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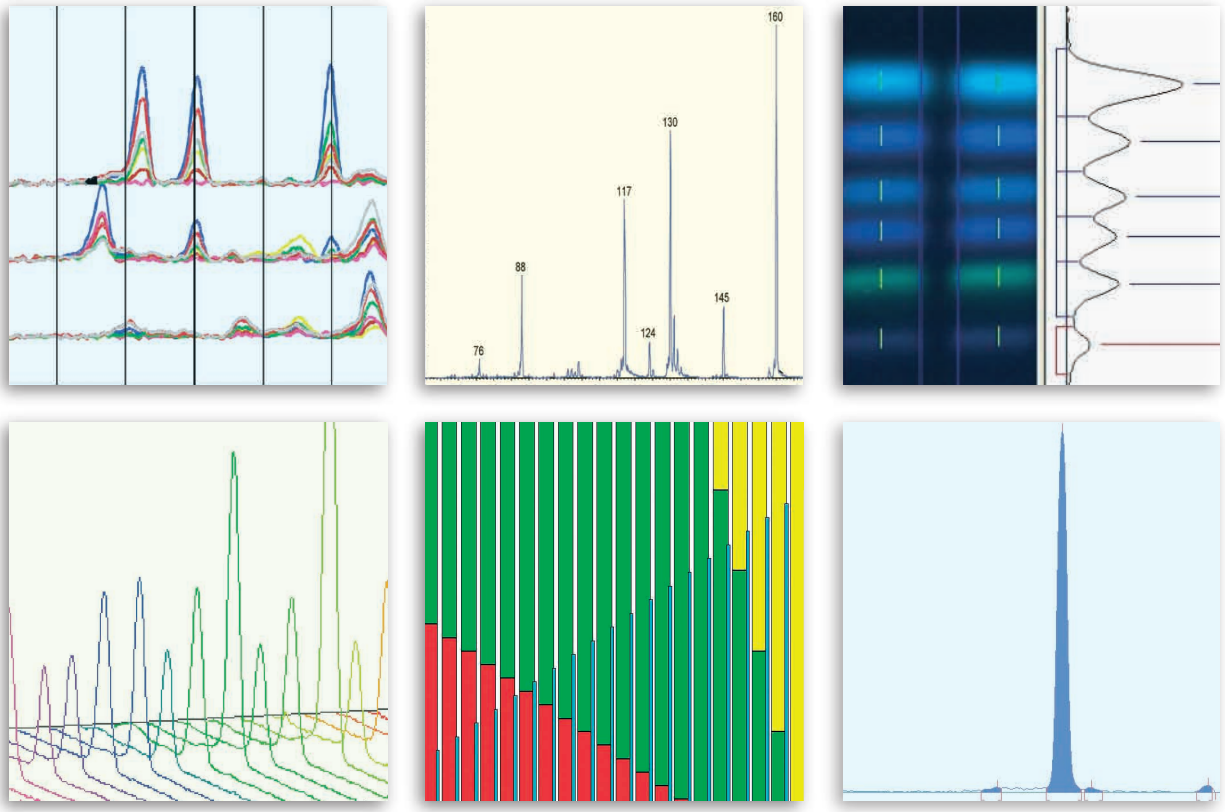

Planar-Chromatographie – ein wichtiger Baustein der modernen Analytik

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Planar-Chromatographie
Herausgegeben von Gerda Morlock
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In dieser Ausgabe hervorgehobene Produkte und Dienstleistungen

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CAMAG (Schweiz)
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Tel. +41 61 467 34 34 • Fax +41 61 461 07 02
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CAMAG (Deutschland)
Bismarckstr. 27–29 • D-12169 Berlin
Tel. +49 30 516 55 50 • Fax +49 30 795 70 73
info@camag-berlin.de

www.camag.com

Bestimmung von Aminopropanol in dermatologischen Produkten

1

▲ *Bayer Santé Familiale, Gaillard, Frankreich*

2

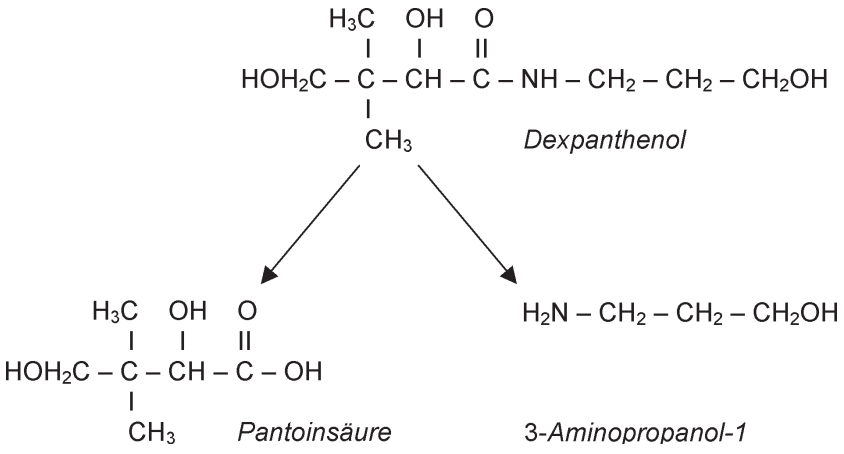
▲ *Caroline Petitti, Chemikerin in der analytischen Entwicklung*

Das analytische Entwicklungslabor von Bayer Consumer Care International Technical Center in Gaillard, Frankreich, entwickelt analytische Methoden für rezeptfreie Arzneimittel. Die meisten Analysen werden mit der HPLC, AAS (Atomabsorptions-Spektroskopie), ICP-OES (Optische Emissionsspektrometrie mit induktiv gekoppeltem Plasma) und Potentiometrie durchgeführt und einige mit UV- oder IR-Spektroskopie, GC und HPTLC.

Aminopropanol wird üblicherweise mit der HPLC analysiert, jedoch ist diese Methode sehr zeitaufwendig. Deshalb wurde die Methode durch eine einfache, schnelle und genaue HPTLC-Methode bei Bayer Consumer Care ersetzt. Die Validierungsdaten belegen die sehr gute Eignung (Korrelation der Kalibrierfunktion 0.9979, mittlere Wiederfindung 102% ±4.9%, Laborpräzision ±5.7%) und erlauben dabei einen um den Faktor 3 höheren Probendurchsatz mit deutlicher Reduzierung der Arbeitszeit.

Einleitung

Aus Dexpanthenol (Provitamin B5), einem der Wirkstoffe in den dermatologischen Bayer-Produkten Bepanthen® und Bepanthol®, entstehen durch hydrolytische Spaltung Pantoinsäure und Aminopropanol gemäss folgendem Schema. Über die Bestimmung von Aminopropanol ist eine Stabilitätskontrolle des Dexpanthenols möglich. Hierbei können höhere Probenzahlen anfallen.



Probenvorbereitung

Creme oder Salbe wurden in Ethanol gelöst und erhitzt. Unlösliche Bestandteile werden durch Zentrifugieren für 5 min bei 10 000 rpm abgetrennt.

Standardlösung

Aminopropanol wurde für eine 5-Punkte-Kalibration zwischen 12.5 µg/mL und 200 µg/mL in Ethanol gelöst.

Anmerkung des Herausgebers: Wenn die Sprühtechnik genutzt wird, können besser unterschiedliche Mengen (Volumina) einer Kalibrierlösung aufgetragen werden.

Schicht

HPTLC-Platte Kieselgel 60 F_{254} (Merck), vorgewaschen mit Methanol (durch Entwicklung) und getrocknet bei 105 °C für 30 min auf dem DC-Plattenheizer III.

Probenauftragung

Als 6 mm Bänder mit dem DC Probenautomat, max. 23 Bahnen, Bahnabstand mind. 7.8 mm, Abstand vom unteren Plattenrand 8 mm, Abstand von den Seiten mind. 14 mm, Auftragevolumen 2 µL für jede Kalibrierlösung und Probelösung.

Chromatographie

Dem Fliessmittel aus Ethanol – Wasser – Eisessig 16:3:1 (v/v/v) wurde das Derivatisierungsreagenz hinzu gefügt (0.5 g Ninhydrin pro 100 mL Fliessmittel). Die Chromatographie wurde in der Horizontalentwicklungskammer (HDC) von beiden Seiten bis zu einer Laufstrecke von 40 mm (von der Plattenkante) durchgeführt.

Derivatisierung

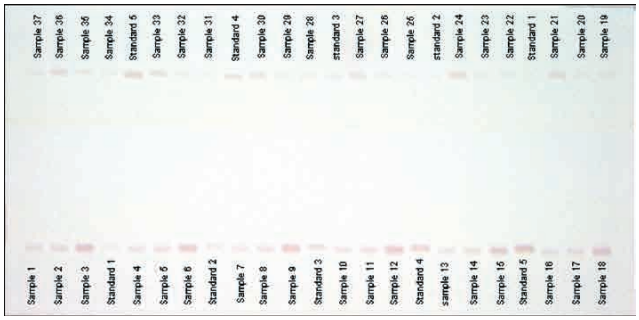
Die Platte wurde bei 105 °C für 5 min auf dem DC-Plattenheizer III erhitzt. Aminopropanol war als hellviolette Zone bei hR_F 50 sichtbar.

Densitometrische Auswertung

Absorptionsmessung bei 486 nm mit dem TLC-Scanner 3 und winCATS Software; Auswertung über die Peakfläche mittels Michaelis Menten 2-Regression.

Dokumentation

Mit dem DigiStore 2-System (Reflektion) im Weisslicht (Auflicht).



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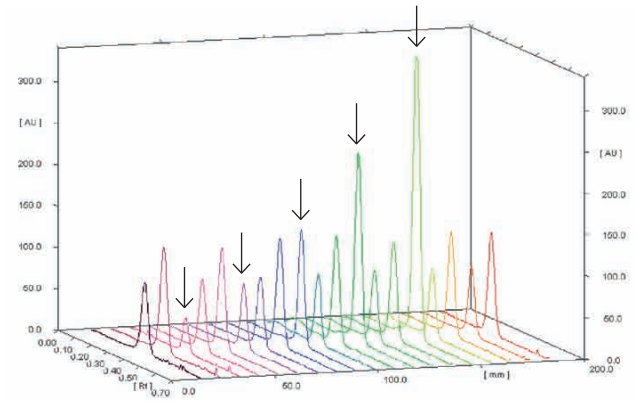
▲ *Beispiel einer HPTLC-Platte: Die hellviolette Aminopropanol-Zone ist bei hR_F 50 sichtbar; entsprechende Proben- und Standardbahnen wurden von beiden Seiten in der HDC entwickelt und zur Derivatisierung nur erhitzt.*



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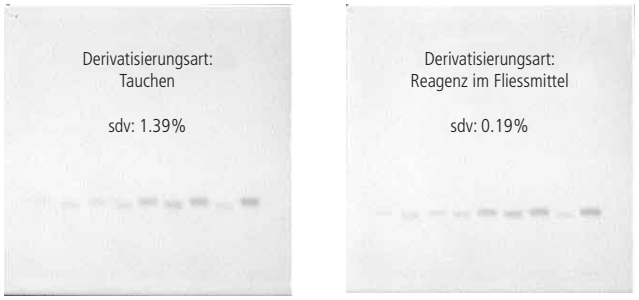
▲ *3D-Grafik im Nahbereich des Aminopropanol-Peaks von allen Bahnen (pro Plattenseite), gemessen bei 486 nm (5-Punkte-Kalibration ist markiert)*

Ergebnisse und Diskussion

Es wurde eine schnelle, alternative Bestimmung zur HPLC-Methode entwickelt. Die HPTLC-Methode ist einfacher und schneller, da bis zu 36 Bahnen simultan entwickelt werden. Ein weiterer grosser Vorteil ist die Leichtigkeit, mit der die Derivatisierung durchgeführt werden kann: Dem Fliessmittel wurde das Derivatisierungsreagenz Ninhydrin zugesetzt, welches gleichzeitig mit der Chromatographie homogen auf die Platte aufgebracht wurde. Durch diese Art der Reagenzaufbringung wurde auch die relative Standardabweichung der Kalibrierfunktion (sdv ±0.19 %) verbessert im Vergleich zu der bereits sehr guten Präzision, die man durch das automatische Tauchen erreicht (sdv ±1.39 %).



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▲ *Vergleich der Derivatisierungsarten: Die relative Standardabweichung der Kalibrierfunktion (sdv) wurde durch die homogene Reagenzaufbringung während der Entwicklung von sdv ±1.39 % auf sdv ±0.19 % verbessert.*

Für die Analyse von 30 Proben benötigte die HPLC-Methode 3 Tage, während die HPTLC-Methode nur 1 Tag beanspruchte. Dadurch konnte mit der HPTLC der Analysen-Durchsatz um den Faktor 3 verbessert werden.

Verwendete Methode	HPLC	HPTLC
Zeitdauer für 30 Proben	3 Tage	1 Tag

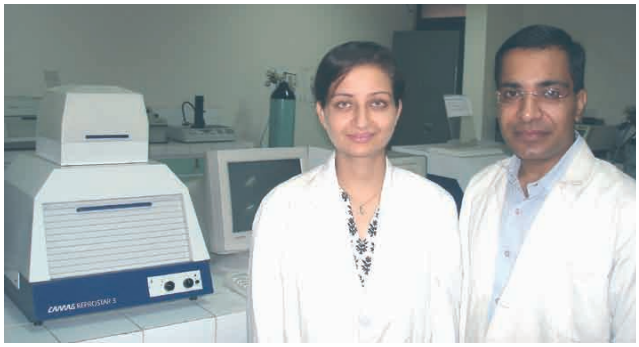
Die sehr gute Eignung der HPTLC für diese Analyse zeigen die Validierungsdaten. Die Detektions- und Bestimmungsgrenze (LOD und LOQ) waren 4.5 µg/mL bzw. 15 µg/mL, bezogen auf ein Auftragevolumen von 2 µL (falls notwendig, kann die LOD und LOQ verbessert werden durch das Auftragen grösserer Volumina). Der Korrelationskoeffizient der Kalibrierfunktion war 0.9979. Die Genauigkeit wurde durch die mittlere Wiederfindung von 102 % (n = 15) belegt. Die mittlere Wiederholbarkeit wurde mit ±4.9 % über 5 unterschiedliche Konzentrationen (n = 3 pro Kalibrierniveau) bestimmt. Die relative Standardabweichung der Vergleichspräzision über mehrere Tage (Laborpräzision, *RSD*, n = 9) war ±5.7 %.

Validierungsparameter	Ergebnisse
LOD LOQ *(beide bezogen auf ein Auftragevolumen von 2 µL)*	4.5 µg/mL 15 µg/mL
Linearität *(Korrelationskoeffizient)*	0.9979
Mittlere Wiederfindung	102 %
Mittlere Wiederholbarkeit (*RSD*) *(bestimmt über 5 unterschiedliche Konzentrationen, n= 3 pro Kalibrierniveau)*	±4.9 %
Laborpräzision (*RSD*, n= 9)	±5.7 %

Weitere Informationen sind auf Anfrage von der Autorin erhältlich.

*Caroline Petitti, Bayer Santé Familiale, 33 rue de l'industrie, F-74 240 Gaillard, Frankreich, caroline.petitti@bayerhealthcare.com

Stabilitätstest von Gatifloxacin und Bestimmung in polymeren Nanopartikeln



6

▲ *Shruti Chopra und Sanjay Motwani*

In CBS 97 stellten wir die HPTLC-Gruppe der Pharmazeutischen Fakultät von Jamia Hamdard in New Delhi vor, die sich intensiv mit der Entwicklung und Validierung von HPTLC-Methoden zur Analytik von Wirkstoffen und Biomarkern aus pflanzlichen Produkten beschäftigt. Die pharmazeutische Industrie benötigt schnelle, zuverlässige und zugleich ökonomische Methoden, um den Gehalt pflanzlicher Inhaltsstoffe in den unterschiedlichen pharmazeutischen Formulierungen sicherzustellen. Daher wird die Entwicklung von Hochdurchsatz-Methoden geschätzt, wobei die HPTLC nebst anderen Methoden zur Qualitätskontrolle unter GMP-Bedingungen perfekt geeignet ist.

In Indien gehört die HPTLC zu den routinemässig eingesetzten analytischen Methoden in vielen Produktentwicklungs- und analytischen Laboratorien. Die HPTLC-Methode ist für die Analytik von pharmazeutischen Darreichungsformen ökonomischer als andere chromatographische Verfahren, da sie weniger Lösungsmittel verbraucht und minimale Anforderungen an die Probenvorbereitung stellt. Viele Proben können zeitgleich in kürzester Zeit mit automatisierten Systemen analysiert werden.

In dieser Arbeit entwickelte und validierte die Gruppe von Sanjay Motwani* gemäss den ICH-Richtlinien eine präzise und robuste HPTLC-Methode [1] für Gatifloxacin-Stabilitätstests und dessen Quantifizierung in polymeren Nanopartikeln.

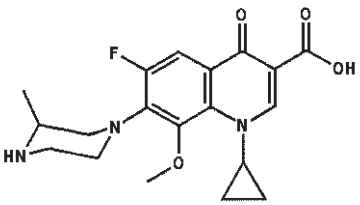
Einleitung

Gatifloxacin ist ein neues Fluorchinolon-Antibiotikum, das in USA weit verbreitet eingesetzt wird, in Europa jedoch beschränkt zugelassen ist. Es richtet sich mit einem breiten Wirkungsspektrum gegen grampositive, gramnegative, aerobe und atypische Erreger von Infektionen der Harn- oder Atemwege, indem es die Gyrase und Topoisomerase IV der bakteriellen DNA hemmt.

Der Leitfaden zu Stabilitätstests von neuen Arzneimitteln und deren Wirkstoffen Q1A (R2) der International Conference on Harmonization (ICH) legt Wert auf die Durchführung von Stresstests zu einem neuen Wirkstoff, um dessen Stabilitätseigenschaften aufzuklären. Zu den geforderten Tests gehören die Stabilität gegen Sauerstoff sowie Säure- und Base-katalysierte hydrolytische und photolytische Stabilitätsuntersuchungen. Eine ideale Methode für Stabilitätstests sollte die Quantifizierung der Abbauprodukte ermöglichen. Als analytische Methoden für die Bestimmung von Gatifloxacin werden bisher die Fluorimetrie, HPLC, HPLC/ESI-MS(-MS) und HPTLC verwendet. Da es bis dato keine HPTLC-Methode für Stabilitätstests von Gatifloxacin und zu dessen Bestimmung aus polymeren Nanopartikeln gibt, wurde nachfolgend eine Methode entwickelt, die diese Anforderungen vollständig erfüllt.

Probenvorbereitung

Gatifloxacin-Bulkware wurde in Methanol gelöst (1 mg/mL). Polybutylcyanoacrylat-Nanopartikel (Gehaltsangabe: 2.48 mg Gatifloxacin in 100 mg Nanopartikel), entsprechend 5.5 mg Gatifloxacin, wurden eingewogen und in einen 10 mL-Messkolben mit 5 mL Aceton überführt. Um eine vollständige Extraktion sicherzustellen wurde für 30 min im Ultraschallbad beschallt, auf 10 mL mit Aceton aufgefüllt und für 5 min bei 2000 rpm zentrifugiert. Der filtrierte Überstand wurde analysiert.



▲ *Chemische Strukturformel von Gatifloxacin [1-Cyclopropyl-6-fluor-1,4 dihydro-8-methoxy-7-(3-methyl-1-piperazinyl)-4-oxo-3-chinolin-carbonsäure]*

Standardlösung

Zur Bestimmung der funktionellen Korrelation wurde Gatifloxacin in Methanol gelöst (100 µg/mL). Für die Analytik der Nebenkomponenten wurde Gatifloxacin in Methanol gelöst (1 mg/mL) und 1:5 mit Methanol verdünnt (200 µg/mL).

Schicht

HPTLC-Alufolien Kieselgel 60 F_{254} (Merck) 20 × 10 cm, vorgewaschen mit Methanol (durch Entwicklung) und für 30 min bei 90 °C getrocknet

Probenauftragung

Bandförmig mit Linomat V, Bandlänge 6 mm, Auftragevolumen 2 µL für Proben und 4–12 µL für die Standardlösung (400–1200 ng/Band), für die Nebenkomponenten-Bestimmung 3 µL für Proben und 2 µL für die Standardlösung, Bahnabstand 10 mm, unterer Randabstand 10 mm, seitlicher Randabstand mind. 15 mm

Chromatographie

In der Doppeltrogkammer mit n-Propanol – Methanol – Ammoniak (25 %) 5:1:0.9 (v/v/v) nach 30 min Kammersättigung mit der mobilen Phase, Laufstrecke 80 mm vom unteren Plattenrand. Die Platten werden nach der Chromatographie 1 min im Warmluftstrom getrocknet.

Anmerkung des Herausgebers: Aufgrund der zunehmenden Diffusion der Zonen sollten Laufstrecken vom unteren Plattenrand bis max. 70 mm gewählt werden.

Densitometrische Auswertung

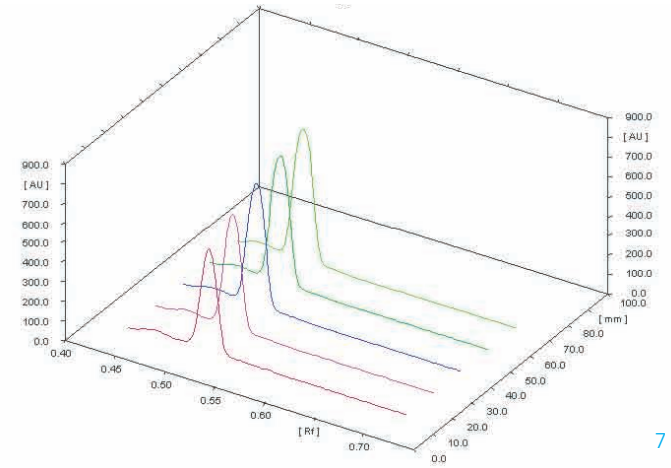
Absorptionsmessung bei 292 nm mit dem TLC-Scanner 3 und winCATS-Software, lineare Kalibration über die Peakfläche.

Ergebnisse und Diskussion

Der Ammoniakgehalt im Fliessmittel war ausschlaggebend, um einen scharfen und symmetrischen Gatifloxacin-Peak bei hR_F 60 ± 2 zu erhalten. Die Regressionsanalyse (Mittelwert von 3 Kalibrierkurven) zeigte einen guten linearen Zusammenhang mit einem Korrelationskoeffizienten von 0.9954 in einem Konzentrationsbereich von 400–1200 ng/Band.

