College, Kalyan 421304, India, dravinashpatil@rediffmail.com): Quantification of beta-carotene from *Diplocyclos palmatus* jeff. fruits rind by using high-performance thin-layer chromatography. Asian Journal of Chemistry 23(2), 788-790 (2011). A simple HPTLC method has been developed for estimation of beta-carotene in fruit rind of *Diplocyclos palmatus* (Cucurbitaceae). The rind of fruits was extract with acetone. HPTLC on silica gel with petroleum ether as mobile phase. The  $hR_f$ -value of beta-carotene was 30. Densitometric evaluation at 450 nm. The method was linear in the range of 6-60 ng/band. The recovery was 99.4 % for beta-carotene.

traditional medicine, herbal, HPTLC, densitometry, quantitative analysis 17a

- 106 032 U. PAWAR et al., see section 8

- 106 100 B. PRATHAP\*, G. NAGARAJAN, C. ROOSEWELT, V. GOPAL (\*Faculty of Pharmacy, PRIST Univeristy, Thanjavur 614904, Tamil Nadu, India): Simultaneous estimation of gatifloxacin and ambroxol HCl in tablet formulation by HPTLC method. Scholars Research Library 2(3), 163-167 (2010). TLC on silica gel with *n*-butanol - water - methanol - ammonia 8:1:1:2 with chamber saturation for 10 min. The  $hR_f$ -value was 33 for gatifloxacin and 89 ambroxol HCl. The method was linear in the range of 100-600 ng/band for gatifloxacin and 12.5-62.5 ng/band for ambroxol HCl. The recovery was 99.3-100.3 %. Densitometric evaluation at 310 nm.

pharmaceutical research, quality control, densitometry, quantitative analysis 17a

- 106 101 A. RAMADAN\*, A. BODAKJI, I. MAHMOUD (\*Dept. of Chemistry, Faculty of science, University of Aleppo, Syria, dramadan@scs-net.org): TLC-densitometric determination of vitamins B1, B6 and B12 in puree and pharmaceutical formulations using treated aleppo bentonite. Asian Journal of Chemistry 22(4), 3283-3291 (2010). TLC of vitamins B1, B6, and B12 in formulations on bentonite-coated TLC plates (0.3 mm) with iso-butylalcohol - methanol - chloroform - acetic acid - ammonia 75:10:35:4:5. The  $hR_f$ -value of vitamin B1 was 26, of B6 37 and of B12 68. The method was linear in the range of 500-4000 ng/band for all three vitamins. Densitometric quantification at 275 nm for B1, B6, and B12 and at 525 nm for B12.

pharmaceutical research, quality control, densitometry, quantitative analysis 17a

- 106 102 Madhuri RAMTEKE\*, A. KASTURE, N. DIGHADE (\*V.R.M. Institute of Diploma in Medical Lab. Technology, R-2, Electronic Zone, MIDC, Hingna Rd., Nagpur 441110, India, shende\_madhuri@rediffmail.com): Development of high-performance thin-layer chromatographic method for simultaneous estimation of atenolol and nifedipine in combined dosage form. Asian Journal of Chemistry 22(8), 5951-5955 (2010). TLC on silica gel with cyclohexane - methanol - ethyl acetate - 25 % ammonia 10:3:6:1. The  $hR_f$  value was 7 for atenolol and 69 for nifedipine. Densitometric evaluation at 230 nm. The linearity was in the range of 3-21  $\mu\text{g}/\text{band}$  for atenolol and 1.2-8.4  $\mu\text{g}/\text{band}$  for nefidipine. The recovery was 100.7 % for both drugs. The method was found suitable for analysis of the combined dosage form of atenolol and nefidipine without interference of excipients.

pharmaceutical research, quality control, densitometry, quantitative analysis 17c

- 106 103 N.M. RAO\*, K.R. KUMAR, J. BHAGYALAKSHMI, T.K. RAVI (\*Dept. of Pharmaceutical Analysis, College of Pharmacy Institute of Paramedical Sciences, Coimbatore TN, India, mallim-pharmmba@gmail.com): Development and validation of a stability indicating TLC method for the estimation of voglibose in bulk and tablet dosage forms. International Journal of Pharma World Research 1(2) (2010). A validated stability indicating assay method is described for estimation of voglibose in bulk and dosage formulation. TLC on pre-washed silica gel plates with acetonitrile - methanol - 25 % ammonia 150:40:1. The  $hR_f$  value of voglibose was 66. Densitometric evaluation at 284 nm. The method was linear in the range of 100-450 ng/band. The average recovery was 101.4 %.

pharmaceutical research, quality control, densitometry, quantitative analysis 17a

- 106 104 W. SOLOMON\*, P. ANAND, R. SHUKLA, R. SIVAKUMAR, R. VENKATNARAYANAN (\*Dept. of Pharmaceutical Analysis, RVS College of Pharmaceutical Sciences, Suler, Coimbatore 641402, Tamil Nadu, India, samwd\_2000@yahoo.com): Application of TLC-densitometric method for simultaneous estimation of tramadol HCl and paracetamol in pharmaceutical dosage forms. International Journal of ChemTech Research 2(2), 1188-1193 (2010) TLC on silica gel with chloroform - ethanol 7:3. The  $hR_f$  value of tramadol HCl was 48 and of paracetamol 85. Densitometric evaluation at 254 nm. The method was linear over a concentration range of 2.5-32.5  $\mu\text{g}/\text{band}$  for tramadol and 10-50  $\mu\text{g}/\text{band}$  for paracetamol. The recovery was 100.7-101.8 %. There was no interference with excipients from the dosage form.

pharmaceutical research, quality control, densitometry, quantitative analysis 17c

### 18. Amino acids and peptides, chemical structure of proteins

- 106 105 D. DAS\*, A. SAHANA, R. SAHA, P. KUNDU, S. LASKAR (\*Department of Chemistry, The University of Burdwan, Burdwan 713104, India; ddas100in@yahoo.com): Anthracene-anchored derivatized methionine: A new ligand for detection of amino acids, and estimation of binding constants. J. Planar Chromatogr. 23, 255-259 (2010). Synthesis of a new ligand by anchoring anthracene to L-methionine. The ligand allows identification of amino acids on TLC plates based on distinguishable colors. A theoretical calculation (Hartree-Fock) was performed to investigate interaction of the ligand with the amino acids. TLC of 22 amino acids on silica gel with *n*-propanol - water 7:3. Detection by spraying with 0.01 % anthracenylmethyl L-methionine in acetone, followed by drying and spraying with 0.25 % ninhydrin in acetone. After drying and heating at 110 °C for 10 min the zones were evaluated visually. Detection limits were between 1 and 200 ng/zone.

qualitative identification 18a

- 106 106 A. MOHAMMAD\*, N. HAQ (\*Analytical Research Laboratory, Department of Applied Chemistry, Faculty of Engineering and Technology, Aligarh Muslim University, Aligarh 202 002, India; alimohammad08@gmail.com): TLC separation of amino acids with a green mobile phase. *J. Planar Chromatogr.* 23, 260-264 (2010). TLC of 15 amino acids on silica gel and alumina (with or without impregnation) with micellar solutions of cetrimide and cetylpyridinium chloride and aqueous solutions of dextrose with chamber saturation for 10 min. A TLC system comprising of silica gel impregnated with micellar solution of cetrimide (5.0 mM) as stationary phase and 40 % aqueous solution of dextrose as mobile phase was best suitable for the separation of amino acids. Detection by spraying with 0.3 % ninhydrin solution in acetone and heating for 15-20 min at 60 °C.
- qualitative identification 18a

- 106 107 J.D. VASTA, B. FRIED\*, J. SHERMA (\*Department of Biology, Lafayette College, Easton, PA, 18042, USA; fried@lafayette.edu): Determination of estivation-induced changes in the amino acid content of *Biomphalaria glabrata* snails by high-performance thin-layer chromatography-densitometry. *J. Liq. Chromatogr. Relat. Technol.* 33, 1028-1037 (2010). HPTLC of amino acids (e. g. alanine, arginine, glycine, leucine/isoleucine, lysine, serine, and valine) on silica gel or cellulose pre-washed with dichloromethane - methanol 1:1 using either 2-butanol - pyridine - glacial acetic acid - water 39:34:10:26 or 2-butanol - pyridine - 25 % ammonia - water 39:34:10:26 in a saturated twin-trough chamber. Detection by treatment with ninhydrin reagent (0.3 g ninhydrin in 100 mL of *n*-butanol with 3 mL of glacial acetic acid) and heating at 110 °C for 10 min. Quantitative determination by densitometric absorbance measurement at 610 nm.
- densitometry, quantitative analysis, HPTLC 18a

## 21. Purines, pyrimidines, nucleic acids and their constituents

- 106 108 Claudia CIMPOIU\*, A. HOSU, L. SESERMAN, M. SANDRU, V. MICLAUS (\*Babes-Bolyai University, Faculty of Chemistry and Chemical Engineering, 11 Arany Janos, 400028 Cluj-Napoca, Romania, ccimpoi@chem.ubbcluj.ro): Simultaneous determination of methylxanthines in different types of tea by a newly developed and validated TLC method. *J. Sep. Sci.* 33, 3794-3799 (2010). HPTLC of caffeine (1), theobromine (2) and theophylline (3) in different types of tea on silica gel with chloroform - dichloromethane - isopropanol 4:2:1. Quantitative determination by absorbance measurement at 254 nm. The  $hR_f$  of (1), (2) and (3) was 65, 45 and 56, respectively. Limits of detection and quantification were 22 and 45 ng for (1), 23 and 46 ng for (2) and 22 and 43 ng for (3), respectively. The intra-day and inter-day precisions had a %RSD lower than 2.55 % ( $n = 6$ ) for all substances. Recoveries (by standard addition) were between 95.1-101.5 % for all the three methylxanthine derivatives. The values of LOD and LOQ obtained are similar with those obtained by HPLC.
- food analysis, quality control, HPTLC, quantitative analysis, comparison of methods, densitometry 21a

- 106 109 CH. JENDRESEN\*, M. KILSTRUP, J. MARTINUSSEN (\*Center for Systems Microbiology, Department of Systems Biology, Technical University of Denmark, 2800 Lyngby, Denmark): A simplified method for rapid quantification of intracellular nucleoside triphosphates by one-dimensional thin-layer chromatography. *Anal. Biochem.* 409 (2), 249-259 (2011). Presentation of a less time-consuming, more sensitive, and more precise method for the quantitative determination of nucleoside triphosphates (NTPs), 5-ribosyl-1-pyrophosphate (PRPP), and inorganic pyrophosphate (PPi) in cell extracts by TLC: Separation of an acid extract of *L. lactis* by charcoal filtration into a filtrate and an eluate, which then was separated by TLC either in the Cashel solvent (0.85 M potassium phosphate, pH 3.4) or in the AFC solvent (3 M ammonium formate [pH 2.4] and 0.7 M ammonium chloride). Two-dimensional separation of 18  $\mu$ L of the eluate sample using the

AFC solvent in the first dimension and using 0.75 M LiCl in 7.5 % lithium borate (pH 6.8, borate solvent) in the second dimension.

food analysis, environmental, agricultural, clinical chemistry research, HPTLC, qualitative identification, quantitative analysis, comparison of methods, radioscanning 21

- 106 110 P. SAINI\*, R. SINGH, S. MATHUR, G. SINGH, S. TUTEJA, U. SINGH (\*Indian Pharmacopoeia Commission Ministry of Health & Family Welfare, Government of India, Sector 23 Rajnagar, Ghaziabad 201002, UP, India): Simultaneous HPTLC method for estimation of lamivudine and zidovudine in bulk and in tablet dosage forms. *Indian Drugs* 47(5), 42-45 (2010). HPTLC of lamivudine and zidovudine on silica gel with acetone - toluene - methanol 2:1:2. The  $hR_f$  of lamivudine was 34 and of zidovudine 74. Densitometric quantification at 267 nm for zidovudine and 272 nm for lamivudine. The method was linear in the range of 10-210 ng/band for lamivudine and 30-420 ng/band for zidovudine. The recovery was between 99.5-100.4 % for the compounds. The method is suitable for estimation of compounds in combined formulations.

pharmaceutical research, HPTLC, densitometry, quantitative analysis 21a

- 106 111 V. VENKATESH\*, A. PRABHAKAR, P. SURESH, U. MAHESHWARI, N. RAO (\*Dept. of Pharmaceutical Analysis, Chalapathi Institute of Pharmaceutical Sciences, Vhalapathi Nagar, Lam, Guntur 522034, India, vedullapalli.pharma@gmail.com): HPTLC method for the simultaneous estimation of etophylline and theophylline in tablet dosage form. *Asian Journal of Chemistry* 23(1), 309-311 (2011). TLC on silica gel with toluene - isopropyl alcohol - acetic acid 12:12:1. The  $hR_f$  values was 63 for etophylline and 75 for theophylline. Densitometric evaluation at 261 nm. The method was linear in the range of 200-400 ng/band for etophylline and 60-80 ng/band for theophylline.

pharmaceutical research, quality control, densitometry, quantitative analysis 21a

## 22. Alkaloids

- 106 112 H. MISRA\*, D. MEHTA, B. MEHTA, M. SONI, D. JAIN (\*School of studies in Chemistry & Biochemistry, Vikram University, Ujjain 456010, India): Study of extraction and HPTLC-UV method for estimation of caffeine in marketed tea (*Camellia sinensis*) granules. *International Journal of Green Pharmacy* 3 (1), 47-17 (2009). HPTLC of caffeine on silica gel with ethyl acetate - methanol 9:1. Densitometric quantification at 274 nm. The method was linear in the range of 2-14 µg/band. The sample extracted with 5 % diethyl amine in water gave the maximum yield of caffeine (2.1 %). The proposed chromatography gave the best resolution with caffeine at an  $hR_f$  value of 40. The method can be used for quality control of tea samples in respect of caffeine contents.

quality control, herbal, densitometry, HPTLC, quantitative analysis, qualitative identification 22

- 106 113 A. SUTHAR\*, K. KATKAR, P. PATIL, P. HAMARAPURKAR, G. MRUDULA, V. NAIK, G. MUNDADA, V. CHAUHAN (\*Pharmacognosy Dept., Manipal College of Pharmaceutical Science, Manipal University, Karanataka, India, ashish.suthar@piramal.com): Quantitative estimation of vasicine and vasicinone in *Adhatoda vasica* by HPTLC. *Journal of Pharmacy Research* 2(12), 1893-1899 (2009). HPTLC of vasicine and vasicinone in methanolic aqueous extracts of the dried herb and in cough syrup on silica gel with chloroform - methanol 9:1. The  $hR_f$  value of vasicine was 11 and of vasicinone 45. Densitometric quantification at 280 nm. The method was linear in the range of 2-100 ng/band for vasicine and 25-1000 ng/band for vasicinone. The recovery was 95-102 %. The limit of detection of vasicine was 1 ng and of vasicinone 25 ng. Vasicine and vasicinone were well separated from other constituents of *Adhatoda vasica*.



quality control, herbal, HPTLC densitometry, quantitative analysis

22

### 23. Other substances containing heterocyclic nitrogen

- 106 114 L.S. ABDEL-FATTAH, Z. EL-SHERIF, K.M. KILANI, Dalia A. EL-HADDAD\* (\*National Organization for Drug Control & Research, 6&7 AboHazem St, Pyramids, P. O. Box 29, Giza, Egypt; daliaelhaddad@hotmail.com): HPLC, TLC, and first-derivative spectrophotometry stability-indicating methods for the determination of tropisetron in the presence of its acid degradates. J. AOAC Int. 93, 1180-1191 (2010). TLC of tropisetron and its acid-induced degradation products on silica gel with methanol - glacial acetic acid 22:3 with chamber saturation for 45 min. Detection under UV light at 254 nm. Quantitative determination by scanning at 285 nm. The  $hR_f$  values were 44, 84, and 92 for tropisetron and degradates I and II, respectively. Linearity was between 1 and 10  $\mu\text{g}/\text{zone}$ . Mean accuracy was 100.2 %. The precision was 0.64 %. The limit of detection and quantification for tropisetron was 0.26 and 0.80  $\mu\text{g}/\text{zone}$ , respectively. The intraday and interday precisions were evaluated by assaying freshly prepared samples of tropisetron in triplicate concentrations, i. e. 3, 5, and 9  $\mu\text{g}/\text{zone}$ , resulting in 99.8 %, 100.3 %, and 100.4 %, respectively, for interday precision and 99.7 %, 100.2 %, and 99.7 %, respectively, for intraday precision (with a precision of 0.9 %, 0.6 %, 0.8 % and 0.6 %, 0.4 %, 0.6 %, respectively).

quality control, pharmaceutical research, densitometry, quantitative analysis

23

- 106 115 Mrinalini DAMLE\*, M. DANGI, Darshana CHAUDHARI, M. SINKAR, Veena RACHA (\*AIS-SMS College of Pharmacy, Kennedy Rd., Pune 411001, India): Stability indicating HPTLC method for estimation of nebivolol hydrochloride and amlodipine besylate in combination. Eurasian J. Anal.Chem. 5(1), 21-24 (2010). A stability-indicating HPTLC method has been developed for simultaneous estimation of amlodipine besylate and nebivolol hydrochloride. HPTLC on silica gel with ethyl acetate - methanol - 25% ammonia 17:2:2. The  $hR_f$  value of amlodipine besylate was 40 and of nebivolol hydrochloride 60. Densitometric quantification at 240 nm for amlodipine and 260 nm for nebivolol. The method was linear in the range of 500-2000 ng/band for both compounds. Degradation products formed under stress conditions (acid, alkali, oxidative, dry heat, photo degradation) were well separated from the main compound. The method was suitable for quality control and stability studies.

pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC

23e

- 106 116 B. DHANDAPANI\*, N. THIRUMOORTHY, P. JOSH (\*Dept. of Pharmaceutical Analysis, A. M. Reddy Memorial College of Pharmacy, Petturivaripalem, Narasaraopet, Guntur 522601, A.P., India, dhandapani@gmail.com): Development and validation for the simultaneous quantification of nebivolol hydrochloride and hydrochlorothiazide by UV spectroscopy, RP-HPLC and HPTLC in tablets. E-Journal of Chemistry 7(2), 341-348 (2010). HPTLC of nebivolol hydrochloride and hydrochlorothiazide on silica gel with ethyl acetate - methanol - 25 % ammonia 17:2:1 with chamber saturation. The  $hR_f$  value of hydrochlorothiazide was 21 and of nebivolol 41. Densitometric evaluation at 285 nm. The method was linear in the range of 200-1000 ng/band for nebivolol and 500-2500 ng/band for hydrochlorothiazide. The recovery was 98.9-102.4 %. Comparison of the HPTLC method with a RP-HPLC method and a UV spectroscopic method gave comparable results. The HPTLC method is suitable for routine quality control.

pharmaceutical research, quality control, HPTLC, densitometry, comparison of methods, quantitative analysis

23e

- 106 117 H. KALÁSZ\*, E. MINGSOVICS, N. RAM, K. KUCA (\*Department of Pharmacology and Pharmacotherapy, Faculty of Medicine, Semmelweis University, 1089 Budapest, Nagyvárad tér 4, Hungary; drkalasz@gmail.com): Thin-layer chromatography of pyridinium aldoximes using dis-

tinct techniques for development. *J. Liq. Chromatogr. Relat. Technol.* 33, 922-935 (2010). TLC and HPTLC of 22 different pyridinium aldoximes on silica gel with water - acetonitrile - acetic acid 8:1:1, water - acetone - acetic acid 8:1:1, and acetone - 50 mM aqueous sodium acetate 1:4 using unsaturated and saturated vapor phase as well as OPLC. Detection under UV 254 nm.

toxicology, clinical chemistry research, HPTLC, qualitative identification,  
comparison of methods 23d

- 106 118 W. PARYS, Alina PYKA\* (\*Department of Analytical Chemistry, Faculty of Pharmacy, Medical University of Silesia, 4 Jagiellonska Street, 41-200 Sosnowiec, Poland; apyka@sum.edu.pl): Use of TLC and densitometry to evaluate the chemical stability of nicotinic acid and its esters on silica gel. *J. Liq. Chromatogr. Relat. Technol.* 33, 1038-1046 (2010). TLC of nicotinic acid and methyl, ethyl, isopropyl, butyl, hexyl, and benzyl nicotinate (heated for 1 to 7 h at 120 °C) on silica gel pre-washed with methanol using methanol - benzene (for nicotinic acid) and acetone - hexane 2:3 for nicotinic acid esters. Quantitative determination by densitometric absorbance measurement at the respective absorption maximums (263 nm for nicotinic acid, 220-222 nm for nicotinic acid esters). The  $hR_f$  of nicotinic acid was 44, and of methyl, ethyl, isopropyl, butyl, hexyl, and benzyl nicotinate 48, 46, 51, 54, 56, and 44, respectively.)

pharmaceutical research, quality control, densitometry, quantitative analysis 23d

- 106 119 L. POBLOCKA, Mirosława KRAUZE\*, D. GŁOD, A. KAWIAK, E. LOJKOWSKA (\*Department of Pharmacognosy, Medical University of Gdansk, Gen. J. Hallera 107 Street, Gdansk, Poland, krauze@amg.gda.pl): Chromatographic analysis of simple phenols in some species from the genus *Salix*. *Phytochem. Anal.* 21, 463-469 (2010). HPTLC of pyrocatechol in the barks of different species and clones from the genus *Salix* on diol phase by multiple gradient development with chloroform - hexane 7:3 - ethyl acetate - formic acid in the following gradients: (I) 18:2:0.01, (II) 17:3, and (III) 16:4. Detection by spraying with thymol reagent. Quantitative determination by absorbance measurement at 254 nm. The  $hR_f$  of pyrocatechol was 25. Detection and quantification limits were 30 and 100 µg/mL, respectively. The intra-day and inter-day precisions had a %RSD lower than 2.5 %. Recovery (by standard addition) was 96.3 %. The correlation coefficient of pyrocatechol concentrations determined by HPTLC and HPLC was 0.9932.

herbal, toxicology, HPTLC,  
quantitative analysis, densitometry 23

- 106 120 A. SHIKHEDKAR\*, S. SURANA (\*R. C. College of Pharmacy Dept. of Pharma. Chemistry, Shirpur, Dhule (MS), India): Application of stability-indicating RP-TLC densitometric determination of rabeprazole sodium in bulk pharmaceutical formulations. *Eurasian J. Anal. Chem.* 4(1), 87-97 (2009). HPTLC of rabeprazole sodium on silica gel with acetone - water 7:30. The  $hR_f$  value was 45. Densitometric quantification at 284 nm. Linearity was in the range of 400-2400 ng/band. The recovery was 99.6-101.4 %. The sample was subjected to forced degradation (acid, alkali, oxidative, thermal, photolytic). The degradation products were well separated from the main compound and the method is suitable for stability studies.

pharmaceutical research, quality control, quantitative analysis, densitometry, HPTLC 23e

- 106 121 Shradhanjali SINGH\*, Kirti TOPAGI, Mrinalini DAMLE (\*Dep of Pharmaceutical Chem, AIS-SMS Collage of Pharmacy, Pune 411001, India, mcdamle@rediffmail.com): A validated high-performance thin-layer chromatographic method for simultaneous estimation of nebivolol hydrochloride and valsartan in pharmaceutical dosage form. *Research Journal Pharm and Tech.* 2(4), 746-748 (2009). A validated HPTLC method is reported for simultaneous estimation of nebivolol

hydrochloride and valsartan in combined dosage form. HPTLC on silica gel with ethyl acetate - methanol - 25 % ammonia. The  $hR_f$  value of valsartan was 27 and of nebivolol 75. The method was linear in the range of 800-2400 ng/band for nebivolol and 200-1000 ng/band for valsartan. Densitometric quantification at 280 nm. The method was found suitable for routine quality control of the combined dosage form.

pharmaceutical research, quality control, densitometry, HPTLC, quantitative analysis 23e

106 122 L. SIVASUBRAMANIAN\* DEVARAJAN, K. BELL (\*Dept. of Pharmaceutical Analysis, SRM College of Pharmacy, Kattankulathur, Tamil Nadu, India): HPTLC for the simultaneous determination of tizanidine and valdecoxib in pharmaceutical dosage form. Journal of Pharmacy Research 2(1), 189-195 (2009). A validated HPTLC method has been developed for simultaneous estimation of valdecoxib and tizanidine in dosage form. HPTLC on silica gel with toluene - methanol - ethyl acetate 2:2:1. The compounds were well separated as compact bands. The method was linear in the range of 10-100 ng/band for tizanidine and 100-1000 ng/band for valdecoxib. The recovery was in the range of 99.3–101.2 %. Densitometric quantification at 254 nm. The method is suitable for quality control of combined dosage form.

pharmaceutical research, quality control, densitometry, HPTLC, quantitative analysis 23e

106 076 N. VEKARIYA et al., see section 11

106 123 S. WALODE\*, H. CHAUDHARI, M. SARASWAT, A. KASTURE, S. WADODKAR (\*Sinhgad Institute of Pharmaceutical Science, Kusgaon (BK), Lonavala, Pune 410401, India): Validation high performance thin layer chromatographic determination and content uniformity test for rosiglitazone tablets. Indian J. Pharm. Sci. 72(2), 249-252 (2010). HPTLC of rosiglitazone on silica gel with methanol - toluene - chloroform - triethyl amine 2:161:1. Caffeine was used as internal standard. The  $hR_f$  value of rosiglitazone was 31 and of caffeine 52. Densitometric evaluation at 264 nm. The method was linear in the range of 5-50 µg/band with average recovery of 99.8-100.2 %. The method was suitable for content uniformity testing according to USP and could be used for stability studies as well.

pharmaceutical research, quality control, densitometry, HPTLC, quantitative analysis 23e

## 24. Organic sulfur compounds

106 124 M. ARANDA, Gertrud E. MORLOCK\* (\*Institute of Food Chemistry, University of Hohenheim, Garbenstrasse 28, 70599 Stuttgart, Germany; gmorlock@uni-hohenheim.de): Quantification of pyritinol in solid pharmaceutical formulation by high-performance thin-layer chromatography-ultraviolet detection and selective evaluation by mass spectrometry. J. Liq. Chromatogr. Relat. Technol. 33, 957-971 (2010). HPTLC of pyritinol on silica gel with dichloromethane - methanol - formic acid 9:1:1 in a twin trough chamber. Quantitative determination by absorbance measurement at 300 nm. Repeatability and intermediate precision in matrix were 0.4 % and 3.0 %, respectively. Recoveries of spiked samples at three levels ranged from 98.5 to 101.9 % with intermediate precisions of *RSD* 3.7 to 4.7 %. The limit of detection and quantification was 0.6 and 2.0 µg/mL (6 and 20 ng/band), respectively. Selectivity was evaluated determining the peak purity by UV-spectrophotometry and showed correlation coefficients ( $r$ ) > 0.9997. Additionally the selectivity was proven by mass spectrometry. Mass spectra showed only the analyte signals which indicated a highly satisfying selectivity.

HPTLC, quantitative analysis, densitometry

24

## 26. Organometallic and related compounds

- 106 125 R. BAOSIC\*, A. RADOJEVIC, Z. TESIC (\*Faculty of Chemistry, University of Belgrade, Studentski trg 12, P.O. Box 158, 11000 Belgrade, Serbia): Prediction of the retention of beta-diketono complexes in TLC systems on silica gel by quantitative structure-retention relationships. J. Serb. Chem. Soc. 75(4), 513-521 (2010). A TLC method has been reported for prediction of retention behavior of several beta-diketono complexes of cobalt(III), chromium(III), and ruthenium(III). Aim of this study was to develop a model for accurate quantitative relationships between molecular structures and retention behavior of mixed beta-diketono complexes. TLC of 36 complexes on silica gel with four mono- and five two-component mobile phases. Quantitative structure - retention relationships (QSSRS) shall allow the prediction of retention of new solutes and structurally similar compounds, as well as their molecular description.

pharmaceutical research, qualitative identification

26c

## 27. Vitamins and various growth regulators

- 106 126 K. UETA, M. NISHIOKA, Y. YABUTA, F. WATANABE\* (\*The United Graduate School of Agricultural Sciences, Tottori University, Tottori 680-8553, Japan; watanabe@muses.tottori-u.ac.jp): TLC-bioautography analysis of vitamin B12 compound from the short-necked clam (*Ruditapes philippinarum*) extract used as a flavoring. J. Liq. Chromatogr. Relat. Technol. 33, 972-979 (2010). TLC of vitamin B12 on silica gel with 2-propanol - 28 % ammonia - water 7:1:2 and 1-butanol - 2-propanol - water 10:7:10 in the dark at room temperature. After drying, agar containing basal medium and pre-cultured *E. coli* 215 was overlaid and then incubated at 30 °C for 20 h. Detection by spraying with a methanolic solution of 2,3,5-triphenyltetrazolium salt, B12 compounds appeared as red zones indicating *E. coli* growth.

food analysis, agricultural, qualitative identification

27

## 28. Antibiotics, Mycotoxins

- 106 127 A. AHMAD, M. MUJEEB, B. P. PANDA\* (\*Pharmaceutical Biotechnology Laboratory, Faculty of Pharmacy, Jamia Hamdard (Hamdard University), Hamdard Nagar, New Delhi, India 110062; bibhu\_panda31@rediffmail.com): An HPTLC method for the simultaneous analysis of compactin and citrinin in *Penicillium citrinum* fermentation broth. J. Planar Chromatogr. 23, 282-285 (2010). HPTLC of compactin and citrinin on silica gel with toluene - ethyl acetate - formic acid 3:2:1 in a twin-trough chamber saturated for 30 min at room temperature and a relative humidity of 60 +/- 5 %. Quantitative determination by absorbance measurement at 238 nm and 366 nm. The  $hR_f$  of compactin and citrinin was 47 and 62, respectively. Good correlation was obtained between peak area and concentration, with a determination coefficient  $r^2 = 0.998$  for compactin and 0.996 for citrinin.

pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis, mycotoxins

28b

- 106 128 Monika DABROWSKA\*, J. KRZEK (\*Jagiellonian University, Collegium Medicum, Department of Inorganic and Analytical Chemistry, Medyczna 9, Kraków, Poland; mtylka@cm-uj.krakow.pl) : Separation, identification, and quantitative analysis of the epimers of cefaclor by TLC-densitometry. J. Planar Chromatogr. 23, 265-269 (2010). TLC of the epimers of cefaclor on silica gel (impregnated with beta-cyclodextrin by development with 1:9 aqueous beta-cyclodextrin solution - methanol) with chloroform - ethyl acetate - glacial acetic acid - water 4:4:4:1 with chamber saturation. Chromatograms were developed at 5 °C and dried at room temperature. Quantitative determination by absorbance measurement at 274 nm. The limit of detection was 0.24 and 0.27 µg/band for the epimers at  $hR_f$  26 and 33, respectively. The limit of quantification was 0.74 and 0.83 µg/band, respectively. The recovery was between 100.0 and 100.8 % and the precision

between 0.7 and 1.7 %.

quality control, pharmaceutical research, densitometry

28a

- 106 129 M. DHOKA\*, V. GAWANDE, P. JOSHI (\*Dept. of Q.A. AISSMS College of Pharmacy, Kennedy Rd., near R.T.O. Pune 411001, M.S., India): HPTLC determination of amoxicillin trihydrate and bromhexine hydrochloride in oral solid dosage forms. *J. Pharma. Sci. & Res.* 2(8), 477-483 (2010). TLC on silica gel with ethyl acetate - glacial acetic acid - methanol - water 10:5:3:2. The  $hR_f$  value was 51 and 74 for amoxicillin and bromhexine, respectively. Densitometric evaluation at 260 nm (bromhexine) and at 320 nm (amoxicillin). The method was linear in the range of 200-1000 ng/band and 10-30  $\mu\text{g}/\text{band}$  for bromhexine and amoxicillin, respectively. The recovery was between 98.1-101.5 %.

pharmaceutical research, quality control, densitometry, quantitative analysis

28a

- 106 130 Mária HEVESI\*, A. M. MÓRICZ, Z. KIRÁLY-VÉGHÉLY, M. TÓTH, G. KÁTAY, E. TYIHÁK (\*Corvinus University of Budapest, Faculty of Horticultural Sciences, Department of Pomology, Villányi Str. 35-44, 1118 Budapest, Hungary; maria.hevesi@uni-corvinus.hu): Effect of trans-resveratrol and ascorbigens on the fire blight pathogen *Erwinia amylovora* in the BioArena system. *J. Planar Chromatogr.* 23, 411-414 (2010). OPLC of ascorbigen and 1'-methylascorbigen on silica gel with *n*-hexane for the first step and chloroform - methanol 9:1 for the second step; OPLC of trans-resveratrol on silica gel with chloroform - methanol 10:1. Detection by bioautography (immersion in *E. amylovora* cell suspension, incubation for 2 h at 26 °C and 100 % relative humidity, and staining with a solution of 80 mg MTT and 100 mg Triton X-100 in 100 mL water. Quantitative determination by densitometry.