Kalibrationsdaten im Bereich von 400–1200 ng/Band ($n = 3$)

Korrelationskoeffizient (r ± SD)	0.9954 ± 0.0012
Steigung ± SD	9.6638 ± 0.0491
Vertrauensbereich (95 %) der Steigung	9.516 - 9.760
y-Achsen-Schnittpunkt ± SD	956.3310 ± 27.6714
Vertrauensbereich (95 %) des y-Achsen-Schnittpunktes	936.26 - 987.90



▲ *3-D Grafik der Kalibrierstandard-Bahnen im Bereich von 400–1200 ng/Band, Absorptionsmessung bei 292 nm*

Die Wiederholbarkeit der Peakflächenmessung ($n = 6$) von 3 verschiedenen Konzentrationen (600, 800 und 1000 ng/Band) zeigte eine sehr gute Präzision der Methode, d.h. geringe Standardfehler SE < ±0.8 und geringe relative Standardabweichungen *RSD* < ±0.03 % innerhalb eines Tages sowie SE < ±1.0 und *RSD* < ±0.03 % zwischen mehreren Tagen. Nach kleinen absichtlichen Änderungen bei kritischen Einflussgrössen im Verfahren zeigte die entwickelte HPTLC-Methode noch immer geringe *RSD* ($n = 3$, 800 ng/Band) von < ±0.03 % und SE von ≤ ±0.8 und somit eine gute Robustheit. Die Detektions- bzw. Bestimmungsgrenze von 2.7 ng (S/N 3) bzw. 8.3 ng (S/N 10) war für die Analytik sehr gut geeignet.

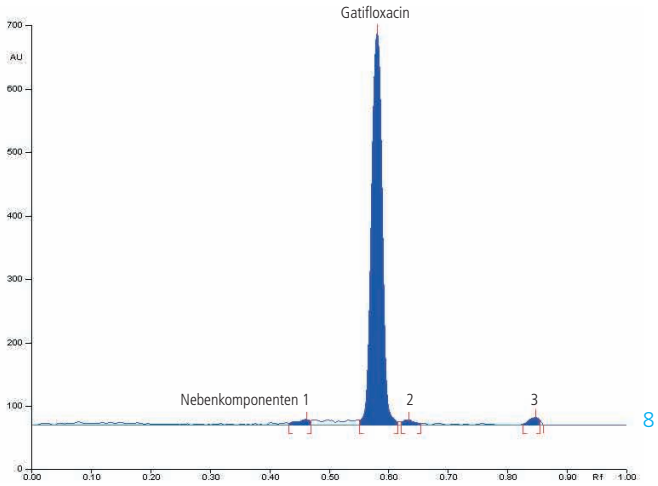
Zur Bestimmung der Wiederfindungsrate wurden 3 unterschiedliche Mengen Gatifloxacin entsprechend 50, 100 und 150 % des Gatifloxacin-Gehaltes in polymeren Nanopartikeln dotiert. Auf jedem Niveau wurde eine 6-fach-Bestimmung durchgeführt. Die Wiederfindungsraten nach Extraktion der polymeren Nanopartikel waren im Bereich von 99.2 und 101.9 %.

Zudem war die HPTLC-Methode perfekt geeignet, um die Einflussgrössen unter unterschiedlichen Stressbedingungen gemäss dem ICH-Leitfaden zu untersuchen. Für potentielle Abbauprodukte waren keine Standards erhältlich. Als Indikator für die Gatifloxacin-Stabilität wurde die Wiederfindungsrate (WFR) herangezogen. Aufgrund der sehr stark reduzierten WFR bei Säureeinwirkung und der leicht reduzierten WFR unter feuchter Hitzeeinwirkung, kann auf 2 Einflussgrössen geschlossen werden. Bei den restlichen 5 Stressbedingungen lag die WFR bei 99 %.

Nr.	Beeinflussung durch	Zeit (h)	hR_F-Wert der Abbauprodukte	Wiederfindungsrate (%)
1	Säure, 5 M HCl, im Rückfluss	3	1, 3, 51, 75	46.1
2	Base, 5 M NaOH, im Rückfluss	3	1	98.9
3	H_2O_2 (30%, v/v), im Rückfluss	3	Keine detektiert	99.2
4	Trockene Hitze von 100 °C	8	54	98.5
5	Feuchte Hitze von 100 °C	3	52	97.7
6	Taglicht – Fotostabilität	24	Keine detektiert	99.3
7	UV 254 nm	8	Keine detektiert	99.0

Die Identität des Gatifloxacin-Peaks durch Vergleich der Probe- und Standardspektren zeigte eine gute Korrelation von 0.9996. Die Peakreinheit durch Spektrenvergleich innerhalb eines Peaks an verschiedenen Positionen (Peakstart, Peakmitte, Peakende) zeigte keine Überlagerung durch Hilfsstoffe und andere aktive Komponenten, die in der polymeren Nanopartikel-Formulierung vorhanden sind.

Bei der Nebenkomponentenbestimmung zeigte die Bulkware (aufgetragen wurden 3000 ng Gatifloxacin) drei zusätzliche Peaks bei hRF 46 ± 2, 63 ± 1 und 84 ± 2. Jedoch waren die Peakflächen der zusätzlichen Banden (Peakfläche von insgesamt 359 AU) signifikant kleiner als die der Gatifloxacin-Standardlösung (400 ng/Band, Peakfläche 9006 AU) und somit < 0.1 %.



▲ *Chromatogram der Nebenkomponentenbestimmung von Gatifloxacin (3 µg/Band), potentielle Nebenkomponenten bei hR_F 46, 63 und 84 sind zusammen unter 0.1 %*

Die entwickelte und validierte HPTLC-Methode zeigte sich als genaue, präzise, spezifische und wiederholbare Methode zur Gatifloxacin-Bestimmung als Bulkware und in pharmazeutischen Formulierungen.

Weitere Informationen sind von den Autoren auf Anfrage erhältlich.

[1] S. K. Motwani, R. K. Khar, F. J. Ahmad, S. Chopra, K. Kohli, S. Talegaonkar, Z. Iqbal, Analytica Chimica Acta 576 (2006) 253–260

*Sanjay K. Motwani, Department of Pharmaceutics, Faculty of Pharmacy, Jamia Hamdard, Hamdard Nagar, New Delhi – 110 062, Indien, Tel.: +91-11-26059688, Fax: +91-11-26059663, sanjay_bcp@rediffmail.com

Wechsel in der Bereichsleitung Vertrieb & Marketing


9

Herr Erwin Malzacher, bei CAMAG seit November 1971, der diesen Bereich seit Ende 1994 erfolgreich geführt hat, äusserte den Wunsch, Mitte 2006 sein Arbeitspensum auf 60 % zu reduzieren. Da dieser Bereich für CAMAG äusserst wichtig ist, suchten wir rechtzeitig eine Nachfolgekraft, um sie entsprechend zu schulen und aufzubauen. Wir sind glücklich, in Frau Claudia Nachbur, die im November 2005 bei CAMAG eintrat, eine hoch motivierte und kompetente Mitarbeiterin gefunden zu haben, und wir haben ihr per 1.1.2007 die Leitung des Bereichs Vertrieb & Marketing übertragen. Herr Malzacher, der sich engagiert der Einarbeitung von Frau Nachbur auf CAMAG-spezifische Belange annahm, wird uns auch noch weiterhin mit seinen langjährigen Erfahrungen zur Verfügung stehen.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Frau Nachbur studierte von 1982 bis 87 an der ETH Zürich und schloss mit dem Diplom als Naturwissenschafterin ab. Vor ihrem Eintritt bei CAMAG erwarb sie sich bei zwei Schweizer Unternehmen langjährige Erfahrungen als Verkaufleiterin im internationalen Vertrieb von Investitionsgütern sowie Aufbau und Pflege eines weltweiten Vertriebsnetzes.

Schon im ersten Jahr ihrer Tätigkeit bei CAMAG hat Frau Nachbur bereits einige internationale Missionen selbständig und erfolgreich durchgeführt. Dazu gehörten ein Kundenseminar in Taschkent, Uzbekistan im März 2006, ihre Mitwirkung an der 9. Colacro in Merida, Mexico im Juni 2006 und ihre anschliessenden Besuche einiger Ländervertretungen im Mittelamerika.


11

◀ *Vortrag von Frau Nachbur beim Seminar am Institute of Chemistry of Natural Plants in Taschkent*


12

◀ *Teilnehmer und Dozenten am HPTLC Workshop der Colacro – ganz rechts Frau Nachbur*

Inzwischen hat Frau Nachbur die Leitung des Bereichs Vertrieb & Marketing fest im Griff. Sie unternahm diverse Reisen nach Osteuropa und ist dabei, den Ausbau und die Reorganisation unseres Vertreternetzes in Europa zu betreiben.

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Liebe Freunde

HPTLC-Symposia im internationalen Massstab haben eine lange Tradition. Das erste »International Symposium on Instrumental High Performance Thin-Layer Chromatography« fand 1979 in Bad Dürkheim statt mit Vortragenden aus 7 Nationen – damals ein beachtliche Internationalität! Das jüngste Symposium im Oktober letzten Jahres lockte über 150 Teilnehmer aus 22 Ländern mit Vortragenden aus 10 Nationen nach Berlin.

Eine erfreuliche Vielzahl jüngerer Teilnehmer sowohl aus dem universitären wie auch aus dem industriellen Bereich präsentierte beeindruckend über 50 Poster, die den Facettenreichtum der HPTLC widerspiegelten.

Nach einem Workshop gab es Präsentationen zu Grundlagen und neuen Aspekten, aber auch hervorragende Anwendungen in der Lebensmittelanalytik, Analytik von pflanzlichen Extrakten und klinischen Analytik. Den Abschluss bildeten innovative Entwicklungen in der HPTLC, Bioaktivitätstests, spezifische Immuno-Detektionen und Kopplungsmethoden. Die Internationalität der Beiträge in den 6 Themenblöcken verdeutlichte die weltweite Aktualität und zeigte das quo vadis der Methode auf.

Wünschen wir der Planar-Chromatographie auch in Zukunft solche Impulse. Für diejenigen, die nicht dabei sein konnten, finden sich Eindrücke unter **www.hptlc.com** – es lohnt sich, da reinzuschauen… übrigens der erste Beitrag in dieser CBS-Ausgabe wurde auf dem Symposium eindrücklich präsentiert. Erfreulich, dass im boomenden Gesundheitsbereich auf die HPTLC rückbesonnen wird.

Herzlichst Ihre

Gerda Morlock

Gerda Morlock
cbs@camag.com

Dear friends

HPTLC symposia on an international platform have a long tradition. The first "International Symposium on Instrumental High Performance Thin-Layer Chromatography" took place in Bad Dürkheim 1979 with scientific presentations from scientists from 7 nations all over the world – at that time a respectable international representation. The latest symposium in Berlin in October last year brought together over 150 scientists from 22 countries and lecturers from 10 nations.

Many young attendees from academia as well as from industry participated, demonstrating that planar chromatography is very much alive and over 50 posters were presented, reflecting a myriad of features of HPTLC.

Presentations were given to fundamentals and new aspects, but also outstanding applications in food analysis, herbal analysis and clinical analysis. The closure part was dedicated to innovative developments in HPTLC, bioactivity tests, specific immuno detection and coupling methods. The internationality of the presentations in the 6 sessions underlined the worldwide up-to-dateness and showed the "quo vadis" of the technique.

We wish planar chromatography more such impulses in future. For those, who could not attend, please go to **www.hptlc.com** for some meeting impressions and hopeful enjoyment. By the way the first CBS article of this issue was impressively presented at the symposium. Nice to hear that in the booming health care business, HPTLC analysis is again being used.

Sincerely,

Gerda Morlock

Gerda Morlock
cbs@camag.com

CAMAG

MÄRZ
MARCH
2007

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THE CBS CLASSIFICATION SYSTEM

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
    f) Clinico-chemical applications and profiling body fluids
    g) Herbal and traditional medicines
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**

XX. (abstract number underlined) refers to HPTLC related publication or application using HPTLC materials

1.Reviews and books

98 004 J. SHERMA (Dept. of Chem., Lafayette Col., Easton, Pennsylvania 18042): Planar chromatography. Anal. Chem. 74, 2653-2662 (2002). This reviews covers the literature of TLC and HPTLC found by computer-assisted searching in Chemical Abstracts and the ICI Web of Science from November 1, 1999 to November 1, 2001. The literature search was augmented by consulting Analytical Abstracts, Chemical Titles and Current Contents, and the following journals were searched directly: J. Chrom. (parts A and B and the bibliography issues), J. Chrom. Science, Chromatographia, Anal. Chem., J. Liq. Chrom. & Rel. Technol., J. AOAC Int., J. Planar Chromatogr. - Modern TLC and Acta Chrom. Publications in the past 2 years on the history, theory, methodology, instrumentation, and applications of TLC are discussed. 198 references are listed.

review 1

98 005 J. SHERMA (Dept. of Chem., Lafayette Col., Easton, Pennsylvania 18042): Planar chromatography. Anal. Chem. 72, 9R-25R (2000). Selective review of the literature of TLC and HPTLC in Chemical Abstracts from November 1, 1997 to November 1, 1999. The literature search was augmented by consulting Analytical Abstracts, Chemical Titles and Current Contents, and the following journals were searched directly: J. Chrom. (parts A and B and the bibliography issues), J. Chrom. Science, Chromatographia, Anal. Chem., J. Liq. Chrom. & Rel. Technol., J. AOAC Int., J. Planar Chromatogr. - Modern TLC and Acta Chrom. The different chapters are: history, student experiments, books and reviews; theory and fundamental studies; chromatographic systems (stationary and mobile phases); apparatus and techniques; detection and identification of separated zones; quantitative analysis; preparative layer chromatography, thin-layer radio-chromatography and applications of TLC/HPTLC. 415 references from the last 2 years.

review 1

98 006 J. SHERMA (Dept. of Chem., Lafayette College, Easton, Pennsylvania 18042): Planar chromatography. Anal. Chem. 78, 3841-3852 (2006). TLC and HPTLC literature found by computer-assisted searching in Chemical Abstracts and the ISI Web of Science (11.2003 - 11.2005) is covered. The literature search was augmented by consulting Analytical Abstracts, and the following journals were searched directly: J. Chrom. (parts A and B and the bibliography issues), J. Chrom. Science, Chromatographia, Anal. Chem., J. Liq. Chrom. & Rel. Technol., J. AOAC Int., J. Planar Chromatogr. - Modern TLC and Acta Chrom. (Papers reporting research on paper chromatography were not included.) 205 references were cited from the last 2 years. The different chapters are: history, student experiments, books and reviews; theory and fundamental studies; chromatographic systems (stationary and mobile phases); apparatus and techniques; detection and identification of separated zones; quantitative analysis; preparative layer chromatography and thin-layer radiochromatography.

review 1

2. Fundamentals, theory and general

98 007 J. GILBERT*, E. ANKLAM (*Department for Environment, Food and Rural Affairs, Central Science Laboratory, Sand Hutton, York, UK, j.gilbert@csl.gov.uk): Validation of analytical methods for determining mycotoxins in foodstuffs. TrAC 21, 468-486 (2002). The article describes the valuable lessons learned from EU while funding a method-validation project (1996-2000) to meet European mycotoxin control in foodstuffs. It shows the performance characteristics of validated and official methods for aflatoxins, the selection and development of methods for validation, and the preparation of naturally contaminated mycotoxin test materials for validation studies. The authors put special emphasis on validation of TLC methods for mycotoxins for developing countries, as the main exporters to Europe of food and food products.

food analysis, toxicology, HPTLC, review 2f

98 008 K. FODOR*, Z. VEGH, B. RENGER (*Gedeon Richter Ltd., P.O.B. 27, H-1475 Budapest, Hungary, k.fodor@richter.hu): Thin-layer chromatography in testing the purity of pharmaceuticals. TrAC 25, 8 (2006). In the individual monographs of drug substances or finished products, only semi-quantitative TLC purity tests are mentioned and the number of TLC applications is steadily decreasing, being replaced by HPLC methods that are considered more appropriate. However, to comply with the latest and current pharmaceutical regulations, TLC manufacturers do not stop developing new equipment and accessories related to sample application, developing chambers, derivatization, documentation, and quantitative evaluation. Numerous examples of TLC/HPTLC applications in analytical research and quality control are mentioned to show the validity of this technique in the description of organic related impurities in drug substances and final products. Finally, authors ask analysts to present excellent, fully-validated and documented GMP/GLP-compliant TLC purity-test procedures to convince experts from pharmacopoeial committees and regulatory bodies of the importance of this analytical tool.

quality control, comparison of methods 2a

98 009 J. K. ROZYLO*, M. JANICKA, R. SIEMBIDA (*Fac. of Chem., Maria Curie-Sklodowska Univ., 20031 Lublin, Poland): Different liquid chromatographic techniques in the study of the process of adsorption chromatography. Acta Chrom. 6, 21-38 (1996). The possibility of using a thermodynamic approach for optimization of mixed mobile phases for systems with binary and ternary solvents is discussed, as well as the method for predicting separation results in column liquid chromatography on the basis of experimental thin-layer chromatographic data. The effect of mobile phase composition on solute retention is described by solute capacity factors (characteristic of the pure solvents), molecular interactions occurring in the bulk phase and adsorption equilibrium in a given adsorbent - binary solution system. Two kinds of chambers were used in the investigation: a saturated Stahl chamber and a sandwich chamber. The study described the use of different TLC techniques as pilot methods for HPLC.

binary/ternary mobile phase, optimization of mobile phase composition 2a

98 010 W. PRUS (Fac. of Textile Engineering and Environmental Protection, Univ. of Bielsko-Biala, 2 Wilowa Street, Bielsko-Biala, Poland): Relationship between raman scattering intensity in the aromatic region and the thermally induced increase in the UV absorption of chemically bonded stationary phases. Acta Chrom. 16, 204-215 (2006). Discussion of the relationship between the thermally induced increase in UV absorption of RP-8 and RP-18 layers and Raman-scattering intensity in the aromatic region when the stationary phases are irradiated with a high-power neodymium laser. The results obtained confirm the assumption of a linear relationship between the growth of UV absorption in this region by the stationary phase as a result of its thermal modification and the density of coverage of the silica gel with alkyl (octyl or octadecyl) ligands capable of aromatization.

UV absorption, Raman scattering 2a

98 011 C. SARBU*, B. TIPERCIUC (*Babes-Bolyai University, Faculty of Chemistry and Chemical Engineering, Arany Janos 11, 400028 Cluj-Napoca, Romania): Modeling, by multivariate regression methods, of the chromatographic retention (lipophilicity) of new oxadiazoline derivatives. J. Planar Chromatogr. 19, 342-347 (2006). HPTLC of 13 oxadiazoline derivatives on RP-18 with methanol - water mixtures containing from 25 to 75 % methanol in steps of 10 %. After development the dried plates were examined under UV light at 254 nm. The retention indices predicted were satisfactory and in very good agreement with the molecular structure of the compounds investigated.

HPTLC 2c

98 012 B. SPANGENBERG (University of Applied Sciences Offenburg, Badstrasse 24, 77625 Offenburg, Germany): Does the Kubelka-Munk theory describe TLC evaluations correctly? J. Planar Chromatogr. 19, 332-341 (2006). In TLC the development step distributes the sample throughout the layer. The essential reqirement for quantitative TLC is a constant sample distribution in each sample spot. The paper shows that quantitative TLC is possible even if the concentration of the sample is not constant. In the absence of uniform sample distribution classical Kubelka-Munk theory must be extended. The extended theory presented is not only capable of describing asymmetrical scattering in TLC layers but also includes a formula for absorption and fluorescence in diode-array TLC. With this new formula all different formulas for diode-array thin-layer chromatographic evaluation are combined in one expression.

2b

3. General techniques

98 013 C. B. FANG (Congbing Fang), X. C. WAN* (Xiaochun Wan), C. J. JIANG (Changjun Jiang), H. R. TAN (Huarong Tan), Y. H. HU (Yinghui Hu), H. Q. CAO (Haiqun Cao) (*Key Laboratory of Tea Biochemistry and Biotechnology, Ministry of Education and Ministry of Agriculture, Anhui Agricultural University, Hefei 230036, China): Comparison of HPTLC, HPLC, and HPCE for fingerprinting of Pueraria Radix. J. Planar Chromatogr. 19, 348-354 (2006). HPTLC of puerarin, daidzein, daidzin, and 3'-methoxypuerarin on silica gel, pre-washed with methanol, in an unsaturated twin-trough chamber with chloroform - methanol - ethyl acetate - water 16.2:18.8:52:3. Quantitative determination by absorbance measurement at 254 nm. The relative standard deviation of Rf values, retention times and peak area percentages all meet the national standards.

traditional medicine, herbal, HPTLC, quantitative analysis, densitometry 3e

98 014 G. GRYGIERCZYK (Inst. of Chem., Silesian Univ., 9 Szkolna Street, 40-006 Katowice, Poland): Chromatographic analysis of organic compounds on impregnated chemically bonded stationary phases. Part I. Acta Chrom. 17, 302-313 (2006). Non-polar (RP-2, RP-8, and RP-18) and polar (amino, cyano, and diol) chemically bonded stationary phases have been impregnated with solutions of organic substances at different concentrations and the effect of impregnation on the mechanism of retention of alcohols, higher fatty acids, amino acids, and medicines has been investigated.

comparison of methods, impregnation, retention behaviour 3b

98 015 T. HALKINA, J. SHERMA* (*Dept. of Chem., Lafayette Col., Easton, PA 18042-1782, USA): Comprative evaluation of the performance of silica gel TLC plates and irregular and spherical-particle HPTLC plates. Acta Chrom. 17, 261-271 (2006). TLC and HPTLC plates have been compared on the basis of theoretical plate number, resolution, linearity, development time, and limit of sensitivity for analysis of a multicomponent analgesic tablet in the fluorescence quenching mode and analysis of a five-component dye mixture in the visible mode.

HPTLC, comparison of methods, irregular and spherical HPTLC plates 3b

98 016 T. HALKINA, J. SHERMA* (*Dept. of Chem., Lafayette Col., Easton, PA 18042-1782, USA) : Use of the ChromImage flatbed scanner for quantification of high-performance thin layer chromatograms in the visible and fluorescence-quenching mode. Acta Chrom. 17, 250-260 (2006). The ChromImage flatbed scanner densitometer with Galaxie-TLC software has been used for the quantification of silica gel HPTLC. The visible mode was evaluated by determination of the recovery of rhodamine B from a four-dye mixture and by determination of the precision of replicate analysis. Determination of caffeine in analgesic tablets and a cola beverage were performed in the fluorescence-quenching mode.

HPTLC, quantitative analysis, flatbed scanner 3f

98 017 A. N. CAMPBELL*, J. SHERMA (*Dept. of Chem., Lafayette Col., Easton, PA 18042, USA): Comparative evaluation of precoated silica gel plates for preparative layer chromatography. Acta Chrom. 13, 102-108 (2003). Five commercial preparative layer chromatography plates precoated with silica gel of 1 mm thickness were compared on the basis of theoretical plate number and resolution by developing a test dye mixture (1.0 - 5.0 µg per zone) with ethyl acetate - methanol - water 4:1:1. Best results were obtained with the Mallinckrodt-Baker layer with 4.5 - 5.5 µm spherical particles. With one exception the efficiency and resolution of the other layers correlated with their particle size.

preparative TLC, comparison of methods, stationary phases 3b

98 018 Nada PERISIC-JANJIC*, T. DJAKOVIC-SEKULIC (*Department of Chemistry, Faculty of Science, University of Novi Sad, Trg D. Obradovica 3, 21000 Novi Sad, Serbia): Study of the characteristics and separation properties of unconventional TLC supports. Part I. J. Planar Chromatogr. 19, 438-442 (2006). TLC of linear aliphatic alcohols with 1 to 20 carbon atoms, as the esters of 3,5-dinitrobenzoic acid, on silica gel impregnated with paraffin oil, aminoplast, microcrystalline cellulose, rice starch, and talc with water - ethyl methyl ketone - dioxane (35:8:X where X = 15-70). Detection by spraying with 0.01 % solution of rhodamine B in methanol, followed by evaluation under UV light at 366 nm. The stationary phases were characterized by use of separation factor, resolution, and delta RF values.

qualitative identification 3b

98 019 W. PRUS (Faculty of Textile Engineering nd Environmental Protection, University of Bielsko-Biala, Bielsko-Biala, Poland): GC-MS study of the products cleaved from RP-18 chemically bonded phases for TLC during thermal modification. J. Planar Chromatogr. 19, 324-326 (2006). GC-MS investigation of dichloromethane extracts from three different types of RP-18 modified silica gel used as TLC adsorbents. Alkenes and carbonyl compounds (two different aldehydes and one ketone) were identified.

3b

98 020 A. V. GERASIMOV, (All-Russian Scientific Institute of Food Flavorings, Acids, and Dyes (GU VNIIPAKK), Liteinyi pr. 55, St. Petersburg, 191104, Russia): Use of the software processing of scanned chromatogram images in quantitative planar chromatography. J. Anal. Chem. 59 (4), 348-353 (2004). Demonstration of quantitative analysis using the software processing of scanned chromatogram images for e.g. food dyes. Digitalization of chromatograms obtained by scanning with a flatbed scanner using the special-purpose software for quantitative analysis.

quantitative analysis, software processing, scanning 3f

98 021 J. WANG (Jie Wang), D. WANG* (Dongyuan Wang), H. ZHANG (Hongxia Zhang), Y. ZHANG (Yanhua Zhang), S. ZHOU (Shuyu Zhou) (*College of Pharmacy, Shenyang Pharmaceutical University, Shenyang 110016, P.R. China): A new plate for planar electrochromatography. J. Planar Chromatogr. 19, 313-318 (2006). A new bonded RP-18 sintered silica gel plate for use in reversed-phase planar electrochromatography (PEC) has been prepared by fusing a mixture of silica gel and glass powder then reaction with octadecyltrichlorosilane. The plates have good chromatographic characteristics and good mechanical stability, and can be regenerated by soaking in acetone. TLC and PEC of p-aminoazobenzene, azobenzene, p-hydroxyazobenzene, Sudan II, and Sudan III with acetonitrile - water 17:3.

electrochromatography 3b

4. Special techniques

98 022 A. CRECELIUS*, M. R. CLENCH, D. S. RICHARDS (*Biomed. Res. Centre, Sheffield Hallam Univ., Sheffield, UK): TLC-MALDI in pharmaceutical analysis. LC-GC Europe 16, Issue 4, 225-229 (2003). A technique for the direct determination of substances from TLC plates by MALDI-MS is discussed. Methods for the generation of quantitative data by adding an internal standard to the development solvent are described and the use of post-source decay-MALDI experiments in conjunction with TLC-MALDI-MS for compound identification is reported.

pharmaceutical research, online TLC-MS 4e

98 023 M. J. FORD, M. A. DEIBEL, B. A. TOMKINS, G. J. VAN BERKEL* (*Org. and Biol. Mass Spectrometry Group, Chem. Science Division, Oak Ridge Nat. Lab., Oak Ridge, Tennessee 37831-6131): Quantitative thin-layer chromatography/mass spectrometry analysis of caffeine using a surface sampling probe electrospray ionization tandem mass spectroscopy system. Anal. Chem. 77, 4385-4389 (2005). TLC of caffeine on RP-8 with methanol - water (7:3) followed by heating at 110° C for 15 min. Detection at 214 nm. Quantification of caffeine at the low-nanogram level is performed directly from a surface of the RP-8 TLC plate using a surface sampling probe and an ESI mass spectrometry system employing selected reaction monitoring detection, and a deuterium-labelled internal standard spotted with the samples. TLC/ESI-MS/MS successfully allowed caffeine quantification in six commercially available beverages using only minimal sample preparation. The resulting calculated analyte concentrations exhibited accuracy comparable to the HPLC/UV method. However, the surface sampling probe used was restricted to unpolar layers.

quantitative analysis, qualitative identification, online TLC-MS, caffeine 4e

98 022 G. J. VAN BERKEL*, A. D. SANCHEZ, J. M. E. QUIRKE (*Org. and Biol. MS Group, Chem. Sc. Div. Oak Ridge Nat. Lab., Oak Ridge, Tennessee 37831-6131): Thin-layer chromatography and electrospray mass spectrometry coupled using a surface sampling probe. Anal. Chem. 74, 6216-6223 (2002). A combined surface sampling probe/electrospray emitter was used for the direct readout of TLC plates by electrospray MS. HPTLC of methylene blue, crystal violet, and rhodamine 6G on RP-18 with methanol - tetrahydrofuran 3:2 containing 50-100 mM ammonium acetate, followed by MS detection in positive ion mode. HPTLC of fluorescein, naphtholblue black, and fast green FCF on PR-18 with methanol - water 7:3, followed by MS detection in negative ion mode. Acquisition of mass spectra of components of individual bands was shown by manual stepping to and sampling from specific locations within the bands. Computer-controlled scanning of lanes was illustrated by using multiple ion monitoring in positive and negative ion modes. Readout resolution, the limits of scan speed, detection levels, TLC phase (restricted to nonpolar phases), and eluting solvents were investigated.

HPTLC, online TLC-MS, surface sampling 4e

98 023 G. J. VAN BERKEL*, V. KERTESZ (*Org. and Biol. Mass Spectrometry Group, Chem. Science Division, Oak Ridge Nat. Lab., Oak Ridge, Tennessee 37831-6131): Automated sampling and imaging of analytes separated on thin-layer chromatography plates using desorption electrospray ionization mass spectrometry. Anal. Chem. 78, 4938-4934 (see correction p.6283) (2006). Modest modifications to the atmospheric sampling capillary of a commercial electrospray mass spectrometer are described. Discussions pf upgrades to a developed surface positioning control software package used to enable the automated sampling and imaging of analytes on and within large area surface substrates using desorption electrospray ionization mass spectrometry. Application of rhodamine B, 6G and 123 on TLC RP-8 plates with methanol - water containing 500 mM ammonium acetate 3:1. Examples are shown for user-defined spot sampling from separated bands on a TLC plate, scanning of the complete development lane, or imaging of analyte bands in

a development lane.

online TLC-MS 4e

98 024 M. LANCASTER, D. M. GOODALL*, E. T. BERGSTROEM, S. MCCROSSEN, P. MYERS (*Dept. of Chem., Univ. of York, York YO10 5DD, UK): Real-time image acquisition for absorbance detection and quantification in thin-layer chromatography. Anal. Chem. 78, 905-911 (2006). First quantitative study of real-time image acquisition of TLC plates during development. Procedures are described for imaging using a CCD camera and for image processing, incorporating corrections for fixed pattern effects and compensation for the moving solvent front, to measure the absorbance of the analyte. Example of use was the TLC and HPTLC separation of Sudan II on silica gel with dichlormethane in a horizontal chamber. The integrated peak areas were found to be independent of time and distance moved. The method gives limits of detection better than those from offline measurements on wetted TLC plates. In addition, obtained information on the degree of solvent permeation into the stationary phase at the analyte position could be useful for TLC and HPLC method development.

quantitative analysis, real-time image acquisition 4e

98 025 I. MALINOWSKA (Dept. of Adsorption and Planar Chrom., Fac. of Chem., Maria Curie Sklodowska Univ., Pl. Marii Curie - Sklodowskiej 3, 20-031 Lublin, Poland): Some aspects of the effect of an electric field in reversed-phase thin-layer chromatography. Acta Chrom. 11, 204-214 (2001). Electric fields have been shown to affect peak width, peak area, and other chromatographic properties in reversed-phase TLC on silanized silica gel and RP-18. To eliminate electroosmotic flow, thin layer electro-chromatography was performed on unwetted layers. Horizontal and vertical modes of thin-layer electrochromatography were investigated, but only for chromatographic systems in which van der Waals forces are the sole interactions. Analytes were polycyclic aromatic hydrocarbons and hexane or heptane was used as mobile phase. The electric field affects the chromatographic process not only by replacing capillary flow by electroosmotic flow.

electrochromatography, electroosmotic flow 4d

98 026 I. MEISEN, A. FRIEDRICH, H. KARCH, U. WITTING, J. PETER-KATALINIC, J. MÜTHING* (*Institute for Medical Physics and Biophysics, University of Munster, Münster, Germany, jm@uni-muenster.de): Application of combined high-performance thin layer chromatography immunostaining and nanoelectrospray ionization quadrupole time-of-flight tandem mass spectrometry to the structural characterization of high-and low-affinity binding ligands of Shiga toxin 1. Rapid Commun. Mass Sp. 19, 3659-3655 (2005). HPTLC of a preparation of neutral Stx1-binding glycosphingolipids from human erythrocytes, comprising 21.4 % and 59.1 % of the high- and low-affinity Stx1-binding ligands Gb3Cer/CD77 and Gb4Cer respectively and their antibody positive bands on silica gel with chloroform - methanol - water 15:30:4. Crude extracts were used without any further purification for an analysis by nanoelectrospray ionization quadrupole time-of-flight mass spectrometry in the negative mode. Only analytical quantities in the microgram scale of a single glycosphingolipid species are required for the structural MS characterization.

clinical chemistry research, HPTLC, preparative TLC 4e

98 027 K. NAKAMURA, Y. SUZUKI, N. GOTO-INOE, C. YOSHIDA-NORO, A. SUZUKI* (*Spingolipid Expression Lab., Supra-Biomolecular System Research Group, Frontier Research System, Inst. of Phys. and Chem. Research (RIKEN), Saitama, Japan): Structural characterization of neutral glycosphingolipids by thin-layer chromatography coupled to matrix-assisted laser desorption/ionization quadrupole ion trap time-of-flight MS/MS. Anal. Chem. 78, 5736-5743 (2006). Structural analysis of neutral glycosphingolipids by direct coupling of TLC to MALDI-QIT-TOF MS/

MS. TLC of neutral glycosphingolipids on silica gel with chloroform - methanol - water 65:35:8. The underivatized glycosphingolipid spots were directly subjected to MALDI-QIT-TOF MS after addition of a matrix solution of 2,5-dihydrobenzoic acid in acetonitrile - water 1:1, which proved to be suitable for MS/MS analysis with high sensitivity. MS/MS and MS/MS/MS spectra reveal the ceramide and long-chain base structures, as well as the sugar sequences. Furthermore, derivatization with primuline (0,01 % in acetone - water, 4:1), a non-destructive fluorochrome for lipid detection, was used to locate glycosphingolipids on a TLC plate prior the MS analysis. The coupling of TLC-immunostaining of glycosphingolipids to MALDI-QIT-TOF MS/MS is shown to be a powerful method to identify both the antibody-specific sugar and the ceramide structures.

online TLC-MS, glycosphingolipids, immunostaining 4e

98 028 S. NYIREDI (Res. Inst. for Med. Plants, Budakalász, Hungary): Multidimensional Planar Chromatography. LC-GC Europe 16, Issue 12a, 52-59 (2003) LC-GC Europe 16, 52-59 (2003). The potential of various multidimensional planar chromatography (MD-PC) techniques, including comprehensive 2D planar chromatography (PC×PC), targeted or selective 2D PC (PC+PC), modulated 2D PC (nPC) and coupled-layer PC (PC-PC) is discussed.

multidimensional planar chromatography 4d

98 029 A. PARENT, T. ANDERSON, D. MICHAELIS, G. JIANG, P. SAVAGE, M. LINDFORD* (*Department of Chemistry and Biochemistry, Brigham Young University, Provo, UT 84602, United States, mrlindford@chem.byu.edu): Direct ToF-SIMS analysis of organic halides and amines on TLC plates. Appl. Surf. Sci. 252, 6746-6749 (2006). To show the application of direct time-of-flight secondary ion mass spectrometry (ToF-SIMS) to analyze a reaction mixture between an alpha-bromoamide (1), and picolylamine, to form the substitution product (3) via SN2 (1 and 3 are indistinguishable by TLC on silica gel with dichloromethane - methanol - ammonia 100:15:2), a series of organic halides with widely differing structures (trifluoromethanesulfonamide, a vinyl bromide, a silyl-protected primary bromide, and iodosobenzene diacetate) were separated on TLC plates under the same conditions. The resulting spots and background were analyzed by positive and negative ion spectra. In all cases, the halide signals were notably stronger than the background signals. In addition, a series of amines (1,1-carbonyl diimidazole, norepinephrine, adenosine, and cinchonidine) was separated on TLC plates and analyzed directly by ToF-SIMS. In all but one (adenosine), even quasi-molecular ions were observed.

pharmaceutical research, qualitative identification 4e

6. Alcohols

98 030 M. WAKSMUNDZKA HAJNOS*, G. JOZWIAK, T. WAWRZYNOWICZ (*Dept. of Inorg. and Anal. Chem., Med. Univ., Staszica 6, 20-081 Lublin, Poland): RP-TLC and RP-HPLC analysis of aliphatic alcohols as the DANS-Cl derivatives. Acta Chrom. 11, 51-61 (2001). A homologous series of C4 - C12 aliphatic alcohols was derivatized with dansyl chloride to introduce a fluorophoric group into the molecule which enables TLC and/or HPLC analysis with UV detection. The progress of the reaction under different sets of conditions was monitored by RP-TLC and it was confirmed that the best yield was obtained after 24 h at ambient temperature. TLC on RP-18 with acetonitrile - water 3:2 in a horizontal chamber. Densitometry at 361 nm.

qualitative identification, densitometry, aliphatic alcohols, dansyl chloride 6

7. Phenols

98 031 U. HUBICKA, J, KRZEK*, J. KALETA, A. NIEDZWIEDZ (*Jagiellonian University, Collegium Medicum, Department of Inorganic and Analytical Chemistry, Medyczna 9, 30-688 Cracow,

Poland): Evaluation of densitometric TLC for quantitative analysis of selected phenolic acids for standardization of propolis concentrates. J. Planar Chromatogr. 19, 449-453 (2006). HPTLC of caffeic, p-coumaric, and ferulic acid on silica gel with dichloromethane - acetonitrile - 90 % formic acid 95:5:1. Quantitative determination by absorbance measurement at 320 nm. The method is precise and accurate.

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

98 122 A. MOHAMMAD et al., see section 33

98 032 A. MORNAR, Marica MEDIC-SARIC*, I. JASPRICA (*Department of Medicinal Chemistry, University of Zagreb, A. Kovacica 1, 10 000 Zagreb, Croatia): ADME data for polyphenols characterized by reversed-phase thin-layer chromatography. J. Planar Chromatogr. 19, 409-417 (2006). The chromatographic behavior of polyphenols (flavonoids and phenolic acids) has been investigated by RP-TLC to establish relationships between chromatographic data and selected ADME (absorption, distribution, metabolism, and elimination) data. TLC of six flavones (flavone, chrysin, tectochrysin, apigenin, acacetin, luteolin), nine flavonols (galangin, kaempferol, kaempferide, morin, quercetin, rhamnetin, isorhamnetin, tamarixetin, myricetin), seven flavanones (flavanone, pinocembrine, pinostrobin, naringenin, sakuranetin, isosakuranetin, hesperitin), and eight phenolic acids (caffeic, ferulic, isoferulic, cinnamic, o-coumaric, m-coumaric, p-coumaric, and sinapic acid) on RP-18. Binary mobile phases were prepared from mixtures of methanol and phosphate buffer; ascending development was performed at room temperature in a saturated glass chamber. Detection at 254 nm.

pharmaceutical research, herbal, qualitative identification 7

8. Substances containing heterocyclic oxygen

98 033 P. K. SALO, A. ESSÉN-SUURONEN, H. SALOMIES, R. A. KETOLA, R. KOSTIAINEN* (*Faculty of Pharmacy, Division of Pharmaceutical Chemistry, P. O. Box 56, FIN-00014 University of Helsinki, Finland): HPTLC, with UV and MS detection, and preparative-layer chromatography for analysis and purification of synthesis products. J. Planar Chromatogr. 19, 371-377 (2006). HPTLC of five isoflavone products and 2-phenylchromone as reference standard on silica gel, pre-washed with acetonitrile, in an unsaturated chamber, twice with chloroform or dichloromethane. Determination by absorbance measurement at 280 nm. Preparative layer chromatography on silica gel. A new device for isolation of sythesis products in sub-milligram amounts was successfully employed.

qualitative identification, preparative TLC, HPTLC, synthetic organic chemistry 8a

98 034 Agnieszka SKALSKA*, A. MATYSIK, M. GERKOWICZ, M. WÓJCIAK-KOSIOR (*Department of Chemistry, Laboratory of Planar Chromatography, Medical Academy, Staszica 6, 20-081 Lublin, Poland): Preparative reversed-phase high-performance thin-layer chromatography for analysis of anthocyanins. J. Planar Chromatogr. 19, 463-466 (2006). Preparative RP-HPTLC of anthocyanin extracts (e. g. malvidine) on silica gel with five-step gradient elution with different concentrations of toluene - acetonitrile - water - formic acid - n-butanol - 2-propanol and resp. addition of tert-butylmethyl ether. After extraction of the third zone from the layer, HPTLC on RP-18 as the best stationary phase with a three-step gradient elution using methanol - water - formic acid in different ratios. Quantitative determination by absorbance measurement at 470 nm.

herbal, quality control, preparative TLC, HPTLC, densitometry, quantitative analysis, qualitative identification 8a

10. Carbohydrates

98 035 S. D. WAGNER*, J. PACHUSKI, B. FRIED, J. SHERMA (*Dept. of Chem., Lafayette Col., Easton, Pennsylvania 18042, USA: Thin layer chromatographic analysis of carbohydrates and amino acids in Schistosoma mansoni (Trematoda) cercariae. Acta Chrom. 12, 159-169 (2002). TLC of carbohydrates on LK5DF silica gel plates containing 19 lanes and a pre-adsorbent sample-application zone, prewashed with dichloromethane - methanol 1:1, with ethyl acetate - glacial acetic acid - methanol - water 12:3:3:2. Detection by spraying with alpha-naphthol-sulfuric acid sugar-detection reagent followed by heating at 110 ºC for 5 min. Densitometry of glucose and raffinose at 515 nm. HPTLC of amino acids with four different TLC systems: on silica gel with preadsorbent zone, on RP-18 with concentrating zone, on cellulose F with n-butanol - acetic acid - water 3:1:1, and on ion-exchange sheets with citrate buffer of pH 3.3 (84 g citric acid monohydrate, 16 g NaOH, and 5.9 mL HCl 37% per L). Detection by spraying with ninhydrin reagent, followed by drying in air for 30 min and heating for 10 min at 110 ºC. Quantification by densitometry at 495 nm for histidine and 610 nm for tryptophan.

pharmaceutical research, quantitative analysis, qualitative identification, HPTLC, carbohydrates, amino acids 10a

98 036 D. J. CLINE, B. FRIED, J. SHERMA* (*Dept. of Chem., Lafayette College, Easton, PA 18042, USA): TLC and GC-MS identification of glucose and maltose in Biomphalaria glabrata (Gastropoda), and use of quantitative TLC to determinate the effect of starvation on the amounts of these carbohydrates. Acta Chrom. 9, 79-86 (1999). HPTLC of the primary carbohydrates in the planorbid snail Biomphalaria glabrata on silica gel with acetonitrile - water 17:3 and ethyl acetate - acetic acid - methanol - water 12:3:3:2, on LK5DF silica gel with ethyl acetate - acetic acid - methanol - water 12:3:3:2, on RP-18 with tetrahydrofuran - water 22:3, on cellulose with ethyl acetate - pyridine - water 2:1:2 and on amino-bonded layers with ethyl acetate - pyridine - water - acetic acid 12:6:2:1. The use of specific detection reagents for reducing sugars (4-aminobenzoic acid detection reagent and aniline - DPA reagent), spiking experiments, and spraying with alpha-naphthol - sulfuric acid reagent for quantitative analysis (densitometry at 515 nm) aided the identification of glucose and maltose. GC-MS analysis confirmed the identification of maltose and glucose on the basis of retention times and spectral fingerprints. A starvation study was conducted to determine changes in sugar levels in B. glabrata digestive gland-gonad complex (DGG) and hemolymph samples during a 12-day starvation period.

pharmaceutical research, quantitative analysis, qualitative identification, HPTLC, densitometry, glucose, maltose 10a

98 037 J. J. SCHARITER*, B. FRIED, J. SHERMA (*Dept. of Chem., Lafayette Col., Easton, PA 18042, USA):: TLC analysis of glucose and maltose in Biomphalaria glabrata (Gastropoda) infected with Schistosoma mansoni (Trematoda). Acta Chrom. 11, 102-107 (2001). The snail Biomphalaria glabrata is medically important because it serves as the intermediate host for the development and transmission of the human blood fluke parasite Schistosoma mansoni. The purpose of this study was to use HPTLC to determine the effects of S. mansoni larval infection on the maltose and glucose content of the hemolymph and digestive gland-gonad complex of B. glabrata. TLC on silica gel with pre-adsorbent zone and 19 channels, with ethyl acetate - acetic acid - methanol - water 13:3:3:2. Detection by spraying with 1-naphthol - sulfuric acid reagent, quantitative determination by absorbance measurement at 515 nm.

pharmaceutical research, quantitative analysis, qualitative identification, densitometry, glucose, maltose 10a

98 038 J. SHERMA*, D. L. ZULICK (*Dept. Of Chem., Lafayette Coll., Easton, PA 18042-1782, USA): Determination of fructose, glucose and sucrose in beverages by high-performance thin layer

chromatography. Acta Chrom. 6, 7-12 (1996). HPTLC of fructose, glucose and sucrose on silica gel with concentration zone. Impregnation of the layer (except concentration zone) by spraying with 0.1 M sodium bisulfate solution, followed by drying and spraying with citrate buffer of pH 4.8. Three-fold development with acetonitrile - deionized water 17:3 after chamber saturation. Visualization by spraying with 1-naphthol - sulfuric acid reagent and heating at 110 °C. Quantification by densitometry at 515 nm.

food analysis, quantitative analysis, densitometry, HPTLC, fructose, glucose, sucrose 10

98 039 A. SZABÓ, A. KÓNYA, I. WINKLER, G. MÁTÉ, B. ERDÉLYI* (*IVAX Drug Research Institute Ltd., P. O. Box 82, 1326 Budapest, Hungary): Simultaneous monitoring of target compounds and carbohydrate patterns during pharmaceutical fermentations. J. Planar Chromatogr. 19, 418-421 (2006). TLC of fructose, glucose, saccharose, maltose, maltotriose, maltotetraose, maltopentaose, maltohexaose on silica gel in a pre-saturated twin-trough chamber with chloroform - carbon tetrachloride - 35 % aqueous formic acid - methanol 20:5:17:22. After drying the chromatogram was immersed in a solution of sulfuric acid reagent (10 mL conc. sulfuric acid in 200 mL of a cooled 1:1 mixture of 1-propanol and carbon tetrachloride) and heated for 8 min at 108 °C. Quantitative determination by absorbance measurement at 290 nm.