agricultural, quality control, HPTLC, densitometry

28

- 106 011 M. KAMINSKA et al., see section 2

## 29. Pesticides and other agrochemicals

- 106 131 R. AKKAD, W. SCHWACK\* (\*Institute of Food Chemistry, University of Hohenheim, Garbenstrasse 28, 70599 Stuttgart, Germany): Multi-enzyme inhibition assay for the detection of insecticidal organophosphates and carbamates by high-performance thin-layer chromatography applied to determine enzyme inhibition factors and residues in juice and water samples. *J. Chromatogr. B* 878 (17-18), 1337-1345 (2010). Use of rabbit liver esterase, *Bacillus subtilis* esterase, and cutinase from *Fusarium solani* pisi for the detection of 21 organophosphorus and carbamate pesticides by HPTLC-enzyme inhibition assays (HPTLC-EI) on silica gel with *n*-hexane - ethyl acetate - dichloromethane 13:4:3. HPTLC-EI assay of three groups of organophosphate and carbamate insecticides with 1) *n*-hexane - ethyl acetate 63:37, 2) chloroform - ethyl acetate 9:1, 3) *n*-hexane - acetone - dichloromethane 15:2:3. Detection by treatment with Fast Blue Salt B and enzymatically coupling to alpha-naphthol released from the respective acetate used as substrate. Quantification by densitometry at 533 nm. Calculation of enzyme inhibition factors derived from HPTLC-EI using linear calibration curves. Comparison to published inhibition constants showed good correlation. The limits of detection ranged from a few pg/zone for organophosphates as strongest inhibitors to a few ng/zone for most carbamates and was around 60 ng/zone for chlorpyrifos and 14 ng/zone for parathion without oxidation. The CUT was able to detect insecticides of high and low inhibitory power in the range of ng to  $\mu\text{g}/\text{zone}$ . The HPTLC-EI with rabbit liver esterase was applied to the analysis of apple juice and drinking water samples spiked with paraoxon (0.001 mg/L), parathion (0.05 mg/L) and chlorpyrifos (0.5 mg/L). The mean recovery was 71-112 % with standard deviations of 2.0-18.3 %.

food analysis, environmental, agricultural, HPTLC, densitometry,  
qualitative identification, quantitative analysis

29c

- 106 132 Vera BAUMGARTNER\*, W. SCHWACK (\*State Laboratory of the Canton Basel-City, Kannenfeldstrasse 2, 4056 Basel, Switzerland; vera.baumgartner@bs.ch): Enhanced quantitative evaluation of the HPTLC-bioluminescence detection. *J. Liq. Chromatogr. Relat. Technol.* 33, 980-995 (2010). HPTLC of bronopol and Kathon CG on silica gel (pre-washed with methanol) with methanol - dichloromethane - *n*-hexane 2:12:7. For detection, luminescent *Vibrio fischeri* bacteria were cultivated. Plates were dried for 30-40 min at 120 °C on a plate heater and then immersed for 1 s in the bacteria suspension using an immersion device and a glass dip tank. Excess bacteria suspension was removed. Images were taken using the BioLuminizer with an exposure time of 55 s. The effective and reasonably fast quantitative method is in routine use at several laboratories.

quantitative analysis, HPTLC

29f

- 106 133 M. BROSZAT, H. ERNST, B. SPANGENBERG\* (\*Institute of Process Engineering, University of Offenburg, Badstrasse 24, 77652 Offenburg, Germany; Spangenberg@FH-Offenburg.de): A simple method for quantifying triazine herbicides using thin-layer chromatography and a CCD camera. *J. Liq. Chromatogr. Relat. Technol.* 33, 948-956 (2010). HPTLC of triazine herbicides (atrazin, terbumelon, simazine, and terbutylazine) on silica gel with methyl-*t*-butyl ether - cyclohexane 1:1 in a vertical developing chamber without chamber saturation. Detection by derivatization using chlorine and starch-iodine. Quantitation with a CCD camera; the range of linearity covers two magnitudes (for atrazine from 10 ng to 1000 ng).

agricultural, environmental, densitometry, quantitative analysis, HPTLC

29d

### 30. Synthetic and natural dyes

- 106 001 Marta KUCHARSKA et al., see section 1

### 32. Pharmaceutical and biomedical applications

- 106 134 R. BAJWA\*, R. SANE (\*Guru Nanak Institute for Research & Development G. N. Khalsa College, Nathalal Parekh Rd., Matunga, Mumbai 40019, India): Studies in phytochemical investigation, anti bacterial activity and acute toxicity study of *Prosopis cineraria* pods. *Indian Drugs* 47(6), 31-38 (2010). Pods of *Prosopis* were extracted with different solvents and the extracts were subjected to phytochemical evaluation using 2D paper chromatography and HPTLC. Analysis of phenolic acids by two-directional paper chromatography with toluene - acetic acid - water 6:7:3 in the first direction and sodium formate - formic acid - water 10:1:200 in the second direction. For detection of phytoconstituents HPTLC on silica gel with different mobile phases and detection wavelengths: 1) beta-sitosterol with toluene - methanol 9:1, measured at 258 nm, 2) coumaric acid with toluene - ethanol 4:1, measured at 292 nm, 3) quercetin with toluene - methanol 4:1, measured at 271 nm, and 4) caffeine with toluene - acetone 7:3, measured at 272 nm.

pharmaceutical research, herbal, qualitative identification, HPTLC

32e

- 106 135 Z. CHUNFANG\*, X. YIN, Y. LONGJIANG, L. SHUO, W. ZEQIANG (\*Resource of Biology and Biotechnical Lab, College of Life Science and Technology, Huazhong University of Science and Technology, Wuhan City, Hubei Province, 430074 China): TLC and HPLC methods to follow the synthesis of vinorelbine. *J. Chromatogr. Sci.* 48(8), 685-689 (2010). TLC combined with HPLC for detection of the intermediates and the end-product from the synthesis process of vinorelbine. Specific TLC separation of vinblastine sulfate, anhydrovinblastine, and vinorelbine on silica gel with petroleum ether - chloroform - acetone - diethyl amine 47:24:4:5. Quantitative evaluation with HPLC.

quality control, comparison of methods, quantitative analysis, qualitative identification 32a



- 106 136 L. CIESLA, D. STASZEK, M. HAJNOS, Teresa KOWALSKA, Monika HAJNOS\* (\*Department of Inorganic Chemistry, Medical University of Lublin, 6 Staszika Street, Lublin, Poland, monika.hajnos@am.lublin.pl): Development of chromatographic and free radical scavenging activity fingerprints by thin layer chromatography for selected *Salvia* species. *Phytochem. Anal.* 22, 59-65 (2011). TLC and free radical scavenging fingerprints of nineteen *Salvia* species on silica gel with toluene - ethyl acetate - formic acid 60:40:1 for the less polar constituents and ethyl acetate - water - formic acid - acetic acid 100:26:11:11 for the medium and highly polar substances. After drying at room temperature for 15 min derivatization with vanillin sulfuric acid reagent (1 g vanillin with 20 % sulfuric acid in methanol) followed by heating for 5 min at 105 °C. Quantitative determination by absorbance measurement at 254 nm. Free radical scavenging properties were investigated by spraying the plate with DPPH radical reagent (0.2 %) in methanol and left at ambient temperature for 30 min. The strongest free radical scavenging activity was observed for rosmarinic acid, with an  $hR_f$  of 70.

herbal, quantitative analysis, postchromatographic derivatization,  
qualitative identification

32e

- 106 137 M. DAMLE\*, S. PHADTARE, H. RASKAR, K. GADGE, R. MEHENDRE, K. BOTHARA (\*Dept. of Pharmaceutical Chemistry, AISSMS College of Pharmacy, Kennedy Rd., Near R.T.O. Pune 411001, M.S., India, mrunal.damle@rediffmail.com): A validated HPTLC method for determination of arbidol from pharmaceutical formulation. *International Journal of ChemTech Research* 2(2), 1042-1046 (2010) HPTLC on silica gel with dichloromethane - methanol 9:1 in a twin-trough chamber saturated for 30 min. The  $hR_f$  value was 70. Densitometric evaluation in absorbance mode at 254 nm. The method was linear in the range of 400-2000 ng/band. The average recovery was 101.2 %.

pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis 32a

- 106 138 B. DHANDAPANI\*, A. SOMASUNDARAM, S. RASEED, M. RAJA & K. DHANABAL (\*Dept. of Pharmaceutical Analysis, K.M.C.H. College of Pharmacy, Kovai Medical Centre Research & Educational Trust, Kovai Estate, Kalapatti Rd., Coimbatore-641045, India, dhandapanirx@gmail.com): Development and validation of HPTLC method for estimation of quetiapine in bulk drug and in tablet dosage form. *International Journal of ChemTech Research* 1(2), 139-141 (2009). TLC on silica gel with toluene - methanol 3:4. The  $hR_f$  value was 41. Densitometric evaluation at 235 nm. The method was linear in the range of 100-500 ng/band. The average recovery was 98.9 %. The proposed method was suitable for estimation of quetiapine in bulk drug and dosage form.

pharmaceutical research, quality control, densitometry, quantitative analysis 32e

- 106 139 B. DHANDAPANI\*, N. ANJANEYULU, Y. VENKATESHWARLU & H. SHAIK (\*K.M.C.H. College of Pharmacy, Kovai Estate, Lalappatti Rd., Coimbatore 641035, Tamil Nadu, Dept. of Pharmaceutical Analysis, Donbosco College of Pharmacy, Guntur, AP, India, dhandapanirx@gmail.com): HPTLC method development and validation for the simultaneous estimation of amlodipine besylate and nebivolol hydrochloride in tablet dosage form. *Journal of Pharmacy Research* 3(2), 332-334 (2010). HPTLC on silica gel with methylene chloride - methanol - ammonia 17:2:1. The  $hR_f$  value of amlodipine was 19 and of nebivolol 41. Densitometric evaluation at 285 nm. The method was linear in the range of 100-500 ng/band for both compounds. The average recovery was 99.9-102.1 for both compounds and there was no interference with additives present in tablet dosage forms.

pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis 32c

- 106 140 P. GAIKWAD, S. BHOPE\*, V. KUBER, M. PATIL (\*Tulip Laboratories Pvt. Ltd., MIDC Ranjangaon, Pune 412220, India, bshrinivas16@gmail.com): Validated TLC method for simultaneous quantitation of kutkoside and picroside-I from kutki extract. *Phytochem. Anal.* 22, 36-41 (2011). TLC of kutkoside (1) and picroside-I (2) in the kutki extract (*Picrorhiza kurroa*) on silica gel with ethyl acetate - methanol - glacial acetic acid - formic acid 25:5:1:1. Quantitative determination by absorbance measurement at 265 nm. The  $hR_f$  of (1) was 42 and of (2) 61. The precision was 0.77 % and 1.01 % for (1) and (2), respectively. Linearity was between 80-480 ng/zone for both substances. Detection and quantification limits were 24 and 79 ng/zone for both. The intra-day and inter-day precisions were 0.4 % and 0.3 % ( $n = 3$ ) respectively. The recovery for (1) was 96.5 % and for (2) 96.0 %, respectively. The results were comparable with those obtained by HPLC.
- herbal, HPTLC, quantitative analysis, comparison of methods, densitometry 32e
- 106 141 S. GANDHI\*, P. DESHPANDE, U. PATIL, P. MUNDHE, P. R. DESHMUKH (\*Dept. of Pharmaceutical Analysis, AISSMS College of Pharmacy, Kennedy Rd., Pune, India): HPTLC determination of cefpodoxime proxetil and potassium clavulanate in combined tablet dosage form. *The Pharma Review* 149-150 (2010). TLC on silica gel with chloroform - methanol - toluene 4:3. The  $hR_f$  value was 16 and 66 for potassium clavulanate and cefpodoxime proxetil respectively. Densitometric analysis was carried out at 215 nm. The method was linear in the range of 500-2500 and 2000-10000 ng/band for cefpodoxime proxetil and potassium clavulanate respectively. The recoveries are in the range of 99.1-100.7 %.
- pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 32a
- 106 142 V. GANGADEVI \*, J. MUTHUMARY (\*Centre for Advanced Studies in Botany, University of Madras, Guindy Campus, Chennai 600 025, Tamil Nadu, India): A simple and rapid method for the determination of taxol produced by fungal endophytes from medicinal plants using high-performance thin-layer chromatography. *Chinese J. Chromatogr.* 26 (1), 50-55 (2008). TLC of some endophytic fungi isolated by column chromatography from selected medicinal plants on silica gel plates with 1) chloroform - methanol 7:1, 2) chloroform - acetonitrile 7:3, 3) ethyl acetate - 2-propanol 19:1, 4) methylene chloride - tetrahydrofuran 3:1, 5) methylene chloride - methanol - dimethylformamide 90:9:1. Detection by spraying with 1 % vanillin sulfuric acid reagent after gentle heating. Also HPTLC on silica gel with chloroform - methanol 9:1. Detection by densitometry at 254 nm and 366 nm. If taxol was present, derivatization by spraying with 1 % vanillin sulfuric acid reagent and heating for 2 min, then detection under UV 366 nm and white light. Only 13 fungal species produced taxol in the artificial culture medium of the 20 screened fungi.
- pharmaceutical research
- traditional medicine, quality control, herbal, densitometry, quantitative analysis, qualitative identification, HPTLC 32
- 106 143 A. GANGWAL\*, S. PARMAL, N. SHETH (\*Dept. of Pharmaceutical Sciences, Suarashtra University, Rajkot 360005, India): Phytochemical investigation and immunomodulatory activity of *Lagenaria siceraria* fruits. *Journal of Natural Remedies* 10(2), 170-174 (2010). The powdered plant material was defatted, extracted with methanol, concentrated and successively extracted with ethyl acetate and *n*-butanol. The resulting fractions were concentrated and subjected to chromatographic fingerprint analysis of the phytoconstituents, i.e. alkaloids, saponins, tannins, flavonoids, anthraquinones and sterols. TLC on silica gel with *n*-butanol - acetic acid - water 4:1:5 for the ethyl acetate fraction and with chloroform - methanol 9:1 for the *n*-butanol fraction. Densitometric evaluation at 254 nm and 366 nm for the *n*-butanol fraction and at 290 nm for the ethyl acetate fraction. Derivatization with vanillin-sulfuric acid reagent for the *n*-butanol fraction and

with  $\text{AlCl}_3$  reagent for the ethyl acetate-fraction, evaluation at 600 nm. The *n*-butanol fraction contained sterols and saponins, the ethyl acetate fraction flavonoids.

quality control, herbal, densitometry, qualitative identification 32e

- 106 144 A. HALKA, P.W. PLOCHARZ, A. TORBICZ, T.H. DZIDO\* (\*Department of Physical Chemistry, Medical University of Lublin, Staszica 6, 20-081 Lublin, Poland; tadeusz.dzido@um-lub.pl): Reversed-phase pressurized planar electrochromatography and planar chromatography of acetylsalicylic acid, caffeine, and acetaminophen. *J. Planar Chromatogr.* 23, 420-425 (2010). TLC and pressurized planar electrochromatography (PPEC) of acetylsalicylic acid, caffeine, and acetaminophen on RP-18 and RP-18 W (prewashed with methanol) using acetonitrile - buffer - water (e. g. 15 % acetonitrile in pH 3.8 buffer) in a horizontal chamber saturated for 15 min. Evaluation by densitometry. The results showed that PPEC of these drugs is characterized by faster separation, better performance, and different separation selectivity. The technique is still under development, however, rather slow because a variety of technical challenges must be overcome.

quality control, comparison of methods 32a

- 106 145 Maha HEGAZY\*, Fadia METWALY, M. ABDELKAWY, NADA ABDELWAHAB (\*Analytical Chemistry Department, Faculty of Pharmacy, Cairo University, Kasr El-Aini Street, 11562 Cairo, Egypt): Validated chromatographic methods for determination of hydrochlorothiazide and spironolactone in pharmaceutical formulation in presence of impurities and degradants. *J. Chromatogr. Sci.* 49, 129-135 (2011). TLC of hydrochlorothiazide (HCT), spironolactone (SPR) and their impurities and degradation products on silica gel with ethyl acetate - chloroform - formic acid - triethyl amine 70:30:1:1. Quantification by densitometry. The different method parameters were optimized for maximum separation. The method was applied for determination of HCT and SPR in commercial tablets. Statistical comparison with the also performed HPLC method showed that there is no significant difference in the performance of the methods.

pharmaceutical research, quality control, comparison of methods, quantitative analysis, qualitative identification, densitometry 32c