qualitative identification, quantitative analysis, densitometry, pharmaceutical production 10

98 040 H. THERISOD, V. LABAS, M. CAROFF* (*Equipe “Endotoxines”, UMR 8619 du Centre National de la Recherche Scientifique, Biochimie, Universitè de Paris-Sud, F-91405 Orsay, France): Direct microextraction and analysis of rough-type lipopolysaccharides by combined thin-layer chromatography and MALDI mass spectrometery. Anal. Chem. 73, 3804-3807 (2001). TLC of lipopolysaccharides from intact bacteria on silica gel with chloroform - methanol - water - triethylamine 12:6:1:0.04. Detection by spraying with 10 % sulphuric acid in ethanol followed by heating at 150 °C for 5 min. For the non-destructive visualization the plates were pre-washed twice with propanol - water 1:1 and developed in methanol - water 1:1 which made the products appear as dull white spots on a bright background. Zones containing lipopolysaccharides or lipid A molecular species were isolated from the plate and analyzed by MALDI mass spectrometery.

lipopolysaccharides 10b

11. Organic acids and lipids

98 041 F. M. HELMY (Dept. of Biol. Sci., Delaware State Univ., Dover, DE, USA).: Cardiolipin, its preferential deacylation in mammalian myocardia. Mini review and chromatographic-computional analysis. Acta Chrom. 17, 9-19 (2006). Comparative correlative TLC analysis conducted on whole-tissue homogenate of control and in-vitro-incubated samples of mammalian myocardia and bullfrog cardiac muscle and thigh skeletal muscle. The focus in the study was on TLC-densitometric analysis of the endogenous cardiolipin of control samples and their respective incubated samples to identify the products, e. g. lysocardiolipin, formed during in-vitro incubation. TLC of the chloroform-methanol extracts (2:1) obtained after the incubation from the freeze-dried tissues homogenate on silica gel with 1-propanol - chloroform - ethyl acetate - methanol - water 50:50:50:21:18. Visualization with thionine reagent, densitometry at 600 nm.

quantitative analysis, densitometry, cardiolipin, monolysocardiolipin, phosphatidylethanolamine, plasmalogen 11c

98 042 M. WAKSMUNDZKA-HAJNOS*, H. D. SMOLARZ, R. NOWAK (*Dept. of Inorg. and Anal. Chem., Med. Acad., Staszica 6, 20-081 Lublin, Poland): Chromatographic separations of phenolic acids by normal-phase TLC. Retention behaviour on polar adsorbents (silica, alumina, polyamide) with non-aqueous mobile phases. Acta Chrom. 9, 38-54 (1999). TLC of benzoic and cinnamic acid derivatives on silica gel 60 H compared with that on alumina oxide 60 G and po-

lyamide layers with non-aqueous ternary mobile phases comprising a non-polar diluent (n-heptane), a polar modifier (2-propanol, dioxane, tetrahydrofuran and ethyl acetate) and 2 % acetic acid in horizontal chambers. Visualization under UV 254 nm, and by spraying with diazotized sulphanilic acid in 10 % sodium bicarbonate solution, or with a 2 % solution of ferric chloride. Chromatogram parameters and the correlation diagrams were used for comparison of separation selectivity.

comparison of methods, qualitative identification, phenolic acids 11a

13. Steroids

98 043 M. BATHORI*, H. KALASZ (*Dept. of Pharmacognosy, Fac. of Pharm., Univ. of Szeged, Hungary): Separation methods for phytoecdysteroids. LC-GC Europe 14, Issue 10, 626-631 (2001). TLC of 20-hydroxyecdysone on silica gel, pre-washed with methanol. Different mobile phases are investigated, the mixture of chloroform - methanol - benzene 25:5:3 was used for quantification. Densitometry at 254 nm. TLC of ecdysteroids under the same conditions, detection at 254 nm, in white light and at 366/>400 nm (fluorescence) after spraying with a vanillin-sulphuric acid reagent. Identification was facilitated with two-dimensional TLC, displacement TLC (D-TLC) and forced flow TLC (FF-TLC). Comparison with HPLC.

pharmaceutical research, quantitative analysis, comparison of methods, ecdysteroids 13

98 044 B. JANOSKA*, T. WIELKOSZYNSKI, K. TYRPIEN, C. DOBOSZ, D. BODZEK (*Med. Univ. of Silesia, Fac. of Med., Dept. of Chem., Jordana 19, 41-808 Zabrze, Poland): Effect of steroid hormones on results from determination of oxycholesterol by TLC. Acta Chrom. 13, 95-101 (2003). Chromatographic systems comprising different stationary and mobile phases were investigated for determination of oxysterols in plasma by TLC. Two chromatographic systems are used to ensure selectivity. TLC on RP-18 with 2-propanol - dichloromethane 3:97, or on silica gel with acetone - chloroform 1:9 in a horizontal chamber. Detection by spraying with Liebermann - Burchard reagent (methanol - conc. H2SO4 - acetic anhydride, 10:1:1) followed by heating at 110 °C for 5 min. Densitometry at 366 nm (oxysterols) and 254 nm (steroid hormones).

clinical chemistry research, pharmaceutical research, quantitative analysis, qualitative identification, oxycholestrols, steroid hormones 13c

14. Steroid glycosides, saponins and other terpenoid glycosides

98 045 H. AGRAWAL, N. KAUL, A. R. PARADKAR, K. R. MAHADIK* (*Dept. of Pharm. Anal. Chem., Bharati Vidyapeeth Deemed Univ., Poona Col. of Pharm., Erandwane, Pune-411038, Maharashtra State, India): Separation of bacoside A3 and bacopaside II, major triterpenoid saponins in Bacopa monnieri, by HPTLC and SFC. Application of SFC in implementation of uniform design for herbal drug standardization, with thermodynamic study. Acta Chrom. 17, 125-150 (2006). HPTLC of bacoside A3 and bacopaside II on RP-18 F254 after pre-washing with methanol and heating at 60° C for 5 min. Development with toluene - methanol - ethyl acetate 15:5:4 in the dark in a controlled humidity chamber (humidity of 55 - 65 %). Densitometry at 344 nm. The method is available for content determination of bacoside A3 and bacopaside II in herbal extracts and commercial formulations.

quantitative analysis, HPTLC, densitometry, saponins 14

17. Amines, amides and related nitrogen compounds

98 046 H. A. KHAN (Department of Biochemistry, College of Science, King Saud University, P.O. Box 2455, Riyadh, 11451 Saudi Arabia): TLC determination of aliphatic polyamines on calcium sulfate layers. Chromatographia 64 (7-8), 423-427 (2006). TLC of six polyamines (ornithine, cit-

rulline, putrescine, cadaverine, spermidine and spermine) on calcium sulfate (CaSO4) and silica gel with 11 different mobile phases, using methanol as the solvent to enhance selectivity and produce differential Rf values. On CaSO4 better separation was achieved than on silica gel, no derivatization is needed, and development time is about 1/3 shorter. Quantitative determination by absorbance measurement at 550 nm after extracting substances from the plates and eluting from the coating material. Limit of detection (LOD) was 750 ng, limit of quantification (LOQ) was 1880 ng. Quantitative separation of underivatized polyamines in spiked human urine samples.

pharmaceutical research, comparison of methods, calcium sulfate layer, separation of polar compounds, biogenic polyamines 17a

18. Amino acids and peptides, chemical structure of proteins

98 047 S. A. NABI*, M. A. KHAN (*Dept. of Chem., Aligarh Muslim Univ., Aligarh, India): Selective TLC separation of lysine and threonine in pharmaceutical preparations. Acta Chrom. 13, 161-171 (2003). Lysine and threonine were separated and quantitatively determined from the mixture of amino acids present in a commercially available drug. TLC of alpha-amino acids on layers prepared from a 1:4 stannic arsenate - cellulose mixture with a variety of mobile phases. Best separation was achieved with n-butanol - formic acid - water 7:2:1; or isopropanol - acetic acid - water 8:1:1, or isopropanol - formic acid - water 7:2:1. Detection by dipping in 1 % ninhydrin in butanol after heating at 60 °C . Quantitative determination by spectrophotometry with hydrindantin-methyl cellosolve reagent.

pharmaceutical research, quality control, quantitative analysis, qualitative identification, lysine, threonine 18a

98 048 F. BUHL, Monika GALKOWSKA* (*Insitute of Chemistry, Department of Analytical Chemistry, Silesian University, 9 Szkolna Street, 40-006 Katowice, Poland): Determination of methionine in pharmaceuticals after chromatographic separation. J. Planar Chromatogr. 19, 401-404 (2006). TLC of methionine, L-cystine, calcium pantothenate, vitamin B1, vitamin B7, and p-aminobenzoic acid on silica gel in a pre-saturated chamber with n-propanol - water - chloroform 5:2:1. Quantification of methionine was based on the oxidation and reaction with leuco xylene cyanol FF solution, which formed a blue dye. Quantitative determination by absorbance measurement at 613 nm.

quality control, qualitative identification, quantitative analysis 18a

21. Purines, pyrimidines, nucleic acids and their constituents

98 049 Jolanta SOCHACKA*, A. KOWALSKA (* Department of General and Analytical Chemistry, Faculty of Pharmacy, The Medical University of Silesia, Jagiellonska 4, 41-200 Sosnowiec, Poland): Comparison of calculated values of the lipophilicity of 2,6-disubstituted 7-methylpurines with values determined by RPTLC. J. Planar Chromatogr. 19, 307-312 (2006). TLC of 2,6-disubstituted 7-methylpurines and 6-mercaptopurine on RP-18 with mixtures of acetone and buffer (sodium acetate - veronal, pH 7.0). Detection in UV light at 254 nm. Partly good agreement was obtained between experimental log P (TLC) values and calculated Clog P values.

qualitative identification 21a

22. Alkaloids

98 050 N. C. NIKOLIC*, M. Z. STANKOVIC (*Fac. of Techn., Bulevar oslobodjenja 124, 16000 Leskovac, Serbia and Montenegro): Acid hydrolysis of potato tuber sprout glykoalkaloids and kinetics of solanidine extraction in three-phase systems. Ital. J. Food Sci. 18, 287-294 (2006). The ratio

of alpha-solanine to alpha-chaconine of tuber sprouts was determined by TLC on silica gel with methanol - chloroform - 1 % aqueous ammonium hydroxide 2:2:1. For quantitative determination zones were isolated from the plate, extracted with 2 % aqueous acetic acid, and measured spectrophotometrically at 420 nm with methyl orange.

food analysis, qualitative identification, autoradiography, potato 22

98 051 A. EVIDENTE*, A. ANDOLFI, A. ABOU-DONIA, S. TOUEMA, H. HAMMODA, E. SHAWKY, A. MOTTA (*Dipartimento di Scienze del Suolo della Pianta e dell´Ambiente, Universitá di Napoli Federico II, Via Universitá 100, I-80055 Portici, Italy, evidente@unina.it): (-)-Amarbellisine, a lycorine-type alkaloid from Amaryllis belladona L. growing in Egypt. Phytochemistry. 65, 2113-2118 (2004) . HPTLC of (-)-amarbellisine in the bulbs of Amaryllis belladona L. on silica gel with chloroform - methanol 9:1 with 1 drop of ammonia. Quantitative determination at 254 nm to estimate the alkaloidal content in the flowering stage (in April), and in the preflowering stage (in November).

herbal, HPTLC, densitometry, quantitative analysis 22

98 052 M. GADZIKOWSKA*, G. GRYNKIEWICZ (*Dept. of Inorg. and Anal. Chem., Med. Acad., Staszica 6, 20-081 Lublin, Poland): Commentary on the chromatographic retention of Chelidonium alkaloids. Acta Chrom. 11, 62-74 (2001). TLC of basic isolates of Chelidonium majus L. (homochelidonine, allocryptopine, chelidonine, protopine, chelerythrine, san-guinarine and berberine) on silica gel with different mobile phases: toluene - ethyl acetate - methanol 17:2:1, toluene - ethyl acetate 4:1, toluene - ethyl acetate - methanol 11:8:1 and toluene - methanol 19:1; and on aluminum oxide 60 type E (basic) with toluene - ethyl acetate 9:1, 4:1, and 3:2; and toluene - methanol 19:1 in horizontal chamber. Evaluation under UV light at 254 and 366 nm, or by spraying with Dragendorff's reagent. The HPLC-UV method was developed in parallel.

herbal, qualitative identification, alkaloids, Chelidonium, qualitative analysis 22

98 053 M. M. GUPTA*, A. SRIVASTAVA, A. K. TRIPATHI, H. MISRA, R. K. VERMA (*Analytical Chemistry Division, Central Institute of Medicinal and Aromatic Plants, P. O. CIMAP, Lucknow-226015, India): Use of HPTLC, HPLC, and densitometry for qualitative separation of indole alkaloids from Rauwolfia serpentina roots. J. Planar Chromatogr.19 , 282-287 (2006). HPTLC of ajmaline, ajmalicine, and reserpine on silica gel, prewashed with methanol, in an unsaturated twin-trough chamber with toluene - methanol 19:6. Detection by dipping in Dragendorff's reagent. Quantitative determination by absorbance measurement at 520 nm.

herbal, traditional medicine, HPTLC, densitometry, quantitative analysis 22

98 054 Ágnes SÁRKÖZI*, Á. M. MÓRICZ, P. G. OTT, E. TYIHÁK, Á. KÉRY (*Department of Pharmacognosy, Faculty of Pharmacy, Semmelweis University, Üllöi Str. 26, 1085 Budapest, Hungary): Investigation of Chelidonium alkaloids by use of a complex bioautographic system. J. Planar Chromatogr. 19, 267-272 (2006). TLC of chelerythrine, chelidonine, and sanguinarine on silica gel with dichloromethane - methanol 97:3. Use of the mobile phase recommended by Ph. Eur. 5, propanol - water - formic acid 90:9:1, did not enable satisfactory separation of Chelidonium alkaloids. Development at 20 - 24 °C and 60 % relative humidity in a presaturated TLC chamber. After drying evaluation under UV light at 254 and 366 nm.

herbal, quality control, qualitative identification, bioautography 22

23. Other substances containing heterocyclic nitrogen

98 055 Joanna NOWAKOWSKA (Medical University of Gdansk, Faculty of Pharmacy, Department of

Physical Chemistry, Al. Gen. J. Hallera 107, 80-416, Gdansk, Poland): The retention behavior of selected porphyrins on silica gel, polyamide, and cellulose TLC plates. J. Planar Chromatogr. 19, 393-397 (2006). TLC of uroporphyrin I, uroporphyrin III, coproporphyrin I, coproporphyrin III, and protoporphyrin IX (as methyl esters) on polyamide 11, cellulose and silica gel with methanol, ethanol, propanol, butanol, acetonitrile and tetrahydrofuran in a saturated chamber. Porphyrins on cellulose were detected by placing in iodine vapor for 5 min; on silica gel and polyamide 11 the porphyrins were detected as red spots in UV light at 254 nm. Chromatographic retention data and a possible retention mechanism are discussed.

clinical chemistry research, qualitative identification 23a

98 056 M. PODGORNA (Inst. of Chem., Silesian Univ., 9 Szkolna Street, 40-006 Katowice, Poland): Separation of porphyrins and metallporphyrins by TLC. Acta Chrom. 12, 226-233 (2002). TLC of porphyrins and their complexes with Cu (II) and Ni (II) on silica gel (activated at 120 ºC for 30 min) with carbon tetrachloride - chloroform 1:1 after chamber saturation for 30 min. After drying the plates at RT, spots were visible on the plates. The effects of Cu (II) and Ni (II) cations on the separation of hydroxy and alkyloxy tetraphenylporphyrin derivatives were determined.

metallporphyrins 23a

27. Vitamins and various growth regulators

98 057 I. BARANOWSKA*, A. KADZIOLKA (*Dept. of Anal. and General Chem., Silesian Technical Univ., Gliwice, Poland): RPTLC and derivative spectrophotometry for the analysis of selected vitamins. Acta Chrom. 6, 61-71 (1996). TLC on RP-18 in Shandon chamber with water - methanol 5:4 and water - acetic acid 7:1 for separation of water-soluble vitamins (nicotinic acid, nicotinamide, C, B1 and rutin) and with acetonitrile - benzene - chloroform 10:10:1 for fat-soluble vitamins (A-acetate, E, E-acetate). Visualization of rutin with 25 % lead (II) acetate, of other water-soluble vitamins with potassium hexaiodoplatinate (IV) solution prepared by mixing 10 % potassium iodide with 5 % hexachloroplatinic acid 9:1. Fat-soluble vitamins were detected with a 10 % solution of antimony chloride. Derivative spectrophotometry was applied to the determination of vitamins B1, B6 and A-acetate in mixtures with other vitamins.

food analysis, qualitative identification, vitamins 27

28. Antibiotics, Mycotoxins

98 058 S. A. NABI*, M. A. KHAN, S. N. KHOWAJA, A. ALIMUDDIN (*Dept. of Chem., Aligarh Muslim Univ., Aligarh, India): Thin-layer chromatographic separation of penicillins on stannic arsenate-cellulose layers. Acta Chrom. 16, 164-172 (2006). TLC of ampicillin, amoxicillin, penicillamine, benzylpenicillin and penicillin in commercial drugs (megaphen and hipenox) on self-made stannic arsenate - cellulose 1:4 layers containing 10 % CaSO4 as binder, with acetonitrile - acetic acid - chloroform 2:3:9, and acetone - acetic acid - chloroform 5:5:6. Detection by treatment with iodine vapour. The zones were extracted with methanol and spectrophotometrically determined at 460 nm using 2,3-dichloro-5,6-dicyano-p-benzoquinone as derivatization reagent.

pharmaceutical research, qualitative identification, penicillin 28a

29. Pesticides and other agrochemicals

98 059 S. A. NABI*, A. GUPTA, M. A. KHAN, A. ISLAM (*Anal. Chem. Div., Dept. of Chem., Aligarh Muslim Univ., Aligarh-202002, India): Thin-layer chromatographic separations of some common pesticides on mixed stannic oxide-silica gel G layers. Acta Chrom. 12, 201-210 (2002). TLC of important organophosphorus, organochlorine, and pyrethroid pesticides (dichlorvos, endosulfan, malathion, monocrotophos, parathion, phorate, phosphamidon, quinalphos, dimethoate, anilofos,

chlorpyrifos, fenvalerate, methyl oxydemeton, and methyl parathion) on stannic oxide-silica gel G layers with a variety of mixed aqueous and organic mobile phases. Detection by treatment with iodine vapor. For quantification corresponding zones were removed from the plate and etracted with ethanol. Quantitative determination by spectrophotomety with the molybdenum blue method. Monocrotophos, dimethoate, and malathion in soil samples were analyzed to test the applicability of the simple method.

environmental, qualitative identification, pesticides 29b

98 060 S. A. NABI*, A. SIKARWAR (*Anal. Research Lab., Dept. of Chem., Aligarh Muslim Univ., Aligarh-202002, India) : Separation of phenolic compounds on stannic arsenate - silica gel layers - quantitative separation and determination of phenolic pesticide residues in bananas. Acta Chrom. 9, 123-132 (1999). TLC of phenolic pesticide residues (m-nitrophenol, p-nitrophenol and p-aminophenol) in banana fruits and plant-tissues on stannic arsenate-silica gel layers. Several mobile phases of different polarity were evaluated; separation of m-nitrophenol, p-nitrophenol, picric acid and p-aminophenol was best achieved with ethanol - 1.0 M citric acid 1:3. Visualization by spraying with AgNO3 - reagent (saturated AgNO3 solution in acetone 1:20 diluted with water until the precipitate of AgNO3 was dissolved) followed by heating at 105 °C, or by spraying with a mixture of 15 % FeCl3 and 1 % K4Fe(CN)6 1:1.

qualitative identification, pesticid residues 29, 32e

98 061 B. B. DAUNDKAR, R. R. MAVLE, M. K. MALVE, R. KRISHNAMURTHY* (*Directorate of Forensic Science Laboratories, State of Maharashtra, Hans Bhugra Marg, Kalina, Vidyanagari, Santa Cruz (E), Mumbai-400098, India): Detection of carbaryl insecticide in biological samples by TLC with a specific chromogenic reagent. J. Planar Chromatogr. 19, 467-468 (2006). TLC of carbaryl (1-naphthylmethylcarbamate), a common derivative of the widely used N-methylcarbamate insecticide, on silica gel with n-hexane - acetone 4:1 in a saturated twin-trough chamber. After drying the plate was heating at 100 °C for 5 min, cooled, and sprayed successively with 5 % sodium hydroxide solution and with a 1:1 mixture of 2 % diphenylamine solution and 5 % formaldehyde solution. Visual detection of the blue-green zones carbaryl.

toxicology, qualitative identification 29c

98 062 J. ESPINOZA, M. BAEZ* (*Departamento de Química Inorgánica y Analítica, Facultad de Ciencias Químicas y Farmacéuticas, Universidad de Chile, Casilla 233, Santiago, Chile): Determination of atrazine in aqueous soil extracts by high performance thin-layer chromatography. J. Chil. Chem. Soc. 48, 19-23 (2003). HPTLC validation of atrazine in aqueous extracts of soils on silica gel (layer thickness 100 µm) previously activated at 120 ºC for 30 min. The elution program applied to aqueous soil matrices started with 10 short isocratic runs (0.8 min) with acetonitrile - dichloromethane 30:70. Mixer was emptied after the tenth step and refilled to continue with 4 successive isocratic runs (2.5, 5.0, 7.5 and 25 min) with dichloromethane. The plate was dried for 1 min between each step and for 3 min after the last one. The plate was preconditioned with nitrogen for 15 s before each run. Quantitative determination by absorbance at 210 nm. Linearity is between 5 and 100 ng and recoveries ranging from 98.7 to 103.5 %. The detection limit is 1.5 ng and the quantification limit is 4.9 ng. Precision analysis shows an intra-assay variation between 1.48 and 5.47 %. The method can be applied to a broad range of soils including those with high organic matter content.

environmental, HPTLC, densitometry, quantitative analysis, AMD 29d

98 063 Malgorzata JANICKA (Faculty of Chemistry, Department of Physical Chemistry, Maria Curie-Sklodowska University, M. Curie-Sklodowska Sq. 3, 20-031 Lublin, Poland): Comparison of

different properties - log P, log kW, and phi 0 - as descriptors of the hydrophobicity of some fungicides. J. Planar Chromatogr. 19, 361-370 (2006). Log P, log kW, and phi 0 are proposed and compared as descriptors of the hydrophobicity of twenty-two dihydroxythiobenzanilides. Chromatographic data log kW and phi 0, were calculated from experimental TLC results obtained on RP-18W and cyano phases with methanol or acetone as organic modifiers. The calculated hydrophobicity was compared with the biological activity of the test substances. Development in saturated sandwich chambers at 20 °C. Detection at 320 nm.

agricultural, qualitative identification 29e

30. Synthetic and natural dyes

98 064 J. SECHRIST, J. PACHUSKI, J. SHERMA* (*Dept. of Chem., Lafayette Col., Easton, PA 18042, USA): Quantification of lutein in dietary supplements by reversed-phase high-performance thin-layer chromatography with visible-mode densitometry. Acta Chrom. 12, 151-158 (2002). HPTLC of lutein in nutrition supplements on RP-18 (with concentration zone, pre-washed with dichloromethane - methanol 1:1) with petroleum ether - acetonitrile - methanol 1:2:2. All sample solutions and developing chambers were wrapped in aluminum foil to protect the labile pigment from photo-decomposition. Densitometry at 448 nm. Three products containing lutein as the free alcohol or dipalmitate ester plus other ingredients were analyzed.

herbal, food analysis, quantitative analysis, HPTLC, densitometry, lutein 30b

32. Pharmaceutical and biomedical applications

98 066 G. AKOWUAH*, I. ZHARI, I. NORHAYATI, A. MARIAM (*Herbal secretariat, School of Pharmaceutical Sciences, Universiti Sains Malaysia, 11800 Minden, Penang, Malaysia, wuahmy@yahoo.com).: HPLC and HPTLC densitometric determination of andrographolides and antioxidant potential of Andrographis paniculata. J. Food Comp. Anal. 19, 118 - 126 (2006). HPTLC of andrographolide (AP) and 14-deoxy-11,12-didehydroandrographolide (DIAP) in the aerial parts of Andrographis paniculata Nees on silica gel with chloroform - methanol 4:1 with chamber saturation for 2 h. Quantitative determination at 254 nm. Good resolution of AP and DIAP was obtained together with symmetrical and reproducible peaks at Rf 0.55 and 0.43, respectively. Linearity is between 10 and 2000 µg/mL; LOD is 3.0 and 3.6 µg/mL; mean recoveries are 97.7 % and 97.8 % and precision analysis shows an intra-assay variation between 0.89 - 0.99 % and an inter-assay variation between 0.86 - 0.98 %. HPTLC method leads to accurate results when compared to the HPLC method.

traditional medicine, HPTLC, quantitative analysis, densitometry 32e

98 067 S. ANANDJIWALA, J. KALOLA, M. RAJANI* (*B. V. Patel Pharmaceutical Education and Research Development Centre, Pharmacognosy and Phytochemistry Department, Thaltej, Ahmedabad-380 054, Gujarat, India; rajanivenkat@hotmail.com): Quantification of eugenol, luteolin, ursolic acid, and oleanolic acid in black (Krishna Tulasi) and green (Sri Tulasi) varieties of Ocimum sanctum Linn. using high-performance thin-layer chromatography. J. Assoc. Off. Anal. Chem. 89, 1467-1474 (2006). HPTLC of eugenol, luteolin, ursolic acid, and oleanolic acid on silica gel in a twin trough chamber with toluene - ethyl acetate - formic acid 35:15:1 at 25 °C and 40 % relative humidity. Quantitation in absorbance mode at 280 nm for eugenol, at 350 nm for luteolin, and at 530 nm for ursolic acid and oleanolic acid after derivatization with anisaldehyde-sulfuric acid reagent and heating at 105°C. The methods were validated for precision, repeatability, and accuracy.

traditional medicine, herbal, HPTLC, densitometry 32e

98 068 S. BABU, K. KUMAR, G. SUBBARAJU* (*Laila Impex Research Center, Vijayawada, India, subbarajugottumukkala@hotmail.com) : Estimation of trans-resveratrol in herbal extracts and dosage forms by high-performance thin-layer chromatography. Chem. Pharm. Bull. 53, 691-693 (2005). HPTLC of trans-resveratrol in the roots of Polygonum cuspidatum and in dosage forms on silica gel with chloroform - ethyl acetate - formic acid 25:10:1. Quantitative determination by absorbance measurement at 313 nm. Linearity of determination of trans-resveratrol is between 0.5 and 3.0 µg with a correlation coefficient of 0.9989. LOD is 9 ng, and LOQ is 27 ng. The average percentage recovery is 99.9 - 100.7 %.

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

98 069 S. BADAMI*, M. GUPTA, S. RAMASWAMY, S. RAY, M. NANJAIN, D. BENDELL, R. SUBBAN, S. BHOJARAJ (*J.S.S. College of Pharmacy, Ootacamund, India, shribadami@yahoo.com): Determination of betulin in Grewia tiliaefolia by HPTLC. J. Sep. Sci. 27, 129-131 (2004). HPTLC of betulin in the bark of Grewia tiliaefolia Vahl. on silica gel with toluene - ethyl acetate 9:1. Quantitative determination by absorbance measurement. Linearity of determination of betulin is between 1000 and 1800 ng and its average percentage recovery is 96.09 - 98.87 %.

herbal, quality control, densitometry, quantitative analysis, HPTLC 32g

98 070 A. BAFNA, S. MISHRA* (*Department of Pharmacy, Faculty of Technology and Engineering, The M.S. University of Baroda, Gujarat, India, shmishra48@rediffmail.com) : Protective effect of bioactive fraction of Sphaeranthus indicus Linn. against cyclophosphamide induced suppression of humoral immunity in mice. J. Ethnopharmacol. 104, 426-429 (2006). HPTLC of the methanol fraction (bioactive fraction) of the fresh flower heads of Sphaeranthus indicus, on silica gel with three different solvent systems: 1) toluene - ethyl acetate 7:3; 2) ethyl acetate - methanol - water 200:27:20; and 3) n-butanol - glacial acetic acid 3:1:1. Detection by spraying with flavone reagent and qualitative determination by absorbance measurement at 254 nm and by fluorescence measurement at 366 nm. The compound having Rf 0.07 reacted to the reagent in solvent system 1. Two compounds at Rf 0.09 and 0.36 reacted in solvent system 2, whereas five compounds having Rf 0.11, 0.35, 0.38, 0.49 and 0.72 reacted to the reagent in solvent system 3.

herbal, traditional medicine, qualitative identification, HPTLC 32e

98 071 G. BALOGH, E. CSIZÉR, G. G. FERENCZY, Z. HALMOS, B. HERÉNYI, P. HORVATH, A. LAUKÓ, S. GÖRÖG* (*Chemical Works of Gedeon Richter Ltd. , P. O. B. 27, H-1475, Budapest, Hungary): Estimation of impurity profiles of drugs and related materials. 12. Isolation and identification of an isomeric impurity in danazol. Pharm. Research 12, 295-298 (1995). TLC of isodanazol according to The United States Pharmacopoeia XXII, 1990, pp. 379-380 and K. Ferenczi-Fodor, Z. Végh, Z. Pap-Sziklay, J. Planar chromatogr. 6, 198-203 (1993). Ferenczi-Fodor described a validated method for the selective determination of three individual impurities in danazol based on TLC-densitometric measurement at 252 nm. The Rf values of danazol and isodanazol in the TLC systems of USP XXII are 0.29 and 0.33, respectively, while in the system of Ferenczi-Fodor et al. 0.47 and 0.58.

quality control, densitometry 32a

98 072 R. BHUSHAN*, D. GUPTA, A. JAIN (*Department of Chemistry, Indian Institute of Technology Roorkee, Rorkee-247 667, India): TLC supplemented by UV spectrophotometry compared with HPLC for separation and determination of some antidiabetic drugs in pharmaceutical preparations. J. Planar Chromatogr. 19, 288-296 (2006). TLC of metformin hydrochloride, pioglitazone hydrochloride, rosiglitazone maleate, gliclazide, and glibenclamide on silica gel in a pre-equilib-

rated chamber at 26 - 30 °C with toluene - ethyl acetate - methanol 17:2:17 and n-butanol - acetic acid - water - methanol 12:4:1:2. Detection by exposure to iodine vapor. Highly reproducible RF and compact spots were obtained in normal-phase TLC which was found to be the least expensive method.

quality control, comparison of methods 32a

98 095 A. N. CAMPBELL, J. SHERMA* (*Dept. of Chem., Lafayette Col., Easton, PA 18042, USA) : Development and validation of a high-performance thin-layer chromatographic method with densitometric detection for determination of bisacodyl in pharmaceutical tablets. Acta Chrom. 13, 109-116 (2003). HPTLC of the laxative bisacodyl in enteric-coated tablets on silica gel with concentrating zone and 19 channels, pre-cleaned with dichloromethane - methanol 1:1, with ethyl acetate-methanol-glacial acetic acid 17:2:1. Quantitative determinaion by absorbance measurement at 254 nm. The method was validated and can be used for routine analysis of the pharmaceutical preparation in industry quality control and regulatory laboratories. An alternative extraction procedure and mobile phase are suggested for analysis of bisacodyl tablets with different formulations.

pharmaceutical research, quality control, HPTLC, qualitative identification, quantitative analysis, bisacodyl 32a

98 111 V.V. DIGHE, R. T. SANE, S.N. MENON, H. N. TAMBE*, S. PILLAI (*TDM Laboratories, Plot No. 194, Scheme No. 6, Road No. 15, Sion (E), Koliwada, Mumbai-22, India): High-performance thin-layer chromatographic determination of itopride hydrochloride in its pharmaceutical preparation and in the bulk drug. J. Planar Chromatogr. 19, 319-323 (2006). HPTLC of itopride hydrochloride on silica gel with methanol - ethyl acetate - toluene - triethylamine 2:5:12:1. Quantitative determination by absorbance measurement at 230 nm. The method was validated for accuracy and precision.

quality control, HPTLC, quantitative analysis, densitometry 32a

98 074 V.V. DIGHE, R. T. SANE, S. N. MENON, H. N. TAMBE*, S. PILLAI, V. N. GOKARN (*TDM Laboratories, Plot No. 194, Scheme No. 6, Road No. 15, Sion (E), Sion Koliwada, Mumbai-22, India): Simultaneous determination of diclofenac sodium and paracetamol in a pharmaceutical preparation and in bulk drug powder by high-performance thin-layer chromatography. J. Planar Chromatogr. 19, 443-448 (2006). HPTLC of diclofenac sodium and paracetamol (with aceclofenac as internal standard) on silica gel, pre-washed with methanol, in a presaturated twin-trough chamber with toluene - ethyl acetate - methanol - formic acid 50:40:10:1. Quantitative determination by absorbance measurement at 260 nm. The method was validated regarding accuracy and precision.

quality control, HPTLC, densitometry, quantitative analysis 32a

98 087 D. M. DIGREGORIO*, H. D. HARNETT, J. SHERMA (*Dept. of Chem., Lafayette College, Easton, PA 18042, USA): Quantification of dextromethorphan hydrobromide and clemastine fumarate in pharmaceutical caplets, gelcaps and tablets by HPTLC with ultraviolet absorption densitometry. Acta Chrom. 9, 72-78 (1999). The method is developed for determination of the cough suppressant dextromethorphan hydrobromide in multi-component flu-relief solid caplets and liquid gel caps, and of the antihistamine clemastine fumarate in tablets. HPTLC on silica gel (pre-washed with dichloromethane - methanol 1:1) with ethyl acetate - methanol - ammonia 17:1:2 for dextromethorphan hydrobromide or dichloromethane - methanol - ammonia 90:10:1 for clemastine fumarate. Densitometry at 225 nm for dextromethorphan hydrobromide and at 216 nm for clemastine fumarate. Multiple samples of the three dosage forms were analyzed to

confirm agreement between content of the active ingredients and label declarations.

pharmaceutical research, quantitative analysis, densitometry 32a

98 075 M. FAN (Fan Minwei), (Shanghai Municipal Inst. TCM, Shanghai 200127, China): (Study of the quality standard of Ruyi Jinhuang Tie ointment) (Chinese). J. Chinese Trad. Patent Med. (Zhongchengyao) 27(7), 864-867 (2005). TLC of extracts of Ruyi Jinhuang Tie ointment on silica gel with 1) n-butanol - glacial acetic acid - water 7:1:2; 2) petroleum ether (30 - 60 ºC) - ethyl formate - formic acid 15:5:1; 3) toluene - ethyl acetate - acetic acid 13:9:1; 4) chloroform - methanol - formic acid 96:3:1; 5) ethyl acetate - methanol - water 100:17:3 and toluene - ethyl acetate - formic acid water 20:10:1; 6) n-hexane - benzene - ethyl acetate 14:3:3. Detection under UV 365 nm and by spraying with 1 % FeCl3 solution in ethanol or with a solution of 5 % p-dimethylaminobenzaldehyde 10 % ethanolic H2SO4 followed by heating at 80 ºC. Identification of the fingerprint. Quantitative determination of berberine chloride by HPLC. The results of three batches of real life samples are given.

pharmaceutical research, traditional medicine, quality control, qualitative identification, berberine chloride 32c

98 076 A. FAYED*, M. SHEHATA, N. HASSAN, S. EL-WESHAHY (*Analytical Chemistry Department, Faculty of Pharmacy, Cairo University, Cairo, Egypt, fayedaeg@yahoo.com): Validated HPLC and HPTLC stability-indicated methods for determination of alfuzosin hydrochloride in bulk powder and pharmaceutical formulations. J. Sep. Sci. 29, 2716-2724 (2006). HPTLC of alfuzosin hydrochloride subjected to stress conditions (alkaline, acidic, oxidative, thermal, and photo-degradation) on silica gel with methanol - ammonia 125:3. Quantitative determination by absorbance measurement at 245 nm. Linearity of determination of alfuzosin is between 0.5 and 7.0 µg and its average percentage recovery is 98.5 - 101.8 %. The drug is well separated from its degradation products upon applying the two methods.

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

98 077 J. FISHER, J. SHERMA* (*Dept. of Chem., Lafayette Col., Easton, PA 18042, USA).: Analysis of carisoprodol tablets by HPTLC with visible absorbance densitometry. Acta Chrom. 11, 96-101 (2001). HPTLC of the muscle relaxant carisoprodol on silica gel uniplates with inorganic binder and fluorescent indicator, prewashed with dichloromethane - methanol 1:1, with chloroform - acetone 4:1 as mobile phase. Detection by spraying with conc. sulfuric acid - methanol 1:1 followed by heating at 150 ºC for 5 min. Quantitative determination by absorbance measurement at 550 nm. The method was applied to tablets containing carisoprodol as the only active ingredient and to tablets containing carisoprodol with aspirin and with aspirin plus codeine phosphate.

densitometry, quantitative analysis, HPTLC, carisoprodol 32a

98 078 Jolanta FLIEGER*, M. TATARCZAK (*Department of Inorganic and Analytical Chemistry, Medical University of Lublin, Staszica 6, 20-081 Lublin, Poland): Effect of inorganic salts as mobile-phase additives on lipophilicity values determined by reversed-phase thin-layer chromatography for new 1,2,4-triazole derivatives. J. Planar Chromatogr. 19, 386-392 (2006). TLC of 11 new 1,2,4-triazole derivatives on RP-18 with mixtures of methanol and water containing from 20 to 70 % methanol in steps of 10 %. Mobile phases containing different additives were prepared by adding the sodium salts of different anions (sodium hexafluorophosphate, perchlorate, trifluoroacetate, nitrate, chloride, iodide, and dihydrophosphate) to the mobile phase. Addition of iodide anions proved a key factor in obtaining lipophilicity indexes which correlated better with the log PG scale for all solutes investigated.

pharmaceutical research, qualitative identification 32a

98 065 S. A. GOSAVI, A. A. SHIRKHEDKAR*, Y. S. JAISWAL, S. J. SURANA (*Department of Pharmaceutical Chemistry, R. C. Patel College of Pharmacy, Karwand Naka, Shirpur-Dhule, M. S.-(425405), India): Quantitative planar chromatographic analysis of pantoprazole sodium sesquihydrate and domperidone in tablets. J. Planar Chromatogr. 19, 302-306 (2006). HPTLC of pantoprazole sodium sesquihydrate and domperidone on silica gel, prewashed with methanol, at 23 - 27 °C in a pre-saturated twin-trough chamber with methanol - water - ammonium acetate 8:2:1. Quantitative determination by absorbance measurement at 286 nm. The method was validated in accordance with ICH guidelines.

quality control, HPTLC, densitometry 32a

98 079 Z. HASSANKHANI-MAJD, V. GHOULIPOUR, S. W. HUSAIN* (*Anal. Lab., Dept. of Applied Chem., Fac. of Chem., Univ. of Tarbiat Moallem, 49 Mofatteh Avenue, Tehran-15614, Iran): Chromatographic behaviour of performance-enhancing drugs on thin layers of bismuth silicate ion exchanger. Acta Chrom. 16, 173-180 (2006). TLC of 14 performance-enhancing drugs (amphetamine, bemegride, caffeine, chlorphentermine, ephedrine, ethylamphetamine, isoproterenol, methadone, methyllendioxyamphetamine, pentazocine, pethidine, pemoline, strychnine and salbutamol) on bismuth silicate gel (prepared from 75 mL bismuth nitrate gel with 14 g silica gel powder) with thickness of 300 µm. 21 organic, aqueous and organic-aqueous mobile phases were investigated. Detection with iodine vapours.

doping, qualitative identification 32c

98 080 X. HOU (Hou Xiuzhen)*, X. QI (Qi Xihong) (*Ningxia Inst. Drug Cont., Yinchuan, Ningxia 750004, China): (Determination of betaine in Lycium barbarum L. by thin-layer chromatography) (Chinese). J. Chinese Trad. Patent Med. (Zhongchengyao) 27 (8), 941-944 (2005). TLC of betaine on silica gel with acetone - ethanol - hydrochloric acid 10:6:1. Detection by spraying with a solution of potassium iodobismuthate. Quantitative determination by densitometry at 515 nm. Validation regarding linearity range (8.0 - 39.0 µg, r = 0.9992), precision (RSD = 1.62 %, n = 6), reproducibility (RSD = 2.14 %, n = 6), and recovery (99.75 %, RSD = 1.92 %, n = 6). Results are given for 12 real life samples. Discussion of the advantages of the method compared to HPLC.

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

98 081 X. JIA (Jia Xiansheng)*, H. WU (Wu Hongfei), Q. MAO (Mao Qing) (*Guiyang Coll. TCM, Guiyang 550002, China): (Determination of fulvotomentoside A in Lonicera Fulvotomentosa Hsu et S. C. Cheng by thin-layer chromatography) (Chinese). Chinese J. Pharm. Anal. (Yaowu Fenxi Zazhi) 25 (6), 719- 721 (2005). TLC of fulvotomentoside A on silica gel with chloroform - methanol - water 61:32:5. Detection by spraying with 10 % H2SO4 in ethanol followed by heating at 105 ºC for 5 min. Quantitative determination by densitometry at 518 nm. The procedure was validated regarding linearity range (1.20 - 7.20 µg/spot, r = 0.998), repeatability (0.51 %, n = 5), precision (0.21 %, n = 5 within plate and 0.87 % n= 5 plate-to-plate), and recovery (99.7 %, RSD = 2.45 %, n = 5). The results are given for three batches of real life samples.

pharmaceutical research, herbal quality control, traditional medicine, qualitative identification, quantitative analysis, densitometry, fulvotomentoside A 32c

98 084 P. KANETKAR, R. SINGHAL, K. LADDHA, M. KAMAT* (*Food Engineering and Technology Department, Institute of Chemical Technology, University of Mumbai, Mumbai, India, mykamat@udct.org): Extraction and quantification of gymnemic acids through gymnemagenin

from callus cultures of Gymnema sylvestre. Phytochem. Analysis 17, 409-413 (2006). HPTLC of gymnemic acid as of gymnemagenin from callus cultures of Gymnema sylvestre on silica gel with chloroform - methanol 4:1. Quantitative determination by absorbance measurement at 205 nm. Linearity of determination of gymnemagenin is between 2 and 10 µg and its average percentage recovery from leave callus extracts is 98.6 - 99.2 %.

herbal, quantitative analysis, HPTLC 32e

98 085 N. KAUL, H. AGRAWAL, P. MASKE, J. RAO*, K. MAHADIK, S. KADAM (*Department of Quality Assurance Techniques, Bharati Vidyapeeth Deemed University, Maharashtra State, India, janhavirao@rediffmail.com) : Chromatographic determination of itopride hydrochloride in the presence of its degradation products. J. Sep. Sci. 28, 1566-1576 (2005). HPTLC of itopride (subjected to acid and alkaline hydrolysis, oxidation, dry and wet heat treatment, UV and photodegradation) on silica gel with toluene - methanol - chloroform 5:3:6 and 1 drop a of ammonia. Quantitative determination by absorbance measurement at 270 nm. The drug is well separated from its degradation products upon applying the two methods.

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

98 073 Y. C. KUO (Yuh-Chi Kuo), C. K. LU (Chung-Kung Lu), L. W. HUANG (Li-Wei Huang), Y. H. KUO (Yueh-Hsiung Kuo), C. CHANG (Chen Chang), F. L. HSU (Feng-Liu Hsu), T. H. LEE* (Tzong-Huei Lee) (*Graduate Institute of Pharmacognosy, Taipei Medical University, Taipei, Taiwan 110, Republic of China; e-mail: thlee@tmu.edu.tw): Inhibitory effects of acylated kaempferol glycosides from the leaves of Linnamomum kotoense on the proliferation of human peripheral blood mononuclear cells. Planta Med. 71, 412-415 (2005). TLC of kaempferol 3-O-alpha-L-[2-(Z)-p-coumaroyl-4-(E)-p-coumaroyl]rhamnopyranoside and kaempferol 3-O-alpha-L-[2,4-di-(E)-p-coumaroyl]rhamnopyranoside on silica gel with dichloromethane - methanol 6:1. Observation under UV light at 254 nm and dipping in vanillin - sulfuric acid resulting in yellow-green spots.

traditional medicine, herbal, qualitative identification 32e

98 086 L. LIU (Liu Lifang)*, SH. CAO (Cao Shuping), Y. JI (Ji Ying), X. ZHANG (Zhang Xiaojun), T. HUANG (Huang Ting) (*China Pharm. Univ., Nanjing 210038, China): Qualitative and quantitative analysis of aristolochic acids in Chinese materia medica and traditional Chinese patent medicines. J. Chinese Trad. Patent Med. (Zhongchengyao) 27 (8), 938-941 (2005). TLC on silica gel with ethyl acetate - water - formic acid 10:1:1. Detection under UV 254 nm. Quantification of aristolochic acid l and ll by HPLC with gradient elution. Results are given for six batches of real life samples.

pharmaceutical research, traditional medicine, quality control, qualitative identification, aristolochic acids 32c

98 088 X. MAO (Mao Xiuhong)*, K. WANG, SH. JI (Ji Shen) (*Shanghai Municipal Inst. Drug Cont., Shanghai 200233, China): (Study of quality standard for Wumei Rendan pills) (Chinese). J. Chinese Trad. Patent Med. (Zhongchengyao) 27 (8), 896-899 (2005). TLC of Wumei Rendan pills on silica gel with ethyl acetate - formic acid - water 2:1:1. Detection by spraying with 10 % ethanolic H2SO4 solution followed by heating at 105 ºC. Identification of the fingerprint. Quantification of menthol by GC, and of ammonium glycyrrhiziate by HPLC.

pharmaceutical research, traditional medicine, quality control, qualitative identification, menthol, ammonium glycyrrhiziate 32c

98 089 W. MARKOWSKI*, K. L. CZAPINSKA, G. MISZTAL, L. KOMSTA (*Department of Physical Chemistry, Medical University Lublin, Staszica St. 6, 20-081 Lublin, Poland): Analysis of some fibrate-type antihyperlipidemic drugs by AMD. J. Planar Chromatogr. 19, 260-266 (2006). AMD-HPTLC of bezafibrate, ciprofibrate, clofibric acid, clofibrate, fenofibrate, and gemfibrozil on diol phase with mixtures of tetrahydrofuran and hexane. An 25 % aqueous solution of acetic acid was used for preconditioning. Quantitative determination by absorbance measurement at 227 and 254 nm.

quality control, AMD, densitometry, HPTLC 32a

98 090 S. MENNICKENT*, M. VEGA, C. GODOY (*Departamento de Farmacia, Facultad de Farmacia, Universidad de Concepción, Casilla 237, Concepción, Chile, smennick@udec.cl): Development and validation of a method using instrumental planar chromatography for quantitative analysis of carbamazepine in saliva. J. Chil. Chem. Soc. 48, 71-73 (2003). HPTLC validation of carbamazepine in saliva samples on silica gel previously activated at 130 ºC for 20 min. Development over 5 cm in a saturated chamber with ethyl acetate - toluene - methanol 5:4:1. Detection by dipping in 60 % perchloric acid in ethanol - water 1:1, followed by heating at 120ºC for 7 min. Quantitative determination by fluorescence measurement at 366 nm. Linearity is between 0.5 and 15.0 ng per spot. The detection limit is 0.18 ng and the quantification limit is 0.54 ng. Precision: The analysis shows an intra-assay variation between 5.1 - 7.4 % and an inter-assay variation between 5.6 - 7.4 %. The method allows separation of carbamazepine from its main metabolites 10,11-dihydrocarbamazepine and carbamazepine-10,11-epoxide.

clinical routine analysis, HPTLC, densitometry, quantitative analysis 32c

98 091 S. MEYYANATHAN*, G. RAMASARMA, B. SURESH (*Department of Pharmaceutical Chemistry, J.S.S. College of Pharmacy, Ootacamund, Tamilnadu, India, meyys@rediffmail.com): Analysis of levofloxacin in pharmaceutical preparations by high performance thin layer chromatography. J. Sep. Sci. 26, 1698-1700 (2003). HPTLC of levofloxacin and lamotrigine (internal standard) on silica gel with water - methanol - n-butanol 1:1:1 and 1 drop of ammonia. Quantitative determination by absorbance measurement at 298 nm. Linearity of determination of levofloxacin is between 0.8 and 3.0 µg and its average percentage recovery is 99.9 %.