- 106 146 K. HULLATTI\*, M. SHARADA (\*Dept. for Pharmacognosy, National College of Pharmacy, Shimoga-577201, DOS in Botany, Univ. of Mysore 570006, India, kkhullatti@gmail.com): Comparative phytochemical investigation of the sources of Ayurvedic drug Patha: a chromatographic fingerprinting analysis. *Ind. J. Pharma. Sci.* 72(1), 39-45 (2010). The Ayurvedic Pharmacopoeia of India recognized roots of the three plants *Cissampelos pareira*, *Cyclea peltata*, and *Stephania japonica* (all Menispermaceae) as source for the marketed drug Patha. HPTLC fingerprint analysis of methanolic extracts of roots of all three plants on silica gel with *n*-butanol - ethyl acetate - formic acid - water 3:5:1:1. Detection under UV 365 nm (crude extract) and 295 nm (total alkaloids). Quantification of the marker berberine by HPLC. The three plants exhibit significantly different physico-chemical features and chromatographic fingerprints. Roots of *C. pareira* contained the highest concentration of berberine, *S. Japonica* contained very low amounts and *C. peltata* no berberine at all.

quality control, herbal, traditional medicine, HPTLC, quantitative analysis, densitometry 32e

- 106 147 Malgorzata JANICKA\*, D. PIETRAS-OZGA (\*Faculty of Chemistry, Department of Physical Chemistry, Maria Curie-Sklodowska University, M. Curie-Sklodowska Sq. 3, 20-031 Lublin, Poland; malgorzata.janicka@poczta.umcs.lublin.pl): Chromatographic evaluation of the lipophilicity of *N*-phenyltrichloroacetamide derivatives using micellar TLC and OPLC. *J. Planar Chromatogr.* 23, 396-399 (2010). TLC of 8 newly synthesized *N*-phenyltrichloroacetamides on RP-18 W with

micellar-organic solutions composed of 0.02, 0.04, or 0.06 M Brij 35 (2-dodecoxyethanol, a non-ionic surfactant) and tetrahydrofuran 4:1 in saturated sandwich chambers. Detection under UV 254 nm. According to the results obtained from the OPLC studies, especially log  $k_m$  values are excellent descriptors of the lipophilicity of the compounds.

pharmaceutical research, HPTLC, qualitative identification, biochemical research 32a

- 106 148 S. KAUSHIK\*, P. SHARMA, A. JAIN, M. SIKARWAR (\*Nagaji Institute of Pharmaceutical Sciences, Gwalior (MP), India): Preliminary phytochemical screening and HPTLC fingerprinting of *Nicotiana tabacum* leaf. *Journal of Pharmacy Research* 3(5), 1144-1145 (2010). The plant material (*Nicotiana tabacum* leaves) was extracted with non-polar and polar solvents. The resulting extracts were subjected to screening for different phyto-constituents (such as terpenes, saponins, flavonoids, alkaloids, and steroids) on silica gel using *n*-hexane - ethyl acetate 5:1 for fingerprint profiling. Detection under UV 254 nm. For derivatization the plate was sprayed with anisaldehyde-sulfuric acid reagent followed by densitometric evaluation at 550 nm. It was observed that fingerprint profiling after derivatization was more suitable than under UV before derivatization. Methanolic extracts of the plant yielded a superior profile.

herbal, HPTLC, qualitative identification, densitometry 32e

- 106 149 A. KHODKE, L. POTALE, Mrinalini DAMLE, K. BOTHARA (Dept. of Q. A. A.I.S.S.M.S College of Pharmacy Kennedy Rd., Near R.T.O. Pune 411001, India, mcdamle@rediffmail.com): A validated stability-indicating HPTLC method for simultaneous estimation of irbesartan and hydrochlorothiazide. *Pharmaceutical Methods* 1(1), 39-43 (2010). TLC on silica gel with acetonitrile - chloroform 5:6. The  $hR_f$  value of irbesartan was 27 and of hydrochlorothiazide 45. Densitometric evaluation at 270 nm. The method was linear in the range of 200-1000 ng/band for irbesartan and 200-600 ng/band for hydrochlorothiazide. The drug was subjected to different stress conditions (acid, base, oxidation, thermal, photolytic). All the degradation products were well resolved from the main drug. The proposed method is stability-indicating and suitable for routine analysis of fixed dose formulation in presence of degradation products.

pharmaceutical research, quality control, densitometry, quantitative analysis, radioscanning 32c

- 106 150 A. KRISHNAVENI\*, S. THAAKUR (\*Dept. of Pharmacognosy, College of Pharmacy, Madurai Medical College, Tamil Nadu, India, akrishmaveni72@rediffmail.com): Quantification of harmaline content in *Passiflora foetida* by HPTLC technique. *Journal of Pharmacy Research* 2(5), 789-791 (2009). TLC of harmaline in methanolic leaf extracts of *Passiflora foetida* on silica gel with chloroform - acetone - diethylamine 5:4:1. The  $hR_f$  value of harmaline was 76. Densitometric quantification at 351 nm. The plant was found to contain 0.75 % w/w of harmaline. The method was linear in the range of 1-10  $\mu\text{g}/\text{band}$ .

herbal, densitometry, quantitative analysis 32e

- 106 151 K. LAKSHMI\*, Lakshmi SIVASUBRAMANIAN, A. PANDEY (\*Dept. of Pharmaceutical Analysis, SRM College of Pharmacy, SRM University, Kattankulathur 603203, Tamil Nadu, India, lakshmiss@hotmail.com): A validated HPTLC method for simultaneous determination of losartan and perindopril in tablet. *Research J. Pharm. and Tech.* 3(2), 559-561 (2010). TLC on silica gel with toluene - acetonitrile - formic acid 50:50:3 with chamber saturation for 30 min. The  $hR_f$  value was 27 for perindopril and 55 for losartan. Densitometric evaluation at 212 nm. The method was linear in the range of 5-30  $\mu\text{g}/\text{band}$  and 1-10  $\mu\text{g}/\text{band}$  for losartan and perindopril respectively. The average recovery was 98.2-99.9 %.

pharmaceutical research, quality control, densitometry, quantitative analysis 32c

- 106 152 M. LI\* (Li Min), J. CHEN (Chen Jianrong), L. LI (Li Li) (\*Ningxia Inst. Cont. of Med. For Animal, Yinchuan, Ningxia 750001, China): (Identification of liquorice and *Rhizoma zingiberis* in Siyi decoction for animal by thin-layer chromatography) (Chinese). *J. Tech. & Sci. of Ningxia Agr. & Forest*, 5, 38-39 (2010). For liquorice TLC on silica gel with ethyl acetate - glacial acetic acid - formic acid - water 15:1:1:2, detection by spraying with 10 % sulfuric acid in ethanol and heating at 105 °C. For *Rhizoma zingiberis* TLC on silica gel with cyclohexane - diethyl ether 1:1, detection by spraying with 10% vanillin in sulfuric acid and heating at 105 °C, followed by evaluation in daylight.  
quality control, pharmaceutical research, traditional medicine, herbal, quantitative analysis, qualitative identification 32c
- 106 153 S. MENNICKENT\*, R. FIERRO, M. VEGA, M. DIEGO, C. GODOY (\*Department of Pharmacy, Faculty of Pharmacy, University of Concepción, P.O. Box 237, Concepción, Chile, smennick@udec.cl): Quantitative determination of fluoxetine in human serum by high-performance thin-layer chromatography. *J. Sep. Sci.* 33, 2206-2210 (2010). HPTLC of fluoxetine on silica gel with toluene - glacial acetic acid 4:5. Derivatization of fluoxetine with 4-dimethylamino-azobenzene-4-sulfonyl chloride. Quantitative determination by absorbance measurement at 272 nm. The  $hR_f$  of fluoxetine was 45. Linearity was between 0.05-0.35 ng/μL. Detection and quantification limits were 230 and 700 ng/μL, respectively. The intra-day and inter-day precisions showed a %RSD lower than 3.9 %. Recovery (by standard addition) was 94.7 %. The method was designed to cover the usual serum concentration level of patients taking the drug.  
clinical routine analysis, HPTLC, quantitative analysis, densitometry 32f
- 106 154 P. MIGAS\*, M. SWITKA (\*Department of Pharmacognosy, Medical University of Gdansk, al. Gen. J. Hallera 107, 80-416 Gdansk, Poland; pmig@amg.gda.pl): TLC with an adsorbent gradient for the analysis of taxol in *Taxus baccata* L. *J. Planar Chromatogr.* 23, 286-288 (2010). TLC of taxol with a stationary-phase gradient whereby parts of different TLC plates were connected by use of a MIGAS device. Separated taxol-containing zones were developed with methanol over 1 cm and thus moved to another plate. The lipophilic substance zone was cut out after separation of the sample on a silica gel plate with n-heptane - ethyl acetate 1:1. Further separation of taxol from accompanying hydrophilic substances was carried out on HPTLC RP-18W with methanol - water 4:1. The taxol-containing fraction was finally separated on silica gel with chloroform - acetone 3:1 in a horizontal chamber at constant temperature (30 +/- 1 °C) and humidity (35 +/- 1 %). Detection under UV 254 and 366 nm. Quantitative determination by densitometry at 220 nm. The precision ( $n = 7$ ) was 0.63 % and the repeatability ( $n = 7$ ) 3.35 %. The limit of detection and quantification was 0.50 and 1.00 μg/zone, respectively; the correlation coefficient from linear regression was >0.98, and the linear calibration range was 1–10 μg/zone.  
herbal, pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 32e
- 106 155 F. MIRZAJANI, A. GHASSEMPOUR, M. JALALI\*, M. HOSSEIN (\*Department of Chemistry, Sharif University of Technology, P.O. Box 11155-9516, Tehran, Iran, jalali@sharif.edu): Optimisation of a microwave-assisted method for extracting withaferin A from *Withania somnifera* Dunal. using central composite design. *Phytochem. Anal.* 21, 544-549 (2010). HPTLC of withaferin A in the aerial part of *Withania somnifera* Dunal. on silica gel with ethyl acetate - toluene - formic acid - 2-propanol 14:4:1:1. Quantitative determination by absorbance measurement at 220 nm. The  $hR_f$  of withaferin A was 29. Detection and quantification limits were 18 and 60 ng/zone respectively. The intra-day and inter-day precisions were 4.24 % and 14.28 %, respectively. Recovery (by standard addition) was 88.2 %. The correlation coefficient of the withaferin A determination was 0.9963.

herbal, HPTLC, quantitative analysis, densitometry 32e

- 106 156 H. MISRA\*, B. DWIVEDI, Darshana MEHTA, B. MEHTA, D. JAIN (\*School of Studies in Chemistry & Biochem, Vikram University, Ujjain 456010, MP, India): Development and validation of high-performance thin-layer chromatographic method for determination of alpha-mangostin in fruit pericarp of mangosteen plant (*Garcinia mangostana* L.) using ultraviolet-visible detection. *Rec. Nat. Prod.* 3/4, 178-186 (2009). An HPTLC method has been reported for quantitative estimation of alpha-mangostin in fruit pericarp of *Garcinia mangostana* (Hypericaceae). HPTLC on silica gel with chloroform - methanol 9:1 in a saturated chamber (10 min). The plate was derivatized with anisaldehyde sulfuric acid reagent. Densitometric quantification at 382 nm. Alpha-mangostin was detected as a compact band with  $hR_f$  value of 46. The method was linear in the range of 1-5  $\mu\text{g}/\text{band}$ . The maximum yield of alpha-mangostin was obtained from the plant by hot extraction with methanol.

quality control, herbal, densitometry, quantitative analysis, postchromatographic derivatization, HPTLC 32e

- 106 157 A. MOHAMMAD\*, S. SHARMA, S. BHAWANI, R. SINGH (\*Analytical Research Lab., Dept. of Applied Chem., Faculty of Eng. & Tech., Aligarh Muslim University Aligarh 202002, India): Identification and separation of *Cannabis sativa*, *Embleia ribes*, *Myristica fragrans* and *Piper longum* from organic extract on silica gel surface with anionic micellar solvent system: application in ayurvedic medicine. *The Open Nutraceuticals Journal* 2, 2-6 (2009) A TLC method using a micellar solution of sodium dodecyl sulfate (SDS) as mobile phase has been developed for identification of four herbals present in *Jatiphaladya*, a powdered herbal formulation containing *Cannabis sativa*, *Myristica fragrans*, *Piper longum*, and *Embleia ribes*. The formulation was extracted with 80 % ethanol. TLC on laboratory made plates coated with silica gel and activated at 100 °C for 60 min, with a 5 % solution of SDS as mobile phase. The resolved spots were identified by spraying with a 2 % solution of vanillin in 5 % methanolic sulfuric acid. Spots corresponding to different herbals were well resolved. Different detection reagents were evaluated, i.e. iodine, vanillin sulfuric acid, and anisaldehyde-sulfuric acid. Vanillin sulfuric acid reagent was found to be the most sensitive. Of the different surfactants used, anionic, cationic and nonionic, SDS was found to be most suitable. The most suitable pH of the mobile phase was pH 4.2-5.7, it provided optimum resolution of zones.

traditional medicine, herbal, herbal, quality control, traditional medicine, postchromatographic derivatization, qualitative identification, postchromatographic derivatization, qualitative identification 32e

- 106 158 D. MUKHERJEE, N. KUMAR, T. KHATUA, P. MUKHERJEE\* (\*School of Natural Products Studies, Department of Pharmaceutical Technology, Jadavpur University, Kolkata 700032, India, pulokm@gmail.com): Rapid validated HPTLC method for estimation of betulinic acid in *Nelumbo nucifera* (Nymphaeaceae) rhizome extract. *Phytochem. Anal.* 21, 556-560 (2010). HPTLC of betulinic acid in the rhizomes of *Nelumbo nucifera* on silica gel with chloroform - methanol - formic acid 49:1:1. Detection by spraying with anisaldehyde - acetic acid - methanol - sulfuric acid 5:100:850:50 and drying at 50 °C for 10 min. Quantitative determination by absorbance measurement at 420 nm. The  $hR_f$  of betulinic acid was 30. Linearity was between 2 and 10  $\mu\text{g}/\text{zone}$ . Detection and quantification limits were 0.4 and 2.30  $\mu\text{g}$ , respectively. The intra-day and inter-day precisions were 0.4 % and 0.3 % ( $n = 3$ ) respectively. Recovery (by standard addition) was 98.4 %.

herbal, HPTLC, quantitative analysis, densitometry, postchromatographic derivatization 32e

- 106 159 S. MUNNA\*, K. JAYAVEERA, C. CHETTY, K. GNANAPRAKASH, K. ADINARAYANA (\*Annamacharya College of Pharmacy, Rajampet, A.P., India, sreenivasulu\_munna@yahoo.com): Heavy metal analysis of various parts of *Ficus mollis* (vahl) by TLC. International Journal of ChemTech Research 2(2), 807-812 (2010). Chloroform and ethyl acetate extracts of leaves and bark of *Ficus mollis* (Moraceae) were subjected to TLC fingerprint profiling on silica gel with toluene - ethyl acetate - formic acid 16:2:1. Evaluation under UV 254 nm. Derivatization with vanillin-sulfuric acid reagent, followed by heating at 105 °C until colorization. In the bark 7 well-defined zones were observed, whereas in leaves 10 zones were observed. Heavy metal and mineral analysis was performed by atomic absorption spectroscopy.

pharmaceutical research, quality control, herbal, clinical routine analysis,  
qualitative identification, postchromatographic derivatization

32e

- 106 160 Smita NAGAGOUDA\*, A. KARIGAR, V. JOSHI, M. SIKARWAR (\*Sonia College of Pharmacy, Dharwad, Karnataka, India, smithasanglad@ymail.com): Validated HPTLC method for mangiferin in *Salacia chinesis*. Journal of Pharmacy Research 3(5), 1107-1109 (2010). HPTLC of mangiferin in *Salacia chinesis* (Hippocrateaceae) on silica gel with ethyl acetate - methanol 2:3 at 25 °C with chamber saturation for 30 min. Densitometric evaluation at 254 nm. Derivatization by spraying with acetic anhydride-sulfuric acid-ethanol reagent, followed by heating at 110 °C for 2 min. The method was linear in the range of 10–200 ng/band. The plant was found to contain 1.54 % of mangiferin.

herbal, quality control, HPTLC, quantitative analysis, postchromatographic derivatization 32e