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

98 092 S. MEYYANATHAN*, P. KUMAR, B. SURESH (*J.S.S. College of Pharmacy, Ootacamund, Tamilnadu, India, meyys@rediffmail.com): Analysis of tramadol in pharmaceutical preparations by high performance thin layer chromatography. J. Sep. Sci. 26, 1359-1362 (2003). HPTLC of tramadol and chlorzoxazone (internal standard) on silica gel with ethyl acetate - methanol 7:1 and 1 drop of ammonia. Quantitative determination by absorbance measurement at 275 nm. Linearity of determination of levofloxacin is between 1.0 and 2.5 µg and its average percentage recovery is 104.6 %.

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

98 093 Zlata MRKVICKOVÁ*, P. KOVANKOVÁ, J. KLIMES, M. DOLEZAL (*Faculty of Pharmacy in Hradec Králové, Charles University in Prague, Heyrovského 1203, 500 05 Hradec Králové, Czech Republic): Determination of the lipophilicity of potential antituberculotic compounds by RP-TLC. J. Planar Chromatogr. 19, 422-426 (2006). TLC of 26 substituted anilides of pyrazine-2-carboxylic acids on silica gel impregnated with a solution of silicone oil in diethyl ether 19:1 for 16 h. The aqueous component of the mobile phases was 0.05 M phosphate buffer of pH 7.4 or 3.0; methanol was used as organic modifier; separation in a pre-saturated chamber. Detection under UV light at 254 nm. .

pharmaceutical research, qualitative identification 32a

98 094 Dragana MUTAVDZIC*, S.BABIC, D. ASPERGER, A. J. M. HORVAT, M. KASTELAN-MACAN (*Faculty of Chemical Engineering and Technology, Laboratory of Analytical Chemistry, Marulicev trg 19, 10000 Zagreb, Croatia): Comparison of different solid-phase extraction materials for sample preparation in the analysis of veterinary drugs in water samples. J. Planar Chromatogr. 19, 454-462 (2006). HPTLC of enrofloxazine, norfloxazine, oxytetracycline, trimethoprim, sulfamethazine, sulfadiazine, and penicillin G/procaine on cyano phase with 0.05 M oxalic acid - methanol 81:19. Evaluation under UV light at 254 and 366 nm. Quantitative determination of TMP by absorbance measurement at 254 nm, and fluorescence measurement at 366 nm for the other compounds. The SPE-TLC determination was validated for linearity, precision, quantification, and detection limit.

agricultural, HPTLC, densitometry 32a

98 096 N. NADER*, S. ESMAEILI, F. NAGHIBI, M. MOSADDEGH (*Traditional Medicine and Materia Medica Research Center, Shaheed Beheshti University of Medical Sciences, P. O. Box 14155-6354, Tehran, Iran): HPTLC determination of apigenin in some Iranian liquid products of Matricaria chamomilla L.. J. Planar Chromatogr. 19, 383-385 (2006). HPTLC of apigenin on silica gel, pre-washed with methanol, in a saturated twin-trough chamber with toluene - methanol 5:1. Densitometric evaluation at 343 nm.

herbal, traditional medicine, HPTLC densitometry, qualitative identification 32e

98 097 O. POZHARITSKAYA, S. IVANOVA, A. SHIKOV*, V. MAKAROV (*Interregional Center "Adaptogen", Std Petersburg, Russia, alexs79@mail.ru): Separation and quantification of terpenoids of Boswellia serrata Roxb. extract by planar chromatography techniques (TLC and AMD). J. Sep. Sci. 29, 2245-2250 (2006). HPTLC of four boswellic acids: 11-keto-beta-boswellic acid (1), acetyl-11-keto-beta boswellic acid (2), beta-boswellic acid (3), and acetyl-beta-boswellic acid (4) on silica gel with automated multiple development (AMD) using solvent gradients. Quantitative determination of 1 and 2 by absorbance measurement at 254 nm. 3 and 4 are quantified after derivatization with anisaldehyde sulfuric acid reagent at 560 nm. The AMD system provides good separation and the method is simple, precise, specific, sensitive, and accurate.

herbal, HPTLC, densitometry quantitative analysis, AMD, postchromatographic derivatization 32g

98 098 V. PURATCHIMANI, S. JHA* (*Department of Pharmaceutical Sciences, Birla Institute of Technology, Mesra, Ranchi-835215, India; puratchi_v@rediffmail.com): Standardisation of Gymnema sylvestre R. Br. with reference to gymnemagenin by high-performance thin-layer chromatography. Phytochem. Anal. 17, 164-166 (2006). HPTLC of gymnemagenin on silica gel with chloroform - methanol 9:1. Quantitative determination by absorbance measurement at 290 nm. Linearity of the determination of gymnemagenin was observed in the range of 4 - 10 µg. The average percentage recovery from an extract was 99.1 %, the content of leaves was 1.61 % (dry weight).

quality control, herbal, HPTLC, densitometry 32e

98 099 Alina PYKA*, M. BABUSKA, J. SLIWIOK (*Department of Analytical Chemistry, Faculty of Pharmacy, Silesian Academy of Medicine, 4 Jagiellonska Street, 41-200 Sosnowiec, Poland): Use of liquid chromatography and theoretical computational methods to compare the lipophilicity of selected cortison derivatives. J. Planar Chromatogr. 19, 432-437 (2006). TLC of cortisone derivatives (corticosterone acetate, 11-dehydrocorticosterone acetate, corticosterone, 11-dhydrocorticosterone, allo-diydrocortisone, hydrocortisone, and cortisone) on a mixture of silica gel and kieselguhr impregnated with a 10 % solution of paraffin oil in hexane. The plates were developed

with methanol - water 3:2. Quantitative determination by absorbance measurement at 254 nm.

pharmaceutical research, qualitative identification, densitometry, quantitative analysis 32a

98 100 S. RAI, A. WAHILE, K. MUKHERJEE, B. PADA, P. MUKHERJEE* (*School of Natural Product Studies, Department of Pharmaceutical Technology, Jadavpur University, Kolkata 700 032, India, pknatprod@yahoo.co.in): Antioxidant activity of Nelumbo nucifera (sacred lotus) seeds. J. Ethnopharmacol. 104, 322-327 (2006). HPTLC of Nelumbo nucifera seed extract (standardized 50 % hydro alcoholic extract, containing 30 % of saponins, strength 10:1) on silica gel with chloroform - methanol 7:1 and hexane - ethyl acetate 7:3. Qualitative determination at 254 nm reveals six zones (at Rf 0.19, 0.36, 0.40, 0.48, 0.61 and 0.74) with the first solvent system, and nine zones (at Rf 0.10, 0.15, 0.27, 0.39, 0.42, 0.51, 0.61, 0.77 and 0.85) with the second solvent system.

herbal, traditional medicine, HPTLC 32e

98 101 S. RAI, K. MUKHERJEE, M. MAL, A. WAHILE, B. SAHA, P. MUKHERJEE* (*School of Natural Products Studies, Department of Pharmaceutical Technology, Jadavpur University, Kolkata, India, pknatprod@yahoo.com): Determination of 6-gingerol in ginger (Zingiber officinale) using high-performance thin-layer chromatography. J. Sep. Sci. 29, 2292-2295 (2006). HPTLC of 6-gingerol in the rhizomes of Zingiber officinale on silica gel with n-hexane - diethyl ether 2:3. Quantitative determination by absorbance measurement. Linearity of determination of 6-gingerol is between 250 and 1200 ng and its average percentage recovery is between 99.79 - 99.84 %. The method permits a good resolution and separation of other constituents of ginger.

quality control, herbal, densitometry, quantitative analysis, HPTLC 32g

98 102 E. REICH*, V. WIDMER (*CAMAG Laboratory, Sonnenmattstrasse 11, 4132 Muttenz, Switzerland): HPTLC for rapid identification of Black Cohosh. LC-GC Europe 19, July 2006 Supplement, 15 (2006). HPTLC of Actaea racemosa (black cohosh) extracts on silica gel with toluene - ethyl formate - formic acid 5:3:2 in the CAMAG Automatic Development Chamber ADC 2 with humidity control (saturation 20 min, humidity 5 %). Detection by dipping in sulphuric acid reagent (20 mL of sulphuric acid in 180 mL methanol) followed by heating for 5 min at 100 °C. Evaluation at 366 nm. The method is specific for identification of black cohosh and for discrimination from different species of common adulterants.

herbal, quality control, qualitative identification, HPTLC, black cohosh 32e

98 103 P. RISHA, Z. MSUYA, M. NDOMONDO-SIGONDA, T. LAYLOFF* (*Management Sciences for Health, PO Box 50104, Dar es Salaam, Tanzania; tlayoff@msh.org): Proficiency testing as a tool to assess the performance of visual TLC quantitation estimates. J. Assoc. Off. Anal. Chem. 89, 1300-1304 (2006). TLC of 27 substandard drugs on silica gel with appropriate mobile phases. Routine Minilab (developed by the German Pharma Health Fund) test procedures to screen product quality and a proficiency testing program to determine the competency of health inspectors and reliability of results were established. Although the TLC screening methods provide a rapid means for drug quality assessment, they need to be put in the hands of competent users.

quality control, qualitative identification 32a

98 104 D. RUDDY*, J. SHERMA (*Dept. of Chem., Lafayette Col., Easton, PA 18042, USA): Analysis of the caffeine in alertness tablets and caplets by high-performance thin-layer chromatography with ultraviolet absorption densitometry of fluorescence-quenched zones. Acta Chrom. 12, 143-150 (2002). HPTLC of caffeine in pharmaceutical preparations on silica gel with ethyl acetate -

methanol 17:3. Densitometry at 275 nm. Tablet, coated tablet, and coated caplet products containing caffeine as the active ingredient were analyzed to test the applicability of the new method.

quantitative analysis, densitometry, HPTLC, caffeine 32a

98 105 T. S. REDDY, A. S. REDDY, P. S. DEVI* (*Analytical Chemistry Division, Indian Institute of Chemical Technology, Tarnaka, Hyderabad-500007, India): Quantitative determination of sildenafil citrate in herbal medicinal formulations by high-performance thin-layer chromatography. J. Planar Chromatogr. 19, 427-431 (2006). HPTLC of sildenafil citrate on silica gel, pre-washed with methanol, with toluene - acetone - methanol 3:1:1 in a saturated twin-trough chamber. Quantitative determination by absorbance measurement at 312 nm. The method was validated in accordance with ICH guidelines on the validation of analytical methods.

quality control, HPTLC, densitometry, quantitative analysis 32a

98 106 M. SAJEWICZ*, T. KOWALSKA (*Inst. of Chem., Silesian Univ., 9 Szkolna Street, 40-006 Katowice, Poland): On problems with liquid chromatographic quantification of chiral 2-arylpropionic acids by use of UV-absorbtion-based detection. Acta Chrom. 17, 292-301 (2006). TLC of ibuprofen and naproxen on silica gel F254 layer pre-washed with methanol - water 9:1 and dryed at ambient temperature for 3 h. The plates were impregnated with a 0.03 M solution of L-arginine in methanol. Development with acetonitrile - methanol - water 5:1:1 containing several drops of glacial acetic acid for S-(+)-ibuprofen, and acetonitrile - methanol - water 10:2:3 containing several drops of glacial acetic acid for S-(+)-naproxen. Densitometry at 200, 205, and 210 nm for S-(+)-ibuprofen and at 202, 215, and 225 nm for S-(+)-naproxen. The investigations were performed by using three independent measurement techniques, all based on UV absorption: HPLC-UV, HPLC-DAD, and TLC-densitometry.

comparison of methods, densitometry quantitative analysis, chiral, 2-arylpropionic acids, S-(+)-ibuprofen, S-(+)-naproxen 32a

98 114 M. Y. SALEM*, N. K. RAMADAN, A. A. MOUSTAFA, M. G. EL-BARDICY (*Cairo University, Faculty of Pharmacy, Analytical Chemistry Department, Kasr-El-Aini 11562, Cairo, Egypt; maissas@hotmail.com): Stability-indicating methods for the determination of disopyramide phosphate. J. Assoc. Off. Anal. Chem. 89, 976-985 (2006). TLC of disopyramide phosphate on silica gel with ethyl acetate - chloroform - ammonia 17:2:1 in a pre-saturated chamber. Detection under UV light at 254 nm. Quantitative determination by absorbance measurement at 268 nm.

quality control, densitometry, quantitative analysis, disopyramide phosphate 32a

98 107 Breda SIMONOVSKA, Marija SRBINOSKA, Irena VOVK* (*National Institute of Chemistry, Laboratory for Food Chemistry, Hajdrihova 19, SI-1000 Ljubljana, Slovenia): Analysis of sucrose esters-insecticides from the surface of tobacco plant. J. Chromatogr. A 1127(1-2), 273-277 (2006). HPTLC of sucrose esters from the surface of leaves of Nicotiana tabacum L. on silica gel with n-hexane - ethyl acetate 1:3. Detection by spraying with aniline-diphenylamine reagent. Identification by off-line TLC-MS. Quantitative determination of sucrose esters-insecticides by indirect estimation through TLC of sucrose obtained after alkaline hydrolysis. Application of the method in the screening of sucrose esters in plant extracts in laboratory and field experiments.

pharmaceutical research, HPTLC, ,qualitative identification quantitative analysis, leaf surface compounds, Nicotiana tabacum L. 32e

98 108 B. SUHAGIA, S. SHAH, I. RATHOD, H. PATEL*, D. SHAH, B. MAROLIA (*Department of Quality Assurance, L.M. College of Pharmacy, Gujarat, India, patelhary2001@yahoo.com). :

Determination of gatifloxacin and ornidazole in tablet dosage forms by high-performance thin-layer chromatography. Anal. Sci. 22, 743-745 (2006). HPTLC of gatifloxacin and ornidazole on silica gel with n-butanol - methanol 8:1 and 1 drop ammonia with chamber saturation for 45 min. Quantitative determination by absorbance measurement at 302 nm. Linearity of determination of gatifloxacin is 100 - 500 ng and of ornidazole 250 - 1250 ng, with a correlation coefficient of more than 0.9850. The intra-day and inter-day coefficients of variation are found to be in the range of 0.68 - 2.58 % and 0.37 - 3.62%, respectively.

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

98 109 C. SULLIVAN, J. SHERMA* (*Dept. Of Chem., Lafayette Coll., Easton, PA 18042-1782, USA): Determination of salicylamide in pharmaceutical tablets by high-performance thin-layer chromatography with ultraviolet absorption densitometry. Acta Chrom. 16, 153-163 (2006). HPTLC of salicylamide in diuretic tablets and pain-relief tablets on silica gel with concentrating zones (pre-washed with dichloromethane - acetone 1:1) with dichloromethane - acetone 4:1. Quantification by densitometry at 254 nm. The method is suitable for routine quality-control analysis of pharmaceuticals containing salicylamide.

quality control, HPTLC, quantitative analysis, densitometry, salicylamide 32a

98 082 A. K. TRIPATHI, R. K. VERMA, A. K. GUPTA, M. M. GUPTA*, S. P. S. KHANUJA (*Central Institute of Medicinal and Aromatic Plants, P. O. CIMAP, Lucknow-226015, India; guptammg@rediffmail.com): Quantitative determination of phyllanthin and hypophyllanthin in Phyllantus species by high-performance thin layer chromatography. Phytochem. Anal. 17, 394-397 (2006). HPTLC of phyllanthin and hypophyllanthin from Phyllantus species, e.g. Phyllantus amarus, on silica gel with hexane - acetone - ethyl acetate 37:6:4. Detection by spraying with vanillin in concentrated sulfuric acid and ethanol. Quantitative determination by absorbance measurement at 580 nm. Recovery of phyllanthin is 98.7 % and of hypophyllanthin 97.3 %.The method was validated and the peak purities and limits of detection and quantification were determined.

herbal, quantitative analysis, HPTLC 32e

98 110 K. TYRPIEN*, C. DOBOSZ, A. CHROSCIEWICZ, M. CIOLECKA, T. WIELKOSZYNSKI, B. JANSOKA, D. BODZEK (*Dept. of Chem., Fac. of Med., Med. Univ. of Silesia, Jordana 19, 41-808 Zabrze, Poland): Investigation of nicotine transformation products by densitometric TLC and GC-MS. Acta Chrom. 13, 154-160 (2003). Samples of nicotine were exposed to air and UV light and the products formed (nicotyrine, cotinine, and trans-3'-hydroxycotinine) were separated by TLC on RP-18 with acetonitrile - water 22:3 in a horizontal chamber. Visual evaluation under UV 254 nm, and under white light after derivatization with Dragendorff's reagent. Quantitative determination by absorbance measurement at 260 nm. GC-MS analysis was performed to confirm the identities of the nicotine transformation products.

qualitative identification, densitometry quantitative analysis, comparison of methods, nicotine 32d

98 083 J. K. VERMA*, A. V. JOSHI (*Department of Chemistry, S. J. Somaiya College of Science and Commerce, Vidyavihar, Mumbay 400077, India): Rapid HPTLC method for identification and quantification of curcumin, piperine and thymol in an ayurvedic formulation. J. Planar Chromatogr. 19, 398-400 (2006). HPTLC of curcumin, piperine, and thymol on silica gel, pre-washed with methanol, without chamber saturation with toluene - ethyl acetate - methanol 18:2:1. Quantitative determination by absorbance measurement at 420 nm for curcumin, 333 nm for piperine, and 277 nm for thymol. Limit of detection for curcumin was 25 ng, for piperine 5 ng, and for thymol 50 ng. Rapid identification of the three compounds is also possible by spraying the plate with anisaldehyde in sulfuric acid reagent.

traditional medicine, herbal, HPTLC densitometry, qualitative identification, quantitative analysis, Ayurveda 32e

98 112 M. WLODARCZYK, G. MATYSIK, W. CISOWSKI, M. GLENSK* (*Department of Pharmacognosy, Wroclaw Medical University, Nankiera 1, 50-140 Wroclaw, Poland): Rapid densitometric quantitative screening of the myricitrin content of crude methanolic extracts of leaves from a variety of Acer species. J. Planar Chromatogr. 19, 378-382 (2006). TLC and HPTLC of myricitrin on silica gel, pre-washed with methanol, in a horizontal chamber with chloroform -methanol - formic acid - water 10:4:1:0.95. Quantitative determination by absorbance measurement at 254 nm.

herbal, HPTLC, quantitative analysis, densitometry 32e

98 113 Magdalena WÓJCIAK-KOSIOR*, A. SKALSKA, G. MATYSIK, M. KRYSKA (*Medical Academy, Laboratory of Planar Chromatography, Department of Chemistry, Staszica 6, 20-081, Lublin, Poland; kosiorma@wp.pl): Quantitative analysis of phenobarbital in dosage form by thin-layer chromatography combined with densitometry. J. Assoc. Off. Anal. Chem. 89, 995-998 (2006). HPTLC of phenobarbital on silica gel, prewashed with methanol and then acetone, using dichloromethane - ethyl acetate - formic acid 95:5:1 in a horizontal chamber. Quantitation by densitometry in the absorbance/reflectance mode at 210 nm. The validity of the HPTLC-densitometric method was established through a study of linearity, sensitivity, accuracy, and reproducibility.

quality control, densitometry, HPTLC 32a

98 115 M. YONAMINE*, M. CORTEZ (*College of Pharmaceutical Sciences, Toxicology, University of S. Paulo, Av. Professor Lineu Prestes, 580 B13B, 05509-900 Sao Paulo, SP, Brazil, yonamine@usp.br): A high-performance thin-layer chromatographic technique to screen cocaine in urine samples. Leg. Med. 8, 184-187 (2006). HPTLC on silica gel of cocaine urine samples submitted to solid phase extraction prior to derivatization (methylation) with diazomethane. For methylation samples were mixed with 100 µL of a solution freshly prepared by distillation of 2.14 g N-methyl-N-nitroso-p-toluenesulfonamide with 10 mL potassium hydroxide 96 % in ethanol and 30 mL ethyl ether, and kept at room temperature for 1 min to convert benzoylecgonine to cocaine. Development over 7 cm in a saturated chamber with ethyl acetate - cyclo hexane - ammonia 250:100:1. Detection by spraying with Dragendorff reagent (10 mL of 40 % m/v potassium iodide in water; 10 mL of 1 N solution of bismuth nitrate in glacial acetic acid; 80 mL of 10 % v/v sulfuric acid water solution; 2 g of resublimed iodine). The technique is capable to discriminate cocaine from interfering substances such as nicotine, caffeine and even cocaethylene in urine samples. The limit of detection was 100 ng of cocaine.

toxicology, doping, HPTLC, densitometry, quantitative analysis 32d

98 116 Á. Z. DÁVID, E. MINCSOVICS, I. ANTAL, É. FURDYGA, Z. ZSIGMOND, I. KLEBOVICH* (*Semmelweis University, Department of Pharmaceutics, Högyes Endre Street 7, 1092 Budapest, Hungary): OPLC combined with NIR spectroscopy - a novel technique for pharmaceutical analysis. J. Planar Chromatogr. 19, 355-360 (2006). OPLC of paracetamol, acetylsalicylic acid and caffeine on HPTLC silica gel (prewashed with acetonitrile - water 17:3) with n-hexane - toluene - diethyl ether - glacial acetic acid - methanol 100:50:30:19:1 and toluene - diethyl ether - glacial acetic acid - methanol 50:30:19:1. Quantitative determination by absorbance measurement at 254 nm.

quality control, densitometry, quantitative analysis, OPLC 32a

33. Inorganic substances

98 117 E. ADAMEK (Silesian Acad. of Med., Fac. of Pharm., Dept. of General and Anal. Chem., Jagiellonska 4, 41-200 Sosnowiec, Poland): The influence of the stationary and the mobile phases on the TLC separation of selected metal-ion complexes with sodium diethyldithiocarbamate (Na-DDTC). Acta Chrom. 11, 196-203 (2001). The effects of the stationary and the mobile phases on chromatographic behaviour in the separation of water-insoluble complexes of diethyldithiocarbamate with Cd2+, Co2+, Cu2+, Fe2+, Hg2+, Mn2+, and Pb2+ have been studied by TLC with polar adsorbents (silica gel, previously activated for 40 min at 110 ºC, and neutral aluminum oxide 60 Type E, activated for 4 h at 150 ºC) with benzene - chloroform after chamber saturation for 2,5 h. The proportion of the components of the mobile phase was varied. Visualization by spraying with 5 % aqueous solution of CuSO4 to form a more durable yellow-green chelate (436 nm). The results obtained show that metal ions in the form of their chelates with DDTC can be separated under the presented conditions.

qualitative identification, metal-ion complexes, sodium diethyldithiocarbamate 33a

98 118 M. CURTUI, Maria-Loredana SORAN* (*National Institute of Research and Development for Isotopic nad Molecular Technology, 72-103 Donath Street, 400293 Cluj-Napoca, Romania): TLC separation of metal ions using di(n-butyl)dithiophosphoric acid and neutral organophosphorus ligands. J. Planar Chromatogr. 19, 297-301 (2006). TLC of U(VI), Th(IV), the lanthanides Ln(III), (La(III), Ce(III), Pr(III), Sm(III), Gd(III), Er(III)), Co(II), Ni(II), and Cu(II) on silica gel using mobile phases containing tributylphosphate, trioctylphosphine oxide (TOPO), and di(n-butyl)dithiophosphoric acid (HDBDTP). The results obtained showed that the retention factors of U(VI), Th(IV), and Ln(III) are enhanced when a mixture of HDBDTP and TOPO is used. TLC on silica gel with xylene - ethyl methyl ketone - N,N-dimethylformamide 16:2:1 in an unsaturated chamber. Detection with 0.05 % arsenazo(II) for U(VI), Th(IV), and Ln(III) and with 0.1 % rubeanic acid in ethanol for Co(II), Ni(II), and Cu(II).