- 106 161 I. NAGUIB\*, M. ABDELKAKAWY (\*Analytical Chemistry Department, Faculty of Pharmacy, Beni-Suef University, Beni-Suef, Egypt, inaguieb@bsu.edu.eg): Development and validation of stability indicating HPLC and HPTLC methods for determination of sulpiride and mebeverine hydrochloride in combination. Eur. J. Med. Chem. 45, 3719-3725 (2010). HPTLC of sulpiride and mebeverine on silica gel with ethanol - methylene chloride - triethyl amine 35:15:1. Quantitative determination by absorbance measurement at 221 nm. The  $hR_f$  of sulpiride and mebeverine was 42 and 62, respectively. Linearity was between 0.4-1.4 µg/band for sulpiride and 0.2-1.6 µg/band for mebeverine. Detection and quantification limits were 0.02 and 0.3 µg/band for sulpiride and 0.04 and 0.2 µg/band for mebeverine. The intra-day and inter-day precisions had a %RSD lower than 2.6 %. Recoveries (by standard addition) were 99.3 % and 100.7 % for sulpiride and mebeverine, respectively. The proposed method showed comparable statistical results with the standard HPLC method

quality control, HPTLC, quantitative analysis, densitometry, comparison of methods 32a

- 106 162 R. NANDA\*, S. POTAWALE, V. BHAGWAT, R. DESHMUKH, P. DESHPANDE (\*Dr. D. Y. Patil Institute of Pharmaceutical Sciences & Research, Pimpri, Pune 411018, India, sachin\_potawale@yahoo.co.in, rabindrananda@rediffmail.com): Development and validation of a HPTLC method for simultaneous densitometric analysis of ranitidine hydrochloride and dicyclomine hydrochloride as the bulk drugs and in the tablet dosage form. Journal of Pharmacy Research 3(8), 1997–1999 (2010). TLC on silica gel (plates pre-washed with methanol) with methanol - water - acetic acid 80:20:1. The  $hR_f$  value of ranitidine HCl was 27 and of dicyclomine HCl was 67. Derivatization by exposure to iodine vapor. Densitometric evaluation at 410 nm. The method was linear in the range of 400–2400 ng/band for dicyclomine hydrochloride and 150–900 ng/band for ranitidine hydrochloride. The mean recovery was  $98.8 \pm 0.5$  % for ranitidine HCl and  $99.1 \pm 0.8$  % for dicyclomine HCl.

pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 32a



- 106 163 J. ODOVIC\*, M. ALEKSIC, B. STOJIMIROVIC, D. MILOJKOVIC, Z. TESIC (\*Faculty of Pharmacy, University of Belgrade, 11001 Belgrade, Serbia): Normal-phase thin-layer chromatography of some angiotensin converting enzyme (ACE) inhibitors and their metabolites. *J. Serb. Chem. Soc.* 74(6), 677-688 (2009). An NP-TLC method has been reported to study the lipophilicity of 5 ACE inhibitors (lisinopril, quinapril, fosinopril, enalapril, cilazapril and their metabolites). TLC on silica gel with several non-aqueous mono and binary solvent systems. Binary mobile phases demonstrated a decrease in  $hR_f$  values of all 5 ACE inhibitors, i.e. increased retention with increased amounts of the less polar component in the mobile phase. Metabolites usually exhibited stronger retention, i.e. lower  $hR_f$  values than the corresponding ACE inhibitor. This is probably due to a different interaction with silica gel because of two carboxylic groups in the structure of the metabolites, whereas ACE inhibitors contain only one carboxylic group. Results obtained on NP-TLC were compared with those by RP-TLC and no significant difference was found regarding lipophilicity.
- pharmaceutical research, qualitative identification 32a
- 106 164 P.N. OKUSA\*, C. STÉVIGNY, M. DEVLEESCHOUWER, P. DUEZ (\*Laboratoire de Pharmacognosie, de Bromatologie et de Nutrition Humaine, Université Libre de Bruxelles (ULB), CP 205/09, Bd du Triomphe, 1050 Brussels, Belgium; okusandj@ulb.ac.be): Optimization of the culture medium used for direct TLC-bioautography. Application to the detection of antimicrobial compounds from *Cordia gillettii* De Wild (Boraginaceae). *J. Planar Chromatogr.* 23, 245-249 (2010). TLC of methanol extracts of *Cordia gillettii* on silica gel with dichloromethane, dichloromethane - methanol 9:1 or 4:1, or petroleum ether - diethyl ether - acetic acid 90:10:1. Detection by spraying with 1 % vanillin in ethanol followed immediately by 10 % ethanolic sulfuric acid, and heating at 105 °C for 10 min. For bioautography, 1 mL of 0.5 McFarland microorganism suspension was diluted with 9 mL of the tested mixture of Mueller-Hilton broth and agar 9:1) at 37 °C. After distribution of the inoculated mixture over the plate and solidification of the medium at ambient temperature, the plate was incubated overnight at 37 °C. The bioautogram was subsequently sprayed with an aqueous solution of MTT 0.8 mg mL<sup>-1</sup> and incubated at 37 °C for 4 h. Also application of the Bioluminex assay (a TLC detection method based on the natural bioluminescence of the non-pathogenic bacteria *Vibrio fischerii*). The bioluminescent bacterial coating was visualized by use of the Bioluminizer.
- herbal, quality control, qualitative identification, bioautography 32 e
- 106 165 Mrunali PATEL\*, Rashmin PATEL, J. PARIKH, B. PATEL (\*Indukaka Ipcowala College of Pharmacy, Sardar Patel University, New Vallabh Vidyanagar 388121, India, mrunalipatel@gmail.com): HPTLC method for estimation of tazarotene in topical gel formulations and in vitro study. *Analytical Methods* 2, 275-281 (2010). HPTLC on silica gel with toluene - methanol 9:1. The  $hR_f$  value was 75. Densitometric evaluation at 372 nm. The method was linear in the range of 100-600 ng/band. The recovery was 99.2-100.4 %. The method was suitable for microemulsion based formulations without any interference from excipients.
- pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis 32a
- 106 166 P. PIKUL\*, J. NOWAKOWSKA, P. ROGULSKI (\*Medical University of Gdansk, Department of Physical Chemistry, Al. Gen. Hallera 107, 80-416 Gdansk, Poland; pikul.piotr@gmail.com): Simple thin-layer chromatographic method for analysis of ranitidine in its pharmaceutical formulations: Application to the analysis of photolytic degradation products. *J. Planar Chromatogr.* 23, 304-308 (2010). Chromatographic investigation of ranitidin and degradation products by TLC on silica gel, RP-18, alumina, cellulose, and amino phase and by HPTLC on silica gel and amino phase. HPTLC of ranitidin and degradation products in pharmaceutical preparations on silica gel

with pure solvents (methanol, ethyl acetate, acetonitrile, and DMSO) and binary mixtures (acetonitrile - DMSO, acetonitrile - methanol, and ethyl acetate - methanol) with chamber saturation. The best separation of ranitidine and its photolytic degradation products was obtained on RP-18 W with methanol - water mixtures in a wide range of concentrations as mobile phase. Detection by spraying with sulfuric acid - methanol 1:4 and heating for 10 min at 120 °C.

pharmaceutical research, quality control, qualitative identification, HPTLC 32a

- 106 167 V.J. RAO\*, L. SATHIYANARAYANAN, S. YADAV (\*Dept. of Pharmaceutical Chemistry, Bharati Vidyapeeth University, Poona College of Pharmacy Pune 411038, M.S., India, janhavirao@rediffmail.com): Stability indicating HPTLC method for trandolapril estimation in the bulk drug and tablet dosage form. Indian J. Pharma. Educ. Res. 44(4), 341–344 (2010). HPTLC of silica gel with toluene - ethyl acetate - methanol - formic acid 5:16:21. The  $hR_f$  value was 51. Densitometric evaluation at 220 nm. The method was linear in the range of 300–1800 ng/band. The sample was subjected to different stress conditions (acid, base, oxidative, thermal, and photolytic). Degradation products were well separated from trandolapril, therefore the method can be used for stability studies.

pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis 32a

- 106 168 L. RUSU, C. MARUTOIU, M. RUSU, A. SIMIONESCU, M. RUSU, Z. MOLDOVAN, C. BARBU\* (\*Spiru Haret University, Brazda lui Novac Street, No.4, Craiova, Dolj, Romania, cristina\_barbu2000@yahoo.co.uk): HPTLC and MS for separation and identification of some beta-blockers in urine. Asian Journal of Chemistry 22(6), 4209-4213 (2010). HPTLC of beta-blockers on silica gel with toluene - ethyl acetate - acetone - 25 % ammonia 10:20:20:1. The method was applied to biological samples (urine and blood). The drugs were extracted from biological samples by liquid-liquid extraction. Detection at 254 nm. The corresponding separated spots were scraped from the plate, extracted with methanol and analyzed by mass spectrometry. Finally the identity was confirmed by comparing the spectra with a data library. The proposed HPTLC method coupled with MS is very sensitive and highly suitable for biological samples.

pharmaceutical research, quantitative analysis, HPTLC 32a

- 106 169 G.A. SALEH, F.A. MOHAMED, S.R. EL-SHABOURY, A.H. RAGEH\* (\*Department of Pharmaceutical Analytical Chemistry, Faculty of Pharmacy, Assiut Univeristy, 71526 Assiut, Egypt): Selective densitometric determination of four alpha-aminocephalosporins using ninhydrin reagent. J. Chromatogr. Sci. 48, 68-75 (2010). TLC of cefaclor, cefadroxil, cefalexin and cefradine on silica gel with ethyl acetate - methanol - water in different ratios. The studied drugs had  $hR_f$  values between 40 and 60. Detection of blue to violet colored zones after spraying with ninhydrin reagent. Quantification by densitometry. Linearity was in the range of 2-10 µg/zone with correlation coefficients ranging from 0.9990 to 0.9996 and determination coefficients from 0.9986 to 0.9992 for all studied drugs. The limits of detection and quantification for all studied drugs were between 90 to 230 ng/zone and 270 to 840 ng/zone, respectively.

pharmaceutical research, quality control, HPTLC, densitometry, quantitative analysis, qualitative identification 32c

- 106 170 A. SHARMA, B. PATEL, R. PATEL\* (\*Prof. Parul Institute of Pharmacy, Vaghodiya, Vadodara, Gujarat, India, drbhp@rediffmail.com): Simultaneous estimation of nebivolol hydrochloride and *s*-amlodipine besylate by high-performance thin-layer chromatography. International Journal of Pharma and Bio Sciences 1(4), 339-345 (2010). TLC on silica gel (plates pre-washed with methanol) with chloroform - toluene - methanol - glacial acetic acid 50:20:30:1. The  $hR_f$  value of *s*-amlodipine was 33 and of nebivolol 48. Densitometric evaluation at 271 nm. The linearity range

was 500-2500 ng/zone for nebivolol and 250-1250 ng/zone for s-amlodipine. The method was suitable for routine analysis of the formulation.

pharmaceutical research, quality control, densitometry, quantitative analysis 32c

- 106 171 D. SHEWIYO\*, E. KAALE, C. UGULLUM, M. SIGONDA, P. RISHA, B. DEJAEGHER, J. VERBEKE, Y. HEYDEN (Tanzania Food and Drugs Authority, P.O. Box 77150, Dares Salaam, Tanzania): Development and validation of a normal-phase HPTLC method for the simultaneous analysis of lamivudine, stavudine and nevirapine in fixed-dose combination tablets. *J. Pharm. Biomed. Analysis* 54, 445-450 (2011). An improved HPTLC method has been developed for simultaneous estimation of lamivudine (LVD), stavudine (STV), and nevirapine (NVP) in fixed dose formulation. HPTLC on silica gel with ethyl acetate - methanol - toluene - 25 % ammonia 12:6:12:1. The  $hR_f$  value is 24 for LVD, 38 for STV and 69 for NVP. Densitometric evaluation at 254 nm. The method was linear in the range of 42-209 ng/band (LVD), 38-192 ng/band (STV) and 42-208 ng/band (NVP). The recovery values were in the range of 98.4-102.2 % for all three compounds. The method was found suitable for quality control of LVD, STV and NVP in fixed dose formulation.

pharmaceutical research, quality control, densitometry, quantitative analysis, HPTLC 32c

- 106 172 Malgorzata STAREK\*, J. KRZEK, M. TARSA (\*Department of Inorganic and Analytical Chemistry, Collegium Medicum, Jagiellonian University, 9 Medyczna Str., 30-688 Kraków, Poland; mstarek@interia.pl): TLC-densitometric method for quantification of oxaprozin and its degradation products in pharmaceutical preparations. *J. Planar Chromatogr.* 23, 298-303 (2010). TLC of oxaprozin on silica gel with *n*-hexane - chloroform - glacial acetic acid 4:1:1 with chamber saturation for 15 min at room temperature. The  $hR_f$  value of oxaprozin was 56. Quantitative determination by absorbance measurement at 286 nm. The limit of detection and quantification was 16 and 40 µg/mL, respectively. Linearity was between 33 and 433 µg/mL. The recovery was between 98.9 and 101.6 % at different spiking levels. Intra-day precision (%RSD) at different concentration levels was 0.86-1.03 % and inter-day precision 0.99-1.15 %. Oxaprozin was subjected to stress tests (acidic and alkaline hydrolysis) and the method was able to separate the degradation products from the main compound.

pharmaceutical research, quality control, densitometry, quantitative analysis 32a

- 106 173 T. TUZIMSKI (Department of Physical Chemistry, Faculty of Pharmacy, Medical University of Lublin, Lublin, Poland, tomasz.tuzimski@umlub.pl): Determination of clofentezine in medical herb extracts by chromatographic methods combined with diode array scanning densitometry. *J. Sep. Sci.* 33, 1954-1958 (2010). HPTLC of clofentezine in medical herb extracts on silica gel with tetrahydrofuran - *n*-heptane 3:7. After drying for 20 min, the plate was turned 90 ° and developed with ethyl acetate - *n*-heptane 1:4. Quantitative determination by absorbance measurement at 254 nm. Limits of detection and quantification were 230 and 700 ng/zone respectively. The average recovery (by standard addition) was 55.8 %.

herbal, HPTLC, quantitative analysis, densitometry, comparison of methods 32e

- 106 174 S.J. VARGHESE\*, T.K. RAVI (\*Sri Ramakrishna Institute of Paramedical Sciences, College of Pharmacy, Department of Pharmaceutical Analysis, Coimbatore 641 044, Tamil Nadu, India; susheeljv@yahoo.com): Determination of rosuvastatin and ezetimibe in a combined tablet dosage form using high-performance column liquid chromatography and high-performance thin-layer chromatography. *J. AOAC Int.* 93, 1222-1227 (2010). HPTLC of rosuvastatin (ROS) and ezetimibe (EZE) on silica gel with *n*-butyl-acetate - chloroform - glacial acetic acid 1:8:1 with chamber saturation for 30 min. Quantitative determination by densitometry at 245 nm. The  $hR_f$  value of

ROS was 30 and of EZE 58. The linearity for ROS and EZE was 0.1 to 0.9 µg/zone. The intraday and interday precision was 0.62 % and 0.73 % for ROS and 0.59 % and 0.69 % for EZE ( $n = 3$ ). The LOD values for ROS and EZE were found to be 0.04 and 0.07 µg/spot, respectively, and their LOQ values were 0.07 and 0.1 µg/spot, respectively. The recovery study results ranged from 98 to 101 % for ROS and EZE.

quality control, pharmaceutical research, HPTLC, densitometry, quantitative analysis, comparison of methods 32a

- 106 175 N.E. WAGIEH\*, Maha HEGAZY, M. ABDELKAWY, E.A. ABDELALLEEM (\*Analytical Chemistry Department, Faculty of Pharmacy, Beni-Suef University, Egypt): Quantitative determination of oxybutynin hydrochloride by spectrophotometry, chemometry and HPTLC in presence of its degradation product and additives in different pharmaceutical dosage forms. *Talanta*, 80 (5), 2007-2015 (2010). Presentation of a method for determination of oxybutynin hydrochloride (OX) in presence of its degradation product and additives in its pharmaceutical formulations. UV spectrophotometry using the first derivative of ratio spectra and measurement at 216 nm. Chemometric analysis using principal component regression and partial least-squares. HPTLC of OX and its degradation products methylparaben and propylparaben on silica gel with chloroform - methanol - ammonia - triethylamine 500:15:2.5:1. Quantitative determination by densitometry at 220 nm. Comparison of the results obtained with all three methods showed no significant differences. pharmaceutical research, quality control, qualitative identification, autoradiography, comparison of methods, HPTLC, quantitative analysis 32c

- 106 176 S. WANKHEDE\*, K. RAKA, S. WADKAR, S. CHITLANGE (Dept. of Pharmaceutical Chemistry, Padmashri Dr. D. Y. Patil Institute of Pharmaceutical Science and Research, Sant Tukaram Nagar, Pimpri, Pune 411018, India, drsagarwankhede@rediffmail.com): Development and validation of stability-indicating HPTLC method for analysis of amlodipine besilate, losartan potassium and hydrochlorothiazide in pharmaceutical dosage form. *Journal of Pharmaceutical Research* 9(2), 60-62 (2010). TLC on silica gel with chloroform - ethyl acetate - methanol - ammonia 20:20:10:1 with chamber saturation for 30 min. Densitometric evaluation at 232 nm. The method was linear in the range of 100-800 ng/band for amlodipine, 400-2400 ng/band for hydrochlorothiazide and 1000-8000 ng/band for amlodipine. The average recovery was 99.1-100.4 % for all three compounds. The method was suitable for stability studies (stress conditions: acid, base, thermal, UV). pharmaceutical research, quality control, densitometry, quantitative analysis 32c

- 106 177 H. ZUO (Zuo Hongxiang)\*, Y. JIN (Jin Yong), CH. ZHANG (Zhang Chengyi), J. MA (Ma Jimei) (\*Pharm. Coll. Beihua Univ., Jilin, Jilin 132013, China): (On the quality standard of Kongzhengyizhi tablets) (Chinese). *J. of Beihua Univ. (Natural Sci.)*, 11 (5), 420-423 (2010). TLC on silica gel with 1) petroleum ether (60-90 °C) - ethyl acetate 4:1 and 2) chloroform - propanone - *n*-hexane - acetic acid 80:40:2:5. Detection under UV 365 nm. Derivatization by spraying with 10 % vanillin in sulfuric acid and heating at 105 °C until the zones were visualized. Identification by comparison with the standards of the components in the individual composition drug. pharmaceutical research, traditional medicine, quality control, herbal, HPTLC, qualitative identification, quantitative analysis 32c

### 33. Inorganic substances

- 106 178 A. MOHAMMAD\*, S. BHAWANI (\*Analytical Research Lab., Dept. of Applied Chemistry, Faculty of Eng. & Tech., Aligarh Muslim University, Aligarh, UP, India): On plate resolution of three-component mixture of cationic surfactants with mixed aqueous-organic eluents containing

formate ion. Separation Science and Technology 44, 1007-1021 (2009). A TLC method for mutual separation of three longchain aliphatic quaternary ammonium halides (cationic surfactants) is reported. TLC of dodecyltrimethyl ammonium bromide (DTAB), tetradecyltrimethyl ammonium bromide (TTAB) and hexadecyltrimethyl ammonium chloride (HTAC) on laboratory made TLC plates coated with Kieselguhr (0.25 mm), with methanol - 10 % sodium formate 3:7 as mobile phase. Detection with modified Dragendorff reagent comprised of solution A (containing bismuth subnitrate and KD) and solution B (containing barium chloride). Solutions A and B were mixed 2:1. The compounds appeared as orange-coloured spots. The effects of alcohols (ethanol, *n*-propanol), substitution of the formate ion by benzoate and acetate ion, different adsorbents (silica gel, aluminum), and the interference of metal ions ( $\text{Cu}^{2+}$ ,  $\text{Zn}^{2+}$ ,  $\text{Hg}^{2+}$ ,  $\text{Co}^{2+}$ ,  $\text{Pb}^{2+}$ ) on the resolution of the three surfactants was studied. The limits of detection of DTAB, TTAB, and HTAC estimated were 3.3, 3.1, and 2.8  $\mu\text{g}/\text{zone}$  respectively. The method was applied for separation of these compounds in water samples.

cosmetics, quality control, qualitative identification

33a

### 35. Other technical products and complex mixtures

106 179 V. GHOULIPOUR, S. AMINI, A. HAGHSHENAS, S. WAQIF-HUSAIN\* (\*Chemistry Department, Faculty of Science, Science and Research Branch, Islamic Azad University, P. O. Box 14515-775, Poonak-Hesarak, Teheran, Iran; syedwaqifhusain@yahoo.com): Chromatographic behavior of food additives on thin layers of titanium(IV) silicate ion-exchanger. J. Planar Chromatogr. 23, 250-254 (2010). TLC of 30 food additives (aldehydes, organic acids, esters and alcohols, and various sodium salts) on titanium(IV)silicate ion-exchanger with 10 different mobile phases: e. g. methanol, heptane - diethylether 4:1, aqueous ammonia - methanol - ethyl acetate 1:3:6, methanol - aqueous ammonia 9:1, methanol - ethanol 17:3, 0.5 M ammonium sulfate solution, 0.25 M, 0.5 M and 0.1 M potassium bromide solution, and phosphate buffer pH 2.5 in a twin-trough chamber without chamber saturation. Detection by spraying with 1 % iron chloride solution, 0.5 % potassium permanganate and 3 % barium chloride solution 1:1, 1 % ninhydrin in ethanol, 2 % phosphomolybdic acid in ethanol, and 5 % potassium dichromate in concentrated sulfuric acid. The study shows that the quality of separation depends to a large extent on the mobile phase, and selectivity is achieved by varying the composition of the mobile phase.

food analysis, qualitative identification

35b

106 180 M. IDRIS, S. SRIVASTAVA, T. R. BAGGI, S. K. SHUKLA\* (\*Central Forensic Science Laboratory, Directorate of Forensic Science, Ministry of Home Affairs, Government of India, Ramanthapur, Hyderabad 500 013 India; drskshukla@gmail.com): High-performance thin-layer chromatography analysis of saccharin in foods and beverages. J. Planar Chromatogr. 23, 339-342 (2010). HPTLC of saccharin in foodstuffs (e. g. cola drinks, lemon juices, betel nut powder, mouth fresheners, ice candy, and tabletop sweeteners) on silica gel with chloroform - methanol - acetic acid 64:35:1 or acetone - isopropanol - acetic acid 60:39:1. Quantitative determination by absorbance measurement at 230 nm. Linearity was between 250 - 1250  $\text{ng}/\mu\text{L}$ . The limit of detection and quantification for saccharin were 40 and 130 ng, respectively. Mean recovery from spiked samples was 102.3 % for cola drinks and 98.8 % for lemon juices. Relative standard deviation (% RSD) for cola drinks, lemon juices, ice candy, mouth freshener, betel nut powders, and tabletop sweeteners were 2.1, 4.2, 3.4, 3.0, 4.9, and 4.1 %, respectively.

food analysis, quality control, HPTLC, densitometry, quantitative analysis

35c

106 181 G. XIAO, G. LI, L. CHEN, Z. ZHANG, J.J YIN, T. WU\*, Z. CHENG, X. WEI, Z. WANG (\*The MOE Key Laboratory for Standardization of Chinese Medicines, the SATCM Key Laboratory for New Resources and Quality Evaluation of Chinese Medicines, and the Shanghai Key Laboratory for Compound Chinese Medicines, Institute of Chinese Materia Medica, Shanghai University of

Traditional Chinese Medicine, 1200 Cailun Road, Zhangjiang Hi-Tech Park, Shanghai 201210, China): Isolation of antioxidants from *Psoralea corylifolia* fruits using high-speed counter-current chromatography guided by thin layer chromatography-antioxidant autographic assay. *J. Chromatogr. A* 1217(34), 5470-5476 (2010). Description of a combination method using high-speed counter-current chromatography and TLC as an antioxidant assay to separate antioxidant components from the fruits of *Psoralea corylifolia* by dipping in a ethanolic DPPH radical solution (2.54 mM) for autography. Bands with the DPPH scavenging activity were observed as white yellow bands on a purple background. High-speed counter-current chromatography of five flavonoids and three coumarins from the fruits of *P. corylifolia* with *n*-hexane - ethyl acetate - methanol - water 10:11:13:1. Preparative TLC for clean-up improves the substance yield from the fruits.

herbal, pharmaceutical research, qualitative identification, autoradiography,  
preparative TLC

35

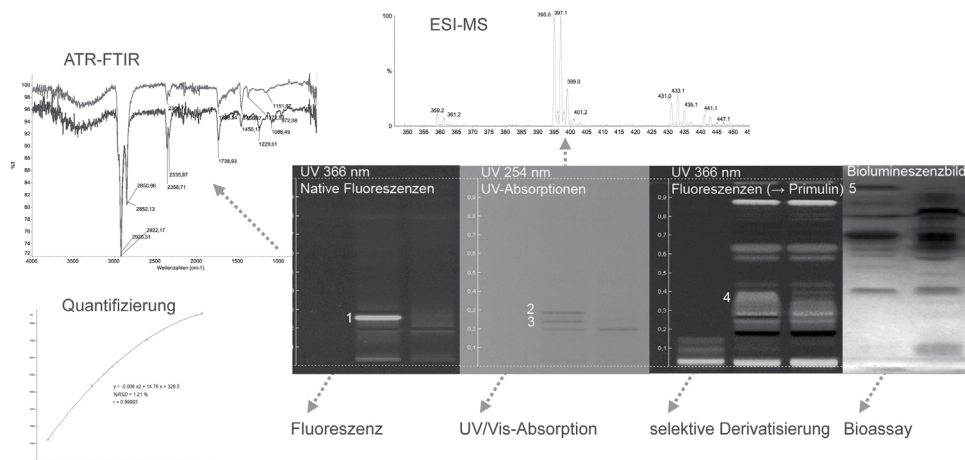
### 38. Chiral separation

106 182 R. BHUSHAN\*, Shivani TANWAR (\*Department of Chemistry, Indian Institute of Technology Roorkee, Roorkee 247667, India): Different approaches of impregnation for resolution of enantiomers of atenolol, propranolol and salbutamol using Cu(II)-L-amino acid complexes for ligand exchange on commercial thin-layer chromatographic plates. *J. Chromatogr. A* 1217(8), 1395-1398 (2010). TLC of atenolol, propranolol and salbutamol and their enantiomers by using different modes of loading/impregnating the Cu(II) complexes of L-proline, L-phenylalanine, L-histidine, N,N-dimethyl-L-phenylalanine, and L-tryptophan on silica gel with 1) the Cu(II)-L-amino acid complex as chiral mobile phase additive, 2) ascending development of plain commercial plates in the solutions of Cu complex, and 3) using a solution of Cu(II)acetate as mobile phase additive for the commercial TLC plates impregnated by development with the amino acid solutions. Detection by exposure to iodine vapor. The detection limit was 0.18 µg for each enantiomer. For the best method performance the Cu(II)cation has to be involved.

quality control, pharmaceutical research, quantitative analysis, qualitative identification 38

# GDCh-Kurs 335/11 Hochleistungs-Dünnschicht-Chromatographie- Massenspektrometrie (HPTLC-MS)

in Zusammenarbeit mit der Universität Hohenheim, Stuttgart



## Ziele

- Potential der HPTLC erkennen
- Aktuelle Kopplungen der HPTLC kennenlernen
- Erkennen, wie Hyphenationen (Kopplungen) in der HPTLC die Analytik effizient unterstützen

## Zielgruppe

Analytiker, Lebensmittelchemiker, Pharmazeuten, Chemiker etc., die das Potential der HPTLC samt ihren Kopplungsmöglichkeiten für ihre Zwecke ausloten möchten

## Veranstaltungsort

Universität Hohenheim, Institut für Lebensmittelchemie, Garbenstrasse 28, 70599 Stuttgart

## Programm am Dienstag, 15.11.2011

- 9:30 Begrüßung und Einführung in die HPTLC (Morlock)
- 10:00 HPTLC erfahren – Experimente Gruppe 1 (Oellig), Gruppe 2 (Morlock)
- 11:00 Kaffeepause
- 11:15 Hyphenationen in der Planar-Chromatographie (Morlock)
- 12:00 Gruppe 1 Experiment DC-HPLC/DAD-ESI/MS (Schwack, Oellig)  
Gruppe 2 Experiment HPTLC-UV/Vis/FLD-MALDI-TOF MS/MS (Schürenberg)
- 12:45 Gruppe 1 Experiment HPTLC-UV/Vis/FLD-Bioaktivität-ESI/MS (Morlock)  
Gruppe 2 Experiment HPTLC-UV/Vis/FLD-ATR FTIR (Dytkiewitz)
- 13:30 Mittagspause (Mensaessen)
- 14:00 Gruppe 1 Experiment HPTLC-UV/Vis/FLD-MALDI-TOF MS/MS (Schürenberg)  
Gruppe 2 Experiment DC-HPLC/DAD-ESI/MS (Schwack, Oellig)
- 14:45 Gruppe 1 Experiment HPTLC-UV/Vis/FLD-ATR FTIR (Dytkiewitz)  
Gruppe 2 Experiment HPTLC-UV/Vis/FLD-Bioaktivität-ESI/MS (Morlock)
- 15:30 Kaffeepause (Umbau ESI auf DART)
- 15:45 Experiment HPTLC-UV/Vis/FLD-DART-MS (Morlock)
- 16:15 Diskussion der Ergebnisse anhand der Vergleichsproben (Morlock)
- 16:45 Ende

**Kursleitung:** apl. Prof. Dr. G. Morlock, Universität Hohenheim, Stuttgart

## Referenten:

- Prof. Dr. Wolfgang Schwack, Universität Hohenheim, Stuttgart
- Dr. Martin Schürenberg, Bruker Daltonik GmbH, Bremen
- Claudia Oellig, Universität Hohenheim, Stuttgart
- Elisabeth Dytkiewitz, Universität Hohenheim, Stuttgart

**Teilnehmerzahl:** max. 16 Personen

**Gebühren:** GDCh-Mitglied € 570.–, Nichtmitglied € 680.–

**Anmeldeschluss:** 18.10.2011

**Anmeldung:** siehe [www.gdch.de/vas/fortbildung.htm](http://www.gdch.de/vas/fortbildung.htm) oder per E-Mail an [fb@gdch.de](mailto:fb@gdch.de)

## High-performance thin-layer chromatography – mass spectrometry (HPTLC-MS)

This course organized by the German Chemical Society will be held November 2011 in German language. The same course in English is tentatively planned for November 2012.

## Objectives

- Recognize the potential of HPTLC
- Learn more about current hyphenations in HPTLC
- Figure out how HPTLC hyphenations efficiently support analyses

## Target group

Analysts, food chemists, pharmacists, chemists etc. who like to evaluate for their projects the benefits of hyphenating HPTLC with mass spectrometry etc.

## Location

University of Hohenheim, Institute of Food Chemistry, Garbenstrasse 28, 70599 Stuttgart

## Program

- 9:30 Welcome and introduction in HPTLC
- 10:00 Realize the power of HPTLC – experiments
- 11:15 Hyphenations in planar chromatography
- 12:00 Group 1 Experiment TLC-HPLC/DAD-ESI/MS  
Group 2 Experiment HPTLC-UV/Vis/FLD-MALDI-TOF MS/MS
- 12:45 Group 1 Experiment HPTLC-UV/Vis/FLD-bioassay-ESI/MS  
Group 2 Experiment HPTLC-UV/Vis/FLD-ATR FTIR
- 13:30 Lunch at the cafeteria
- 14:00 Group 1 Experiment HPTLC-UV/Vis/FLD-MALDI-TOF MS/MS  
Group 2 Experiment TLC-HPLC/DAD-ESI/MS
- 14:45 Group 1 Experiment HPTLC-UV/Vis/FLD-ATR FTIR  
Group 2 Experiment HPTLC-UV/Vis/FLD-bioassay-ESI/MS
- 15:45 Experiment HPTLC-UV/Vis/FLD-DART-MS
- 16:15 Discussion of the results based on a common sample

**Course leader:** Assoc. Prof. Dr. G. Morlock, University of Hohenheim, Stuttgart

## Further tutors:

- Prof. Dr. Wolfgang Schwack, University of Hohenheim, Stuttgart
- Dr. Martin Schürenberg, Bruker Daltonik GmbH, Bremen
- Claudia Oellig, University of Hohenheim, Stuttgart
- Elisabeth Dytkiewitz, University of Hohenheim, Stuttgart

**Number of participants:** max. 16

**Fee:** GDCh members € 570.–, non-members € 680.–

**Interested?** Please email [fb@gdch.de](mailto:fb@gdch.de)



15:3:1, Laufstrecke 6 cm, Laufzeit 15 min; automatisierte Trocknung für je 1 min nach der Auftragung und Chromatographie; durch die Chromatographie wird die Startfläche zu einer Bande fokussiert.

## Postchromatographische Derivatisierung

Mit der Chromatogramm-Tauchvorrichtung (Vertikalgeschwindigkeit 2,5 cm/s, Verweilzeit 0 s) wurde die Platte in das Diphenylamin-Anilin-*o*-Phosphorsäure-Reagenz getaucht und anschliessend auf dem DC-Plattenheizer 20 min bei 120°C erhitzt. Das Reagenz (Je 1,5 g Anilin und Diphenylamin wurden in 150 mL Methanol gelöst und 15 mL *o*-Phosphorsäure wurden tropfenweise hinzugegeben.) ist mindestens 1 Monat im Kühlschrank haltbar.