33a

98 119 V. GHOULIPOUR, S. W. HUSAIN* (*Fac. of Chem., Univ. of Tarbiat Moallem, 49, Mofatteh Avenue, Tehran-15614, Iran): Inorganic ion-exchangers for quantitative TLC of toxic elements. V. Separation and determination of chromium (VI). Acta Chrom. 12, 170-176 (2002). Rapid and selective method for the separation and determination of Cr (VI) from Al (III), Cr (III), Mn (II), Fe (III), Co (II), Ni (II), Cu (II), Zn (II), Se (IV), Sr (II), Zr (IV), Cd (II), La (III), Ce(III), Hg(II), and U (VI). TLC on self-prepared titanic silicate ion-exchange plates with 0.4 M ammonium oxalate - 2.2 M aqueous ammonia 1:1. Detection of Cr (VI) by spraying with a saturated solution of diphenyl-carbazide in iso-propanol. Densitometry at 545 nm.

toxicology, quantitative analysis, toxic cations 33a

98 120 A. MOHAMMAD*, M. AJMAL, S. ANWAR (*Dept. of Appl. Chem., Z. H. College of Engineering and Technology, Aligarh Muslim Univ., Aligarh-202 002, India): Identification of Hg2+ and its separation from UO22+ and Fe3+ on impregnated silica gel layers with formic acid - sodium chloride mobile phases. Acta Chrom. 9, 113-122 (1999). Selective separation of Hg2+ from spiked sea and industrial waste water samples. TLC of some inorganic pollutants on silica gel impregnated with potassium thiocyanate (0.01 -2.0 M) 1:1 mixtures of sodium chloride (0.01 - 1.0 M) and formic acid (0.01 - 1.0 M) as mobile phases. Detection with 5 % potassium ferrocyanide in water (for Fe3+, Cu2+, UO22+, VO2+, and Tl+), 0.05 % dithizone in carbon tetrachloride (for Zn2+, Cd2+, Pb2+, Bi3+, Hg2+, Ag+, and Tl+), 1 % dimethylglyoxime in ethanol (for Ni2+ and Co2+), 0.1 % aluminum (for Al3+) and 0.01 % thorine (for Th4+ and Zr4+). The effect of surfactants, halides and oxyanions on the separation of Hg2+ from UO22+ and Fe3+ was examined.

The method was used to identify Hg2+, Pb2+, Cd2+, and Zn2+ in synthetic heavy metal sludge samples.

environmental, qualitative identification, inorganic pollutants 33a

98 121 A. MOHAMMAD*, M. P. A. NAJAR (*Anal. Res. Lab., Dept. of Appl. Chem., Z.H. Col. of Eng. and Techn., Aligarh Muslim Univ., Aligarh-202002, India): Quaternary separation of some transition metal chlorosulphates on mixed adsorbent layers with water as mobile phase. Quantitative determination of nickel chlorosulphate. Acta Chrom. 11, 154-170 (2001). Several stationary phase - mobile phase combinations were evaluated to identify suitable chromatographic systems for analysis of metal chlorosulphates. The effect of anionic species on the mobility and separation of metal chlorosulphates was examined. Quaternary separation of the chlorosulphates (Mn, Fe, Ni, and Cu or Zn) on silica gel - cellulose 2:1 with double-distilled water. Detection of Cu and Fe by spraying with 1 % aqueous potassium ferrocyanide, of Ni and Co with 1 %, and of Zn and Mn with 0.5 % dithizone in chloroform. For the quantitative determination of nickel the substances were extracted with distilled water, and buffer solution (1 M ammonium hydroxide - 1 M ammonium chloride 1:1) with bromo-pyrogallol red indicator (0.05 g in 100 mL ethanol - water 1:1) was added. The mixture was titrated with 0.05 M aqueous EDTA solution.

metal cations 33a

98 122 A. MOHAMMAD*, N. JABEEN (*Anal. Lab., Dept. of Appl. Chem., Zakir Hussain Col. of Eng. and Techn., Aligarh Muslim Univ., Aligarh-202 002, India): Reversed-phase chromatography of amines, phenols, and metal cations on silica layers impregnated with tributyl phosphate, using surfactant-mediated mobile phases. Acta Chrom. 13, 135-153 (2003). TLC of 17 metal cations (e.g. Cr3+ from Cr6+, Fe3+ from Mn2+ and Cr6+, VO2+ from Mn2+ and Cr6+) and 14 phenol derivatives (e.g. o-cresol from m-cresol, m-aminophenol from o-aminophenol) on silica gel impregnated with 0.001 M tributyl phosphate, with an 0.01 M aqueous micellar solution of N,N,N-cetyltrimethyl ammonium bromide as mobile phase. An aqueous solution (8.3 × 10-6 M) of the non-ionic surfactant, Brij-35 proved suitable for achieving good separations of 16 amines (e.g. p-dimethylaminobenzaldehyde from L-tryptophan, p-dimethylaminobenzaldehyde from indole) on silica layers impregnated with 0.001 M TBP. Visualization of amine or phenol spots by treatment with iodine vapour for 10 min. Metal ions were detected by spraying with the appropriate chromogenic reagent.

qualitative identification, metal cations, phenol derivatives, amines 33a, 7

98 123 A. MOHAMMAD*, R. YOUSUF, Y. HAMID (*Anal. Res. Lab., Dept. of Appl. Chem., Fac. of Eng. and Techn., AMU, Aligarh-202002, India): Thin layer chromatography of inorganic ions on blended inorganic ion-exchangers with tributyl phosphate - formic acid as mobile phase. Acta Chrom. 11, 171-182 (2001). The chromatographic behaviour of some cations (Ni2+, Co2+, Cd2+, Cu2+, UO22+, VO2+, Fe2+/3+, Al3+, Th4+, Mo6+, W6+, Pb2+, Hg+/2+, Bi3+, Ag+, and Tl+) and anions (CrO42-, Cr2O72-, IO3-, IO4-, BrO3- SCN-, Fe(CN)63-, NO2-) was examined on layers prepared from 1:9 mixtures of a synthetic inorganic ion-exchanger (stannic arsenate or tin(IV) molybdosilicate) with silica gel, alumina, or cellulose with 1 % methanolic tri-n-butyl phosphate - 1 M aqueous formic acid 1:4 as mobile phase. Several binary cation separations of analytical interest were achieved. The metal ions were detected by use of conventional spot-test reagents. Separation of IO3- from NO2- and BrO3- under the same conditions, detection with 0.2 - 0.5 % diphenylamine in 2 M H2SO4. For quantitative determination the region corresponding to IO3- was isolated from the plate and extracted with 1.0 M hydrochloric acid. 1 % potassium iodide and conc. hydrochloric acid were added to the extract and the mixture obtained was titrated against Na2S2O3.

qualitative identification, metal cations, anions 33a

98 124 A. MOHAMMAD*, S. SYED, L. M. SHARMA, A. A. SYED (*Dept. of Appl. Chem., Fac. of Eng. and Techn. A.M.U., Aligarh-202002, India): Thin layer chromatographic separation and recovery of gold and silver from secondary sources. Acta Chrom. 11, 183-195 (2001). TLC method for selective separation of Au3+ and Ag+ from accompanying metal ions (Cr6+, Ni2+, Cu2+, Zn2+, Cd2+, and Hg2+). TLC of Au3+ on silica gel G with aqueous 1.2 mM N-cetyl-N,N,N-trimethylammonium bromide and of Ag+ on alumina G layers with 2.5 M aqueous ammonium sulphate. Detection of Au3+ by spraying with 5 % aqueous stannous chloride, of Ag+, Cd2+, and Hg2+ by spraying with yellow ammonium sulphide, and of Cu2+ by spraying with 1 % aqueous potassium ferrocyanide. Ni2+ was detected by spraying with 1 % ethanolic dimethylglyoxime, Cr6+ by spraying with saturated ammonium thiocyanate in acetone, and Zn2+ by spraying with 0.05 % dithizone in CCl4. TLC in combination with spectrophotometry and volumetric analysis was used to determine content of Au3+ and Ag+ in secondary materials.

qualitative identification, cations 33a

98 125 A. MOHAMMAD*, V. AGRAWAL (*Anal. Research Lab., Dept. of Applied Chem., Fac. of Engineering and Technology, Aligarh Muslim Univ., Aligarh 202002, India): Use of cationic micellar mobile phases in normal-phase TLC for enhanced selectivity in the separation of transition metal ions. Simultaneous separation of mixtures of zinc, nickel, mercury and cadmium or manganese cations. Acta Chrom. 12, 177-188 (2002). The effect of surfactant concentrations below and above the critical micellar concentration on the retention behaviour of metal ions was examined. The effect of organic and inorganic additives on the mobility and separation efficiency of the metal ions was also assessed. TLC of mixtures of Zn2+, Ni2+, Hg2+, and Cd2+ or Mn2+ on silica gel with 50 mM aqueous micellar solution of N,N-cetyltrimethyl-ammonium bromide (CTAB). Detection of Ni2+ with a 1 % solution of alcoholic dimethylglyoxime, of Zn2+,Cd2+, and Hg2+ with a 0.5 % solution of dithizone in CCl4, and of Mn2+ with a 1:1 mixture of 2 M sodium hydroxide and 30 % H2O2. The proposed method was used for separation and identification of cations from drag-out nickel-plating solution and a sludge sample containing their hydroxides, as well as for semi-quantitative Ni2+-determination.

qualitative identification, micellar TLC, cations 33a

98 126 A. MOHAMMAD*, Y. H. SIRWAL (*Anal. Lab., Dept. of Applied Chem., Fac. of Engineering and Technology, Aligarh Muslim Univ., Aligarh-202002, India): Novel mobile phase for separation of Cr6+ from Cr3+ and associated heavy metal cations by high-performance thin-layer chromatography. Acta Chrom. 13, 117-134 (2003). HPTLC of 11 heavy metal cations on silica gel with pure organic, mixed organic and mixed aqueous - organic mobile phases. Mobile phases such as methanol - dimethylamine 4:1 and methanol - dimethylamine - formic acid 4:4:1 were found most suitable for rapid separation and identification of mixtures of Cr6+ and Cr3+ and of Cr6+, Ni2+ and Co2+, respectively. Detection of Cd2+, Ag+, Pb2+, Tl+, Bi3+ and Hg2+ by spraying with yellow ammonium sulphide reagent, of VO2+ with a 1 % aqueous solution of potassium ferrocyanide, of Ni2+ and Co2+ with dimethylglyoxime (0.2 % in ammonia), of Cr6+ with a saturated alcoholic solution of AgNO3 and of Cr3+ with a 1 % alizarin red in methanol. The effect of impurities such as inorganic ions, phenols, and surfactants on the separation of Cr6+ and Cr3+ was examined. The proposed method was successfully used for analysis of industrial wastewater samples.

environmental, qualitative identification HPTLC, metal cations 33a

35. Other technical products and complex mixtures

98 127 C. IMARK, M. KNEUBUEHL, S. BODMER* (*Biodyn GmbH, Industriestrasse 31, CH-8305 Dietlikon, Switzerland, bodmer@biodyn.ch) : Occurrence and activity of natural antioxidants in herbal spirits. Innovative Food Science and Emerging Technologies 1, 239-243 (2001). HPTLC

of commercial herbal spirits (alcoholic or hydroalcoholic solutions of volatile substances with flavoring or medicinal properties) and one red wine on silica gel with toluene - ethyl formate - formic acid 79:20:1. Antioxidative components were detected by dipping for 30 s in a soybean oil solution (3 % in n-hexane, previously treated with active carbon). Quantitative determination in UV light at 254 nm after different times of UV-exposure (30 min - 20 h). The antioxidant activity could be evaluated from the fluorescence-persisting time of the respective spots and was correlated with linoleic acid oxidation and DPPH-titration methods. Although the nature of the active herbal antioxidants remains to be established, phenolic compounds seem to be key candidates.

food analysis, herbal, HPTLC, quantitative analysis, densitometry, comparison of methods, post-chromatographic derivatization 35b

98 128 A. MOHAMMAD*, H. SHAHAB (*Anal. Res. Lab., Dept. of Applied Chem., Fac. of Eng. and Tech., Aligarh Muslim Univ., Aligarh-202002, India): Use of a glutamic acid-containing aqueous-organic mobile phase for on-plate separation, detection, and identification of cationic and non-ionic surfactants by thin-layer chromatography. Acta Chrom. 17, 272-291 (2006). TLC on silica gel with 0.1 M glutamic acid - methanol - acetone 1:1:1 has been found to be highly suitable for separation and identification of cationic and non-ionic surfactants. Visualization by use of Draggendorf reagent or iodine vapour. Spectrophotometric determination of tetradecyltrimethylammonium bromide at 670 nm after the spot extraction. The method has been used for identification of tetradecyltrimethylammonium bromide and Triton TX-100 in saline water, river water, and domestic waste water. The effects of sample pH, polarity of the alcohol and nature of the amino acid in the mobile phase, and the presence of alumina, kieselguhr, or cellulose in the silica gel layer have been examined.

qualitative identification, cationic and non-ionic surfactants 35a

98 129 T. WIDLA*, M. SLIWIOK (*Fac. of Law and Admin., Dept. of Criminalistics, Silesian Univ., 40-006 Katowice, Bankowa Street, Poland): Detection and determination of trotyl by HPTLC. Acta Chrom. 6, 113-115 (1996). HPTLC of trotyl on silica gel (activated at 110 °C) with hexane - benzene 1:1. Several selected visualizing agents were investigated: phenol red, bromphenol blue, thymol blue and bromothymol blue. After spraying, the plates were heated at 100 °C. A low detection limit (1 µg) was obtained by application of phenol red and bromphenol blue. This method enables further possibilities for quantitative determination.

HPTLC, qualitative identification, forensic application, trotyl 35

37. Environmental analysis

98 130 D. BODZEK*, C. DOBOSZ, K. TYRPIEN (*Dept. of Chem., Fac. of Med., Med. Univ. of Silesia, 41-808 Zabrze, Jordana 19, Poland): Determination of selected PAH carbonyl derivatives by TLC with densitometric detection. Acta Chrom. 11, 108-117 (2001). TLC of polyaromatic carbonyl compounds most commonly found in environmental samples (acridone, 1,2-naphthoquinone, 9,10-phenanthrenequinone, acenaphthenequinone, xanthone, 1-aminoanthraquinone, anthrone, 1,4-chrysenequinone, anthraquinone and 9-fluorenone) on silica gel and RP-18. Different combinations of solvents were evaluated as mobile phases. The best separation was obtained by use of pure dichloromethane on silica gel and methanol - water - acetonitrile 3:2:1 on RP-18 in a horizontal chamber. Evaluation under UV 254 nm and 366 nm. Quantitative determination of acridone by fluorescence measurement at 390/>400 nm, and by absorbance measurement at 250 nm for the other compounds.

environmental, densitometry, quantitative analysis, PAH, carbonyl derivatives 37c

38. Chiral separation

98 131 M. SAJEWICZ, H. E. HAUCK, G. DRABIK, E. NAMYSLO, B. GLÓD, Teresa KOWALSKA* (*Institute of Chemistry, Silesian University, 9 Szkolna Street, 40-006 Katowice, Poland): Tracing possible structural asymmetry of silica gel used for precoating thin-layer chromatography plates. J. Planar Chromatogr. 19, 278-281 (2006). Discussion of the deviation from the strict vertical of the migration tracks of chiral compounds during TLC on silica gel. Focus is put on possible crystalline asymmetry of the silica gel used in TLC. Description of an attempt to verify the hypothesis by means of circular dichroism spectroscopy. TLC of S-(+)-ibuprofen and S-(+)-naproxen on silica gel with acetonitrile - methanol - water 5:1:1 for S-(+)-ibuprofen and 10:2:3 for S-(+)-naproxen. Quantitative determination by absorbance measurement at 210 nm.

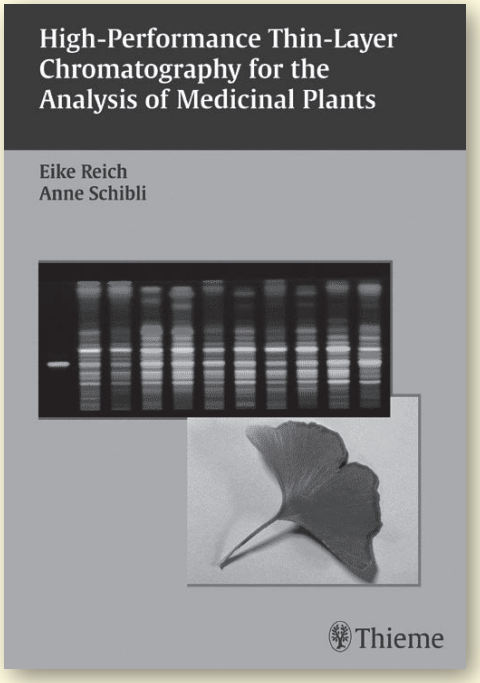
quantitative analysis, densitometry 38

98 132 M. SAJEWICZ, R. PIETKA, G. DRABIK, E. NAMYSLO, Teresa KOWALSKA* (*Institute of Chemistry, Silesian University, 9 Szkolna Street, 40-006 Katowice, Poland): On the stereochemically peculiar two-dimensional separation of 2-arylpropionic acids by chiral TLC. J. Planar Chromatogr. 19, 273-277 (2006). TLC of the 2-arylpropionic acids S-(+)-ibuprofen, S,R-(+/-)-ibuprofen, S-(+)-naproxen, and S,R-(+/-)-2-phenylpropionic acid on silica gel pre-developed with methanol - water 9:1 and impregnated with L-arginine in the cationic form as chiral ion-pairing reagent. Acetonitrile - methanol - water 5:1:1 (with several drops of glacial acetic acid to fix the pH at < 4.8) was used for ibuprofen, 10:2:3 for naproxen and 20:4:3 for 2-phenylpropionic acid. Quantitative determination by absorbance measurement at 210 nm.

qualitative identification, densitometry 38

98 133 J. SZULILK*, A. SOWA (*Inst. of Chem., Silesian Univ., 9 Szkolna Street, 40-006 Katowice, Poland): Separation of selected enantiomers of fatty hydroxyl acids by TLC. Acta Chrom. 11, 233-237 (2001). Conditions for separation of racemic mixtures of fatty acids by use of a mobile phase containing a chiral compound as additive and on a chiral stationary phase. TLC separation of the enantiomers of DL-alpha-hydroxypalmitic acid on silica gel with acetone - hexane - hydrochloric acid 3:5:2 containing 1 % L-alanine, detection by dipping in 2 % aqueous sodium hydroxide solution. TLC of the enantiomers of DL-12-hydroxystearic acid on silica gel F254 with hexane - acetonitrile - acetic acid - hydrochloric acid 5:2:1:2 containing 2 % L-alanine, detection by dipping in 2 % aqueous sodium hydroxide solution. Separation of the enantiomers of DL-12-hydroxyoleic acid on chiral plate with acetone - water 3:2, detection by treatment with iodine vapor. Isomers were characterized by calculation of their optical topological indexes.

qualitative identification, fatty acids, enantiomers 38



E. Reich, A. Schibli

High-Performance Thin-Layer Chromatography for the Analysis of Medicinal Plants

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43 references

Key areas

1. Medical Plants-Analysis
2. Herbs-Analysis
3. Thin-Layer Chromatography
4. Plant Extracts-Analysis
5. Plants, Medicinal-Chemistry
6. Chromatography

This is a book one can fall in love with it reading it from page to page. Well somebody will skip theory. But all practical aspects are dealt with completely, carefully, easy to understand and up to the level where it could be reproduced in practice by the reader.

The book as such is of high production quality cleanly printed on top quality paper throughout the whole volume.

Colour is intensively used for a top level of organization, a truly great help for the normally information overloaded reader. The HPTLC graphics are extremely well reproduced with respect to their original life colour. All technical drawings are excellent done and organized. They contain the messages in a high level of communication to the reader. In summary to what one sees at first: a book of first class, rarely seen elsewhere.