## Dokumentation

Mit dem DigiStore 2 Dokumentations-System, Belichtungszeit 7 ms (Weisslicht, Auf- und Durchlicht kombiniert) und 1200 ms (UV 366, Auflicht), Verstärkung 1

## Densitometrie

TLC-Scanner 3 mit winCATS Software, Absorptionsmessung bei 400 nm, Spaltgrösse 6 × 0.45 mm, Messgeschwindigkeit 20 mm/s

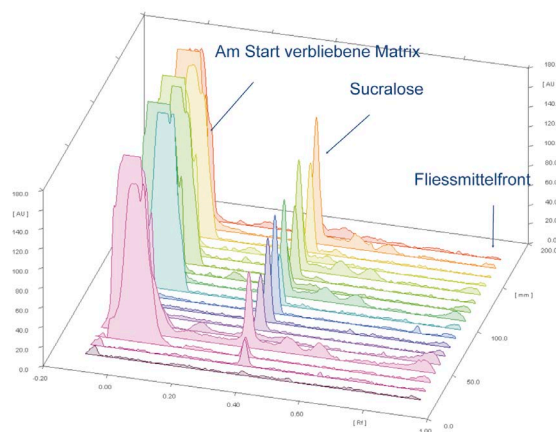
## Massenspektrometrie

Mit dem TLC-MS-Interface wurde die entsprechende Zone mit Methanol – Ammoniumformiat-Puffer (10 mM, pH 4) 19:1 bei einer Flussrate von 0.1 mL/min (HP 1100 Pumpe, Agilent) eluiert und direkt in das Single-Quadrupol-Massenspektrometer (MSD, Agilent) überführt. Das Elektrospray-Ionisations (ESI)-Massenspektrum wurde im full scan-Modus aufgenommen: Kapillarspannung + 4 kV bzw. – 4 kV, Vernebelungsgasdruck 20 psig, Trocknungsgas-Temperatur 300 °C, Trocknungsgasfluss 10 L/min, Fragmentor-Spannung 100 V, Verstärkung 1, Schwellenwert 10 und Schrittgrösse 0.25.

## Ergebnisse und Diskussion

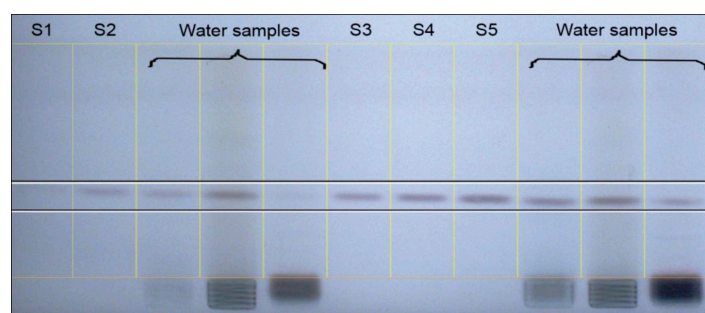
Zur Methodvalidierung wurde die mittlere Wiederfindungsrate von Sucralose in Trinkwasser mit  $84 \pm 7\%$  ( $n = 3$ , Dotierungsniveau  $1 \mu\text{g/L}$ )

bestimmt. Bei Extraktion einer 0,5 L-Probe mit einer Wiederfindungsrate von 80 % und 300  $\mu\text{L}$  Extrakt-Auftragung betrug die Nachweisgrenze 100 ng/L (6 ng/Band). Die Kalibrierkurve (10–300 ng/Zone, Korrelationskoeffizient  $r = 0.9999$ , relative Standardabweichung  $\%RSD = 1.3\%$ ) ermöglichte die Quantifizierung von Sucralose in Wasserproben in einem Konzentrationsbereich zwischen 0.1 und 5  $\mu\text{g/L}$ . Für die Kalibration in Matrix wurden Probe-Startzonen (mehrfach aufgetragen) mit unterschiedlichen Sucralosemengen übersprüht. Die Steigung dieser Kalibrierkurve war vergleichbar mit der der externen Standard-Kalibration, die somit für die Routineanalytik geeignet war und bevorzugt wurde. Allerdings wurde für geringe Sucralosekonzentrationen ( $< 200 \text{ ng/L}$ ) die Kalibration in Matrix gewählt, um potentielle Matrixeinflüsse zu korrigieren.



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Zum Eignungsnachweis der neuen HPTLC-Methode wurden in einem Ringversuch erhaltene Wasserproben analysiert und die Ergebnisse mit den jeweiligen Ringversuchs-Mittelwerten verglichen, die mit HPLC-MS grösstenteils nach Korrektur durch deuterierte Standards bestimmt wurden. Die über die Wiederfindungsrate korrigierten HPTLC-Ergebnisse zeigten eine hohe Genauigkeit, und der Bias (Abweichung) zu den angegebenen Mittelwerten lag zwischen 0 und 33 %. Vergleicht man diese mit dem entsprechenden Ringversuchsergebnis (Sollwert), so waren beide Mittelwerte statistisch nicht unterscheidbar (berechnete t-Werte  $<$  t-Tabellen-Wert von 12,7,  $P = 95\%$ , zweiseitig).



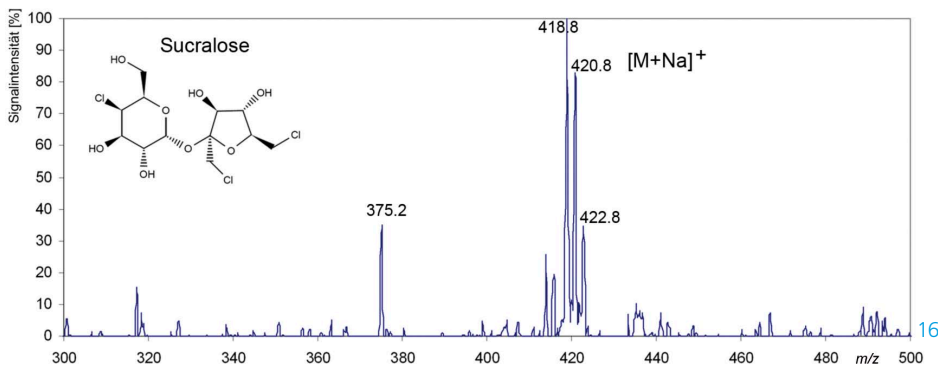
15



Sucralose in Probe (ng/L)	Abwasser I	Abwasser II	Oberflächenwasser I	Oberflächenwasser II
Mittelwerte HPTLC-Vis (%RSD, n = 2)	5863 ± 5	7034 ± 1	247 ± 28	218 ± 13
Mittelwerte HPLC-MS/MS oder HPLC-TOF MS (%RSD, n = 6 Labore*)	5869 ± 17	7302 ± 24	186 ± 21	200 ± 23
Bias in %	0.1	3.7	32.8	9.0
Berechnete t-Werte	0.04	5.42	1.25	0.90

\*meist mit teuren, Isotopen-markierten Standards korrigiert

In den aufgenommenen Massenspektren von der nicht derivatisierten Platte waren das Formiat-Addukt die am deutlichsten ausgeprägte Signalgruppe im negativen Ionisationsmodus und das Natrium-Addukt im positiven Modus, wobei das 3-fache Chlor-Isotopenmuster deutlich zu erkennen war. Mit MS-Standard Einstellungen lag die Nachweisgrenze (S/N von 3) in der Wasserprobe bei etwa 100 ng/Zone.



Da diese HPTLC-Methode darauf ausgelegt ist, Hauptquellen des Sucralose-Eintrags zu finden, ist ein hoher Probendurchsatz notwendig. Bis zu 17 Proben können bei geringen laufenden Kosten innerhalb von 1,4 Stunden (ohne SPE) parallel analysiert werden. Im Vergleich zu den bisher eingesetzten Methoden ist diese neue Methode damit sehr effektiv – zumal die HPTLC mit einer einfachen selektiven Derivatisierung zu vergleichbaren Ergebnissen führte wie die Analytik mit HPLC-TOF MS oder HPLC-MS/MS und Isotopen-markierten Standards.

Weitere Informationen sind vom Autor auf Anfrage erhältlich.

- [1] G. Morlock, S. Prabha J. Agric. Food Chem. 55 (2007) 7217
- [2] G. Morlock, M. Vega J. Planar Chromatogr. 20 (2007) 411
- [3] G. Morlock *et al.* J. Chromatogr. A, doi 10.1016/j.chroma.2010.11.063

Kontakt: apl. Prof. Dr. G. Morlock, Universität Hohenheim, Institut für Lebensmittelchemie, Garbenstrasse 28, 70599 Stuttgart; gerda.morlock@uni-hohenheim.de

## Fingerprint-Analyse von pflanzlichen ätherischen Ölen mit der Planar-Chromatographie und durch das TLC-MS-Interface verbunden mit der Flüssigkeits-Chromatographie



17

Prof. Teresa Kowalska und Dr. Mieczysław Sajewicz

Planar-Chromatographie ist der Forschungsschwerpunkt von Prof. Teresa Kowalska und Dr. Mieczysław Sajewicz im Fachbereich für Allgemeine Chemie und Chromatographie der Schlesischen Universität in Kattowitz, Polen. Die DC/HPTLC wird täglich für analytische und physikalisch-chemische Fragestellungen genutzt. Im gut ausgerüsteten Labor des Fachbereichs haben Prof. Kowalska und Dr. Sajewicz in vielen Vergleichen zwischen der Planar-Chromatographie und anderen chromatographischen Techniken festgestellt, dass die HPTLC auf diversen Anwendungsgebieten mit der HPLC vergleichbar und teilweise sogar überlegen ist. Zurzeit beschäftigt sich die Forschungsgruppe mit den Möglichkeiten des TLC-MS-Interface und untersucht neue, noch nicht vollständig anerkannte Einsatzbereiche dieses nützlichen Geräts.

### Einleitung

Die aktuellsten analytischen Fragestellungen kommen häufig aus den Bereichen der biomedizinischen Analytik, der Umwelt- und der Lebensmittelanalytik. Ein Schwerpunkt ist das schnelle Screening einer grossen Anzahl von Proben aus dem Pharma- und Lebensmittelbereich hinsichtlich Verfälschung oder Kontamination. Die Schwierigkeiten bei der chemischen Analytik pflanzlicher Proben sind einerseits die nicht vorhersehbare chemische Zusammensetzung

eines bestimmten Pflanzenextraktes, andererseits die hohen Kosten der phytochemischen Referenzstandards. Für das schnelle Screening von Pflanzen, die meist in getrockneter Form oder als Granulat gehandelt werden, sowie für chemotaxonomische Studien durch Botaniker, die neben morphologischen auch an chemischen Ähnlichkeiten und Unterschieden zwischen den einzelnen Spezies interessiert sind, erweist sich ein instrumentelles HPTLC-System als sehr wertvoll. Dies kann in Kombination mit einem geeigneten Detektions-System die effiziente Fingerprint-Analyse pflanzlicher Proben zu ihrer raschen und sicheren Identifikation unterstützen.

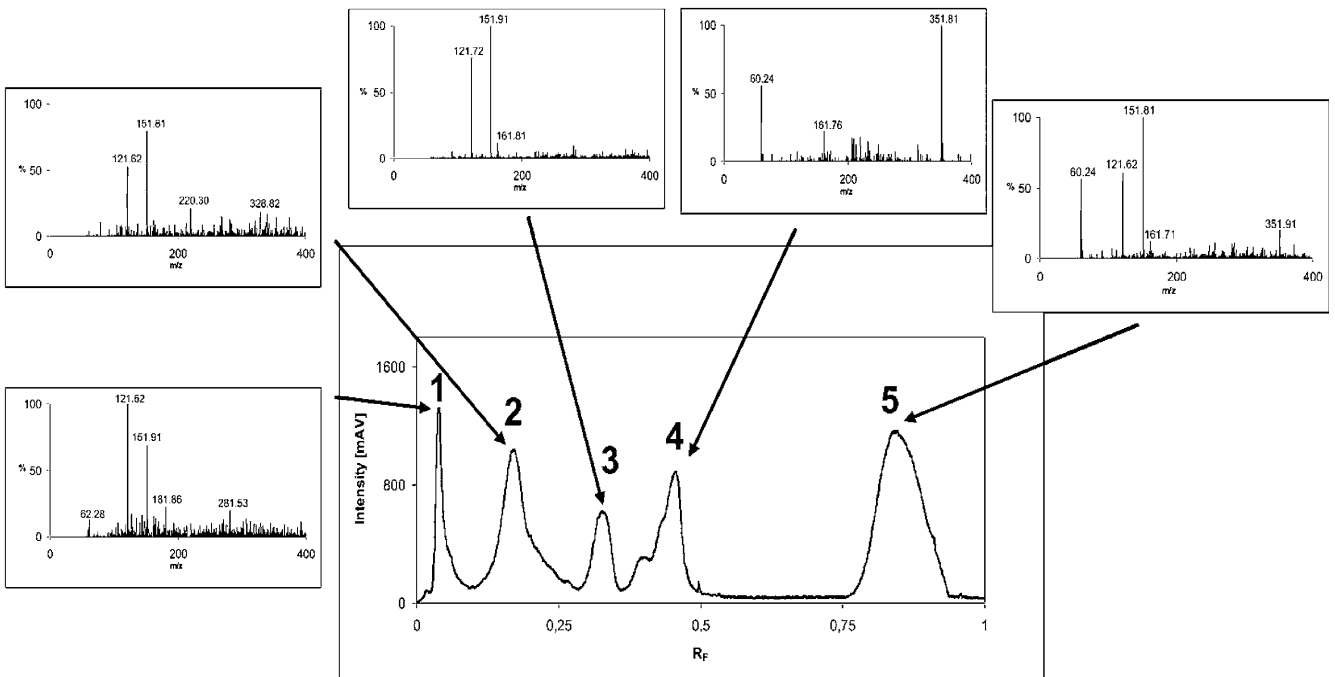
Eine bemerkenswerte Eigenschaft der Planar-Chromatographie ist, dass sie sogar zur Trennung flüchtiger Fraktionen (z.B. ätherischer Öle) eingesetzt werden kann, sofern die Trennung bei tiefen Temperaturen (-20 oder -10 °C) und auf Kieselgel als starkem Adsorbens [1, 2] durchgeführt wird. In unseren Untersuchungen konnten wir dies erfolgreich am Beispiel von ätherischen Ölen verschiedener Arten der Gattung Salbei (*Salvia*) zeigen [3].

**Es ist offensichtlich, dass sich die HPTLC dank ihrer Einfachheit und Vielseitigkeit und in Kombination mit der exakten Detektion durch die Massenspektrometrie in naher Zukunft erfolgreich als besonders vielseitiges und praktisches Werkzeug zum schnellen Screening mit den Techniken der instrumentellen Flüssigkeits- und Gas-Chromatographie messen kann.**

*Anmerkung: Nachfolgend wird es anhand konventioneller DC-Platten gezeigt; es liesse sich die Methode für den Einsatz von HPTLC-Schichten leicht und mit erheblichen Vorteilen anpassen: Bessere Trennung, kürzere Laufzeiten und Flussmittel-Ersparnis.*

### Probenvorbereitung

Die verschiedenen Pflanzeninhaltsstoffe (z.B. ätherische Öle, Phenolsäuren, Flavonoide etc.) können



Densitogramm und Massenspektren der 5 getrennten chromatographischen Zonen der ätherischen Öle von *Salvia lavandulifolia*

unter anderem durch Wasserdampfdestillation, klassische Flüssig-Flüssig-Extraktion oder beschleunigte Lösungsmittel-Extraktion mehr oder weniger selektiv aus pflanzlichem Material isoliert werden.

### Schicht

DC-Platten Kieselgel 60 F<sub>254</sub> (Merck), 20 × 20 cm

### Probenauftragung

Punktförmig, Auftragsvolumen 5 µL des unverdünnten ätherischen Öls

### Chromatographie

In der Doppeltrogkammer bei  $-10 \pm 0.5$  °C mit Toluol – Ethylacetat 95:5 bis zu einer Laufstrecke von 15 cm

### Densitometrie

Absorptionsmessung bei 340 nm

### Massenspektrometrie

Interessierende Zonen wurden markiert und mit dem TLC-MS-Interface direkt in ein Varian 500 Massenspektrometer eluiert. Die Massenspektren wurden im positiven Elektrospray-Ionisations-Modus mit der MS-Software Workstation v.6.9.1 aufgenommen: Full scan, Temperatur der Sprühkammer 45 °C, Trägergas-Temperatur 150 °C, Trägergasdruck 25 psi, Kapillarspannung 70 V, Nadelspannung 5 kV.

## Ergebnisse und Diskussion

Das Densitogramm des durch Wasserdampfdestillation aus *Salvia lavandulifolia* gewonnenen ätherischen Öls weist fünf getrennte Zonen auf, von denen jeweils ein Massenspektrum aufgenommen wurde [4].

Diese Methode kann auf die verschiedensten komplexen Substanzgemische angewandt werden [5, 6]. Im Gegensatz zur vorherrschenden Meinung kann mit der Elektrospray-Ionisations-Massenspektrometrie (ESI-MS) auch unter milden Bedingungen eine Fragmentierung der Substanzen erreicht werden. So können zum Beispiel die zu bestimmten Fragmenten gehörenden Ionen in den verschiedenen Massenspektren des aus *Salvia lavandulifolia* gewonnenen ätherischen Öls zugeordnet werden; in den Spektren sind die Fragmente mit  $m/z$  60, 122 und 152 erkennbar. Trotz der komplexen Fingerprints der untersuchten Fraktionen können die interessierenden Substanzen zuverlässig identifiziert werden.

Die bestehenden Kopplungen mit DC/HPTLC [7, 8] werden durch das TLC-MS Interface erweitert, da es multidimensionale flüssigkeits-chromatographische Konfigurationen ermöglicht. So kann man eine interessierende Zone direkt in ein mit einem beliebigen Detektor ausgerüstetes HPLC-System transferieren und diverse Vorteile nutzen: Eine schnelle Vorfraktionierung komplexer Mischungen mittels

Planar-Chromatographie, gefolgt von einer verbesserten Trennung mit HPLC und unterschiedlichsten Detektionsmöglichkeiten wie:

- MS-Detektor (siehe CBS 103, 2009, 13–15)
- UV/VIS-Detektor
- Diodenarray-Detektor (DAD)
- Lichtstreu-Detektor (ELSD, siehe CBS 105, 2010, 2–5)
- Polarimetrischer Detektor
- Zirkulardichroismus-Detektor
- Weitere Detektionssysteme.

Es wurde eine vergleichende Untersuchung derselben Pflanzenextrakte mittels eindimensionaler (1D) DC-MS und zweidimensionaler (2D) DC-LC-MS durchgeführt [4–6]. Das zweidimensionale System erwies sich als vorteilhafter, da es im Vergleich zum eindimensionalen eine grössere Menge analytischer Daten liefert (in Form der massenspektrometrischen Fingerprints). Die bemerkenswerte Flexibilität des über das TLC-MS-Interface gekoppelten DC-HPLC-Systems basiert auf der selektiven Multidimensionalität, da nicht alle Zonen transferiert werden müssen. Zusätzlich kann die HPLC-Trennung jeder einzelnen Zone des Chromatogramms unter verschiedenen Bedingungen durchgeführt werden: Mit unterschiedlichen HPLC-Säulen bzw. Eluenten-Zusammensetzungen, im isokratischen oder Gradientenmodus und mit dem bestgeeigneten Detektor.

Weitere Informationen sind von den Autoren auf Anfrage erhältlich.

- [1] C. Mathis, G. Ourisson *Phytochemistry* 3 (1964) 115
- [2] A. Koch *et al.* in *Thin Layer Chromatography in Phytochemistry*, M. Waksmundzka-Hajnos, J. Sherma, T. Kowalska (Eds.) CRC Press, Boca Raton, USA, 2008, 451
- [3] M. Sajewicz *et al.* *J. Planar Chromatogr.* 23 (2010) 270
- [4] M. Sajewicz *et al.* *J. Liq. Chromatogr. Relat. Technol.*, in press
- [5] M. Sajewicz *et al.* *J. Liq. Chromatogr. Relat. Technol.*, in press
- [6] M. Sajewicz *et al.* *J. Chromatogr. Sci.*, in press
- [7] G. Morlock, W. Schwack *Trends Anal. Chem.* 10 (2010) 1157
- [8] G. Morlock, W. Schwack *J. Chromatogr. A* 1217 (2010) 6600

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### CAMAG TLC-MS Interface

Dieses Interface ermöglicht die schnelle und kontaminationsfreie Elution von TLC/HPTLC-Zonen mit online Übertragung an das jeweilige Massenspektrometer. Der Vorteil dieses universellen Interface liegt darin, dass es ohne Modifizierung in jedes beliebige HPLC/MS-System plug & play integriert werden kann.

Je nach angeschlossenem Massenspektrometer kann innerhalb einer Minute die Substanz über ihr Massenspektrum identifiziert oder bei unbekanntem Substanzzonen die dazugehörige Summenformel erhalten werden. Die Detektierbarkeit ist vergleichbar zur HPLC-MS, da die gesamte Substanzzone mit ihrem Tiefenprofil extrahiert wird.

Auch kann man interessierende Zonen für weitere Untersuchungen mit z. B. NMR, (ATR) FTIR, Statisches Nanospray, Direkteinlass-EI-MS und MALDI in ein Vial extrahieren.

## HPTLC für die Qualitätskontrolle in der Traditionellen Chinesischen Medizin



Professor Dr. Irmgard Merfort und ihre Arbeitsgruppe

Herr Zhi Li hat im Rahmen seiner Diplomarbeit, betreut durch Frau Professor Irmgard Merfort\*, Lehrstuhl für Pharmazeutische Biologie und Biotechnologie, Institut für Pharmazeutische Wissenschaften der Albert-Ludwigs-Universität Freiburg, im CAMAG Labor an der Entwicklung und Validierung von HPTLC-Methoden für die Qualitätskontrolle von pflanzlichen Kombinationspräparaten gearbeitet.

### Einleitung

Pflanzliche Kombinationspräparate und Mischungen von Arzneipflanzen oder Pflanzenextrakten sind typische Bestandteile der Traditionellen Chinesischen Medizin (TCM) und auch anderer traditioneller Medizinsysteme. Für viele dieser Arzneimittel und der darin enthaltenen Einzelpflanzendrogen bestehen in den jeweiligen Arzneibüchern schon seit langem Monographien. Diese Monographien entsprechen jedoch oft nicht den Richtlinien der heute für die Herstellung von TCM-Arzneimitteln gültigen Guten Herstellungspraxis (cGMP), da sie häufig weder eine eindeutige Identifikation der Einzelkomponenten noch eine geeignete Methode zur Gehaltsbestimmung des Fertigarzneimittels enthalten.

**Am Beispiel des traditionellen chinesischen Tierarzneimittels Cangzhu Xianglian San (CXS) und basierend auf der HPTLC-Fingerprintanalyse wurde ein systematischer Ansatz für die Optimierung der Qualitätskontrolle pflanzlicher Kombinationspräparate entwickelt [1]. Die HPTLC erwies sich dabei als schnelle und leistungsfähige Methode für die korrekte Identifikation, den Nachweis von Verfälschungen sowie die quantitative Analyse pflanzlicher Vielstoffgemische.**

### Probenvorbereitung

Kommerzielle Produkte von CXS und Proben der darin enthaltenen Pflanzendrogen *Coptis rhizoma*, *Aucklandia radix* und *Atractylodes radix* wurden mit Methanol (0.1 g/mL) während 15 min im Ultraschall extrahiert. Für die Quantifizierung von Berberin wurden 25.0 mg CXS mit 50 mL Methanol während 30 min im Ultraschallbad bei 60 °C extrahiert, nach dem Abkühlen wurde das Volumen auf 50.0 mL ergänzt und der Extrakt filtriert.

### Standardlösungen

Methanolische Lösungen (1.0 mg/mL) von Berberin HCl, Palmatin HCl, Jatrorrhizin HCl, Dehydrocostuslacton und Costunolid wurden hergestellt. Für die Quantifizierung wurde eine Lösung von 3.3 µg/mL Berberin in Methanol verwendet.

### Schicht

HPTLC-Platten Kieselgel 60 F<sub>254</sub> (Merck), 20 × 10 cm, vorgewaschen durch Chromatographie mit Methanol, danach während 20 min bei 120 °C im Trockenschrank getrocknet

### Probenauftragung

Bandförmig mit DC-Probenautomat 4, Bandlänge 8 mm, Bahnabstand mind. 10 mm, unterer Randabstand 8 mm, seitlicher Randabstand mind. 15 mm, Auftragevolumina 2–10 µL für Proben und 1–8 µL für Standardlösungen

### Chromatographie

In der Automatischen Entwicklungskammer ADC2 nach Kammersättigung (mit Filterpapier) für 20 min mit Toluol, Ethylacetat, Methanol, Isopropanol, Wasser 60:30:20:15:3 im vorderen Trog und 10 mL 25 % Ammoniaklösung im hinteren Trog der Doppeltrogkammer, Laufstrecke 70 mm vom unteren Plattenrand. Da die Trennung von der Plattenaktivität beeinflusst wird, wurden die Platten während 10 min mit einer gesättigten MgCl<sub>2</sub>-Lösung bei 33 % relativer Feuchte konditioniert.

### Dokumentation

Mit dem TLC Visualizer unter UV 366 nm

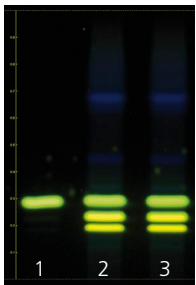
### Densitometrie

Fluoreszenzmessung bei 366/>400 nm mit dem TLC Scanner 3 mit winCATS Software



## Ergebnisse und Diskussion

Zwei kommerzielle CXS-Produkte wurden nach der DC-Methode in der offiziellen Monographie des chinesischen Tierarzneibuchs (Chinese Veterinary Pharmacopoeia 2005, ChVPh) untersucht. Beide Proben wiesen im Chromatogramm an der Position von Berberin eine gelbgrün fluoreszierende Zone auf und entsprachen damit den Anforderungen der Monographie. Zweifel am Nutzen dieser DC-Methode waren jedoch angebracht, da Berberin ein weit verbreiteter Inhaltsstoff vieler TCM-Arzneimittel ist und auch jede beliebige Pflanzendroge, die mit Berberin versetzt wurde, den Identifikationstest bestehen würde.

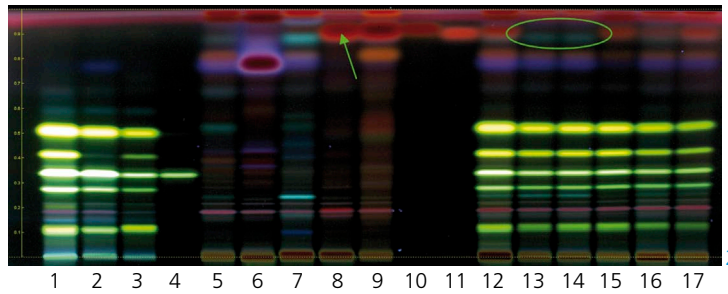


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Identifikation von CXS nach ChVPh 2005. Bahn 1: Berberin HCl, 2,3: CXS (2 Chargen eines Herstellers).

Zunächst wurden HPTLC-Methoden zur Identifikation der einzelnen Komponenten von CXS erarbeitet. Das Ziel war die eindeutige Unterscheidung a) aller drei zugelassenen *Coptis*-Arten, b) von *Aucklandia radix* und ihren Verfälschungen *Vladimiria radix* und *Inula radix*, sowie c) der zwei zugelassenen Arten von *Atractylodes rhizoma* und der präparierten *Atractylodes*-Droge. Dann wurde die Eignung der Methoden untersucht, jede einzelne Komponente im pflanzlichen Kombinationspräparat CXS nachzuweisen. Die Analyse von fünf kommerziellen CXS-Produkten verschiedener Hersteller ergab, dass drei Produkte nicht alle vorgeschriebenen Komponenten enthielten. Die beiden Produkte, die zuvor nach ChVPh untersucht worden waren (Bahn 13 und 14), bestanden jetzt den Identifikationstest nicht mehr, da die für *Aucklandia* spezifische Zone im Chromatogramm fehlte (siehe Pfeil und ovale Markierung). Ein weiteres Produkt enthielt die präparierte *Atractylodes*-Droge anstelle der unbehandelten. Die anderen zwei Produkte bestanden den Identifikationstest.

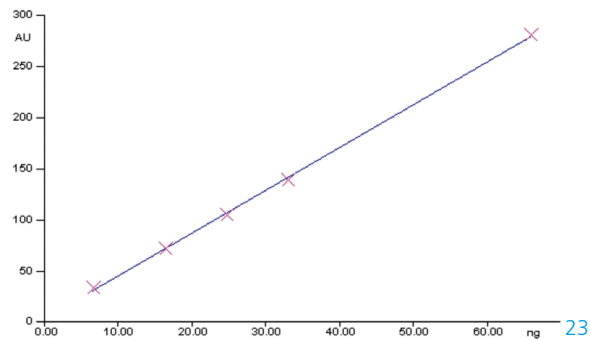
Für die quantitative Analyse von CXS wurde das Alkaloid Berberin ausgewählt, da dies ein



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HPTLC-Analyse von CXS. Bahn 1: *Coptis chinensis*; 2: *Coptis teeta*; 3: *Coptis deltoidea*; 4: Berberin HCl; 5: *Atractylodes chinensis*; 6: *Atractylodes lancea*; 7: *Atractylodes rhizoma* (präpariert); 8: *Aucklandia lappa*; 9: *Vladimiria radix*; 10: *Dehydrocostuslactone*; 11: *Costunolide*; 12: Mischung von Referenzdrogen; 13, 14: CXS-a (Hersteller A, 2 Chargen); 15: CXS-b (Hersteller B); 16, 17: CXS-c (Hersteller C, 2 Chargen).

gut nachweisbarer Inhaltsstoff von *Coptis rhizoma*, dem Hauptbestandteil von CXS ist. Nach ChVPh darf der Berberin HCl-Gehalt von *Coptis rhizoma* nicht unter 3.6 % liegen, das entspricht einem minimalen Gehalt von 0.98 % Berberin im Endprodukt CXS. Der Arbeitsbereich für die densitometrischen Bestimmung lag im Fluoreszenzmodus (366/>400 nm) zwischen 10 und 1500 ng. Die zugrunde liegende Michaelis-Menten-Funktion I wies eine relative Standardabweichung (%RSD) von 2.15 % auf. In einem engeren Arbeitsbereich von 6–60 ng war die Kalibrierfunktion linear ( $r = 0.99970$ ,  $RSD = 2.15\%$  Peakhöhe und  $r = 0.99959$ ,  $RSD = 2.34\%$  Peakfläche).



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Lineare Kalibrierfunktion (Peakhöhe) von Berberin HCl

Die Wiederfindungsrate bei einer mit Berberin aufgestockten Probe des Pflanzenmaterials lag zwischen 95.4 und 97.3 %. Die Wiederholpräzision betrug 3.9 % ( $n = 3$ ). Die beiden positiv identifizierten CXS-Produkte wiesen einen Berberin-Gehalt von 2.4 % (Produkt CXS-b, Bahn 15) bzw. 1.4 % (Produkt CXS-c2, Bahn 17) auf und entsprachen damit der Anforderung nach einem minimalen Gehalt von 0.98 %.

Weitere Informationen erhalten Sie auf Anfrage von den Autoren.

[1] Z. Li, I. Merfort, E. Reich, J AOAC 93 (2010) 1390

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# CAMAG TLC Scanner 4



## Der CAMAG TLC Scanner 4 in Verbindung mit winCATS Software ist der fortschrittlichste Messplatz zur densitometrischen Auswertung von Dünnschicht-Chromatogrammen

Die überaus effiziente und komfortable Software steuert alle Funktionen des Scanners und bearbeitet die Messdaten bis zum Endresultat.

*Der TLC Scanner 4 mit winCATS entspricht den Anforderungen von GMP/GLP und kann IQ/OQ qualifiziert werden.*

Ausser der Quantitativen Chromatogramm-Auswertung mit allen praxisrelevanten Funktionen einschliesslich Scanner-Selbsttest und 21 CFR Part 11 »compliance ready« ermöglicht winCATS:

- Zweiwellenlängen-Scan zur Untergrund-Korrektur Matrix- und/oder Chromatographie-bedingter Schicht-Unregelmässigkeiten
- Mehrwellenlängen-Scan zur quantitativen Bestimmung auch sehr unterschiedlich absorbierender Substanzen in einem einzigen Messablauf
- Bahnoptimierung zur korrekten Auswertung auch nicht ideal gelaufener Chromatogramme
- Spektrenbibliothek, vom Nutzer selbst angelegt zur Unterstützung von Identitätsprüfungen und zum schnellen Screening

Weitere Informationen unter: [www.camag.com/tlc-scanner](http://www.camag.com/tlc-scanner)

# CAMAG

Weltweit führend in der  
Planar-Chromatographie