Quantitative data

Introduction - short but concise	7 % of the total content
Theoretical concepts	15 %
Practical aspects of modern TLC	23 %
Typical applications in herbal analysis	12 %
Method development	2 %
Validation	8 %
Appendices in sum	6 %

Definitions - Standard operating procedure for HPTLC of herbal raw materials - Resources - Common mobile phases for the screening of unknown herbal drugs - most common non specific derivation reagents for analysis of herbal drugs – Worksheet - Validation of method for identification of eleuthero by HPTLC fingerprint.

Index	0.4 % (5 pages).

Some qualitative data – analyzing the content for applicability

The authors avoided to show the beauty of the analysis of Medicinal Plants only, although just the cover page picture invites analysts to immediately start with HPTLC. On page 181 he/she may see what may be ahead of his/heir work in case one is confronted with "unknown botanicals". Under "Common Mobile Phases for the Screening of Unknown Herbal drugs" the reader finds more than 25 precisely and quantitatively given mobile phase mixtures for 10 substance classes which may help him to succeed in an own case. On page 185 he finds a table of miscibility of various mobile phases and sample solvents he might not have in easy reach in his library. Under "Most common Non specific Derivatization Reagents for Analysis of Herbal Drugs" the reader finds four full pages in tabular order "Reagent name/Preparation, use/Examination/Detection of". Again, there is a precise quantitative receipt what to mix, what to check for, what to avoid, how to use. Thus there is immediate help again not so easy to find elsewhere. Reichs/Schiblis book can really be used the classical way: to read, to understand, to do. There is no electronic full text search and text analysis necessary. This book is made for being kept open at the working place.

However it is also highly qualified to inform, teach and change minds. Analysts who know HPLC only should fear to inject any plant extract into their expensive column, especially in the case of HPLC x HPLC for comprehensive separations (which they think it will solve all problems). These HPLC-only analysts may dramatically change their mind. Thus this book is dangerous for them.

Professor Dr. Rudolf E. Kaiser

Institute for Chromatography
Bad Dürkheim

Chlorfreie Fliessmittel für die Bestimmung von PAK in Wasserextrakten

13

▲ *Dr. Holger Hegewald*

Dr. Holger Hegewald* ist Leiter eines Analytiklabors in Évora, Portugal. Für die Analytik von Pestiziden, pflanzlichen Hormonen, Aflatoxin M1 in Milch, Anthocyanen und organische Säuren in Wein sowie polycyclischen aromatischen Kohlenwasserstoffen (PAK) in Wasser setzt er ausschliesslich die Planar-Chromatographie ein. In CBS 95 hat er die zeitsparende Derivatisierung von Glyphosat und AMPA in der Startzone aufgezeigt – in diesem Beitrag stellt er uns eine chlorfreie Alternative zur Bestimmung der PAK vor.

Einleitung

Die Methode orientiert sich an der DIN 38407-7 [1], die die quantitative Bestimmung der 6 PAK auf koffeinimprägnierten HPTLC-Platten mit Dichlormethan bei -20 °C anwendet.

Bei der Auswahl der Fliessmittel sollte an erster Stelle die Trennqualität stehen, aber es sollte auch deren Gesundheits- und Umweltschädlichkeit in Betracht gezogen werden. So wird allgemein der Ersatz des cancerogenen und mutagenen Benzens durch Toluen empfohlen, welches der gleichen Selektivitätsgruppe nach Snyder angehört [2]. Die umweltrelevanten chlorhaltigen Lösungsmittel, die in Verdacht stehen, ebenfalls cancerogen zu sein, sind aber nicht so einfach zu ersetzen, da z. B. Chloroform, ausser Wasser, der einzige Vertreter der Selektivitätsgruppe VIII ist und bei Dichlormethan in der Selektivitätsgruppe V keine chlorfreien Lösungsmittel existieren.

Mit der Verwendung von Isopropylacetat anstelle von Dichlormethan wurde ein chlorfreies Fliessmittel gefunden, das Trennungen hoher Qualität mit günstiger Basislinienform und Matrixretardation liefert und somit für die quantitative Auswertung der Chromatogramme geeignet ist.

Für den qualitativen Nachweis der PAK (Screening-Verfahren) wurde die Trennung mit Isopropylacetat – n-Hexan (3:1, v/v) in der Horizontalentwicklungskammer bei Zimmertemperatur durchgeführt.

Probenvorbereitung

Die Wasserproben werden mittels Festphasenextraktion an C18-Sorbens extrahiert und die adsorbierten PAK mit 3 mL Dichlormethan eluiert. Nach Zusatz des internen Standards (IS) 2-Methylanthracen (2-MA) werden die Extrakte auf etwa 0,1 mL mit Stickstoff eingeengt.

Standardlösung

Verdünnung der zertifizierten Standardlösungen (10 ng/µL) mit Methanol, um eine Standardmischung wie in der Tabelle angegeben zu erhalten:

Substanz	Konzentration (ng/µL)
Benzo(a)pyren	0.1
Benzo(ghi)perylen	0.1
Benzo(b)fluoranthen	0.2
Benzo(k)fluoranthen	0.2
Indeno(1,2,3cd)pyren	0.2
Fluoranthen	0.5
2-Methylanthracen (IS)	1.5

Schicht

HPTLC-Platte Kieselgel 60 F_{254} coffeinimprägniert für die PAK-Bestimmung (Merck) oder Nano-SIL-PAH (Macherey-Nagel), 20 x 10 cm.

Probenauftragung

Bandförmig mit Linomat, 17 Bahnen, Auftragevolumen 1–12 µL der Standardlösung und etwa ein Drittel der Probenextrakte, Bandlänge 7 mm, Bahnabstand 10 mm, seitlicher und unterer Randabstand 20 bzw. 8 mm, Auftraggeschwindigkeit 7 s/µL. Nach

dem Auftragen werden die Startzonen 2 min im kalten Luftstrom getrocknet.

Zum Screening in der Horizontalentwicklungskammer können auch auf der gegenüberliegenden Plattenseite Standards und Proben für die Entwicklung von zwei Seiten aufgetragen werden, insgesamt 34.

Chromatographie

Quantitative HPTLC: In einer vorgekühlten (–20 °C, 30 min) Doppeltrogkammer mit 8 mL Isopropylacetat in einer Troghälfte, ohne Kammersättigung. Die getrocknete Platte wird zunächst in der fliessmittelfreien Troghälfte 10 min bei –20 °C äquilibriert. Start der Chromatographie durch Positionierung der Platte in der fliessmittelhaltigen Troghälfte. Laufstrecke vom unteren Plattenrand 70 mm (Laufzeit 25 min).

Qualitative HPTLC: Bei Zimmertemperatur in der Horizontalentwicklungskammer mit Isopropylacetat – n-Hexan 3:1 (v/v), bei Bedarf von beiden Seiten, S-Konfiguration. Laufstrecke vom unteren Plattenrand 50 mm (Laufzeit 9 min).

Bei beiden: Nach dem Trocknen (2 min im kaltem Luftstrom) wird zur Fluoreszenzverstärkung, vor allem der 3 dunkelblauen Zonen, die Platte mit der Tauchvorrichtung in Paraffin-Toluen 1:1 getaucht (Tauchgeschwindigkeit 4 cm/s, Eintauchzeit 1 s).

Densitometrische Auswertung

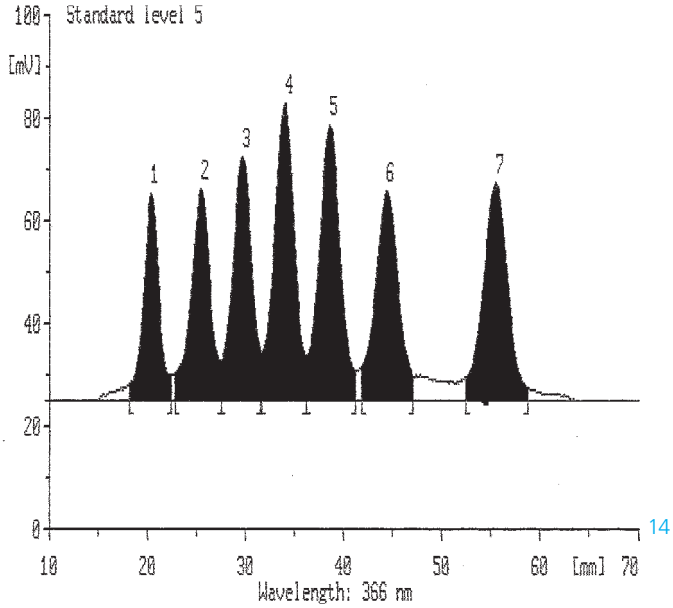
TLC-Scanner mit CATS-Software, Fluoreszenzmessung bei UV 366/>400, lineare Kalibration über die Peakhöhe.

Dokumentation

Mit DigiStore 2 Dokumentationssystem, Aufnahme bei UV 366/>400 nm

Ergebnisse und Diskussion

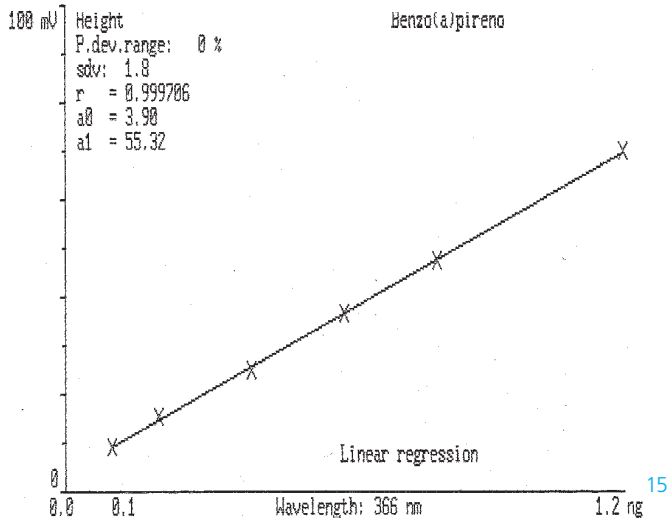
Quantitative HPTLC: Nachfolgendes Densitogramm zeigt die Trennung von 10 µL Standardmischung. Die Sequenz der PAK ist die gleiche wie bei der HPTLC mit Dichlormethan nach DIN 38407-7 und in der Trennqualität mit jener vergleichbar.



14

▲ *Quantitative HPTLC: Densitogramm einer Standardbahn (1–5 ng/Band, IS 15 ng/Band) 1 Benzo(ghi)perylen (BP), 2 Indeno(1,2,3-cd)pyren (IP), 3 Benzo(a)pyren (BaP), 4 Benzo(b)fluoranthen (BbF), 5 Benzo(k)fluoranthen (BkF), 6 Fluoranthen (F), 7 2-Methylanthracen (2-MA) as IS*

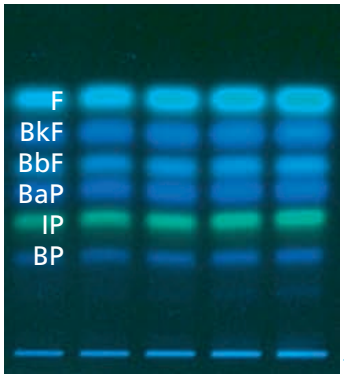
Die Kalibration – hier am Beispiel von BaP im Arbeitsbereich von 0.1–1.2 ng/Band – ist linear mit einer relativen Standardabweichung (sdv) von ±1.8 % (r > 0.9997).



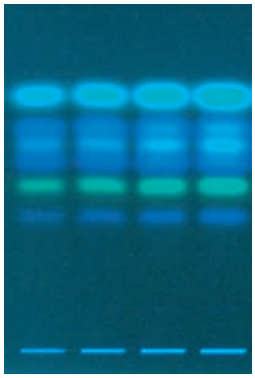
15

▲ *Lineare Kalibration von BaP im Bereich von 0.1-1.2 ng /Zone*

Die beiden hier vorgestellten Chromatographiesysteme scheinen gegen Störungen durch Matrixbestandteile weniger empfindlich zu sein als die Trennung mit Dichlormethan.



16a


16b

▲ *Quantitative (links) und qualitative HPTLC (rechts): Plattenausschnitte mit Standardbahnen (2–12 ng/Band, für die bildliche Darstellung wurden höhere Mengen gewählt) unter Beleuchtung UV366 nm (hier ohne Peak 7, dem IS 2-Methylanthracen)*

Das qualitative System mit Isopropylacetat – n-Hexan hat darüberhinaus den Vorteil, alle sechs PAK für die visuelle Auswertung unter UV 366 nm ausreichend gut zu trennen, während auf den in DIN 38407-7 angegeben Systemen nur bis zu 5 Banden erkennbar sind.

Unterschiede in der Qualität der Densitogramme konnten aufgrund der selektiven Fluoreszenzdetektion nicht feststellt werden, je nachdem ob die Platten vorgewaschen wurden oder nicht. Deshalb wurde auf eine Vorreinigung der Platten verzichtet.

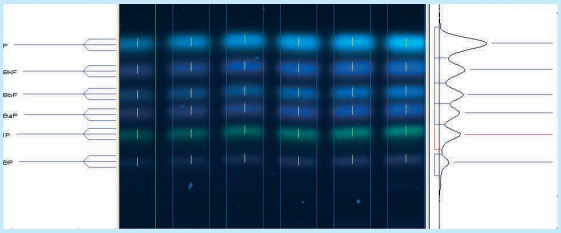
Anmerkung des Herausgebers: Verwendet man Isopentylacetat statt Isopropylacetat, so duftet es im Labor nach Bananen.

Weitere Informationen sind beim Autor auf Anfrage erhältlich.

*Dr. Holger Hegewald, Lacrome Lda, Rua César Batista 6 D, P-7000-715 Évora, Portugal, lacrome@clix.pt

[1] Deutsches Institut für Normung: DIN 38407–7, Bestimmung von sechs polycyclischen aromatischen Kohlenwasserstoffen mittels HPTLC, Beuth Verlag, Berlin 2000

[2] Snyder. L.R., J. Chromatogr. Sci. 16, 223-234, 1978


17

Digitale Bildauswertung VideoScan

Für das PAK-Screening kann die Platte nach Aufnahme mit dem DigiStore 2 Dokumentationssystem mit hochaufösender 12 bit CCD Kamera visuell ausgewertet werden. Zur schnellen Quantifizierung der Wasserproben in Grenzwertnähe kann die digitale Bildauswertung VideoScan eingesetzt werden. Das Programm entspricht den Anforderungen von GMP/GLP und ist IQ/OQ qualifizierbar. Es bietet flexible Anwendungen, z. B. Vergleich von Bahnen verschiedener Chromatogramme und Auswertung von Bahnen mit variierendem Abstand oder schräg liegenden Messbahnen. Die Chromatogramme können zu beliebiger Zeit, auch noch Jahre nach der Bilderfassung, ausgewertet werden.

Die wichtigsten Eigenschaften des VideoScan-Programms in Kürze:

- Einfach und schnell in der Bedienung
- Die Integration der Analogkurven kann wahlweise automatisch oder manuell durchgeführt werden.
- Die quantitative Auswertung erfolgt wahlweise über Peakhöhe und/oderPeakfläche.
- Es besteht die Auswahl zwischen Einstandard- und Mehrbereichs-Kalibrierung (linear oder polynom).

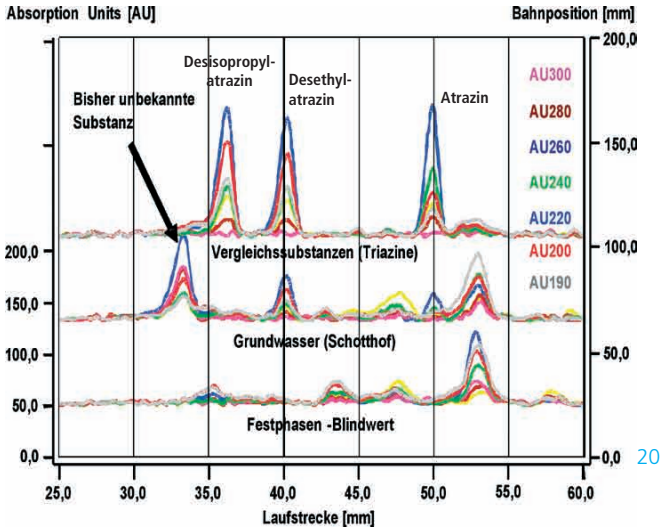
Nachweis der Metaboliten Desphenyl-chloridazon und Methyl-desphenyl-chloridazon in Oberflächen-, Grund- und Trinkwasser

18 ▲ *Dr. Walter H. Weber**

19 ▲ *Dipl.-Ing. (FH) Wolfram Seitz, Dr. Wolfgang Schulz, Anna Aichinger*

Im Betriebs- und Forschungslaboratorium des Zweckverbandes Landeswasserversorgung (LW), das bereits im CBS 94 bzw. 95 vorgestellt wurde, konnten bei der routinemässigen Ressourcenüberwachung im Rahmen des »multidimensionalen Screenings« zwei bis dato nicht bekannte Kontaminanten mittels HPTLC/AMD-UV/VIS erkannt werden [1]. Zur Strukturaufklärung wurde die gefundene Zone mittels der Extraktionsapparatur ChromeXtract® (siehe CBS 93, 94 und 96) der Tandem- bzw. Flugzeit-Massenspektrometrie zugeführt, und es konnten die entsprechenden, protonierten Moleküle der Metaboliten Desphenyl-chloridazon und Methyl-desphenyl-chloridazon mittels HPTLC/MS nachgewiesen werden. Dieses multidimensionale Überwachungskonzept basiert auf der Philosophie, neben dem Einsatz der üblichen Methoden zur Targetanalyse (GC und HPLC) auch organismische Tests zur Beurteilung der biologischen Wirkung einer Probe bzw. deren einzelner oftmals unbekannter Probeninhaltsstoffe anzuwenden. Dabei ist die HPTLC mit automatisierter Mehrfachentwicklung (AMD) von zentraler Bedeutung.

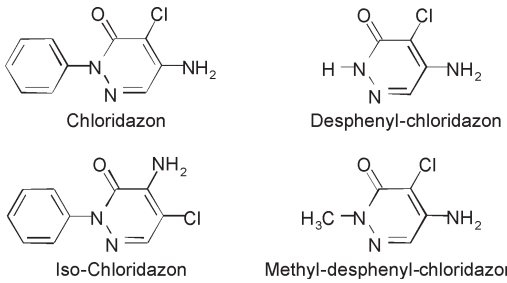
Dieses multidimensionale Überwachungskonzept basiert auf der Philosophie, neben dem Einsatz der üblichen Methoden zur Targetanalyse (GC und HPLC) auch organismische Tests zur Beurteilung der biologischen Wirkung einer Probe bzw. deren einzelner oftmals unbekannter Probeninhaltsstoffe anzuwenden. Dabei ist die HPTLC mit automatisierter Mehrfachentwicklung (AMD) von zentraler Bedeutung. Für die Identifizierung von unbekannten, jedoch toxischen Substanzen ist die HPTLC/MS-Kopplung unentbehrlich.



20 ▲ *AMD-Monitoring (aufgenommen im Mehrwellenlängenscan) im Rahmen des »multidimensionalen Screenings«*

Einleitung

Bei dem Wirkstoff Chloridazon handelt es sich um ein in Deutschland zugelassenes systemisches Herbizid, das zur Kontrolle von Unkräutern beim Anbau von Zuckerrüben, Futterrüben und roten Rüben eingesetzt wird. Desphenyl-chloridazon ist der Primärmetabolit dieses Herbizid-Wirkstoffes und wurde bisher nicht bei der Untersuchung von Oberflächen-, Grund- und Trinkwässern berücksichtigt. Methoden zur Bestimmung der Abbauprodukte sind bisher noch nicht bekannt. In der vorliegenden Arbeit wird ein Verfahren zum Nachweis von Chloridazon und dessen Abbauprodukten unter Verwendung der HPTLC/AMD-Technik beschrieben.



▲ *Strukturformeln des Herbizids Chloridazon, seiner Metaboliten Desphenyl-chloridazon und Methyl-desphenyl-chloridazon sowie von Iso-Chloridazon*

Probenvorbereitung

Zur Anreicherung der Analyten kam die Festphasen-Extraktion (SPE) zum Einsatz, wobei 1 L Wasserprobe mit Schwefelsäure auf pH 3 eingestellt und über das Sorbens geleitet wurde. Die Reinigung und Konditionierung der Kartuschen (200 mg Sorbens, z.B. SDB1, Mallinckrodt-Baker bzw. Isolute ENV+, Separtis) erfolgte mit jeweils 6 mL n-Hexan, Aceton, Methanol und zweimal 6 mL MilliQ-Wasser (pH 3). Zur Elution wurden 4 mL Methanol eingesetzt. Nach dem Einengen im Stickstoffstrom wurde der Rückstand mit 200 µL Methanol aufgenommen. Bei stärker matrixbelasteten Wässern (z.B. bei verschiedenen Oberflächenwässern) ist es notwendig, weitere Aufreinigungsschritte zum Beispiel mittels GPC durchzuführen.

Standardlösung

Die Referenzsubstanzen von Chloridazon, Desphenyl-chloridazon, Iso-Chloridazon (Dr. Ehrenstorfer) bzw. Methyl-desphenyl-chloridazon (BASF) wurden jeweils in Methanol gelöst und bei -20 °C aufbewahrt. Die Konzentration je Analyt beträgt ca. 100 mg/L.

Schicht

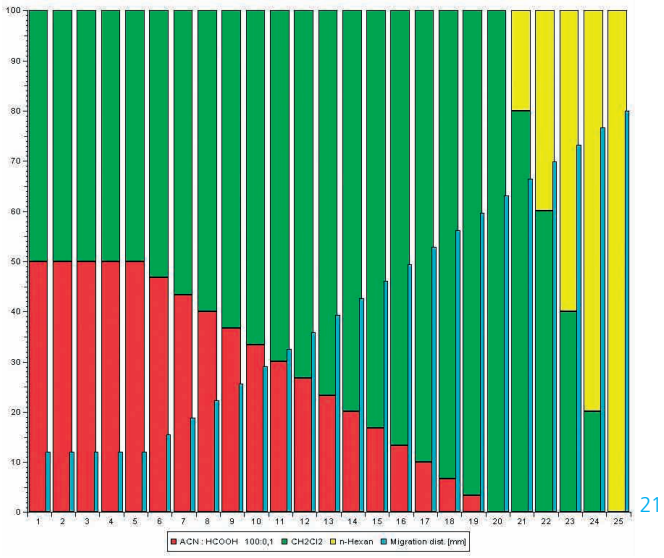
HPTLC-Platte Kieselgel 60 F_{254} (Merck), 20 × 10 cm

Probenauftragung

Mit dem DC-Probenautomat 4, 16 Bahnen, Bandlänge 6 mm, Bahnabstand 10 mm, Abstand vom unteren Plattenrand 8 mm, Abstand von den beiden Seiten mind. 20 mm

Chromatographie

Im AMD2-System mit einem 25-stufigen Gradienten, basierend auf Dichlormethan. Als elutionsverstärkendes Lösungsmittel wird bei den ersten Entwicklungsläufen mit Ameisensäure sauer überlagertes Acetonitril 100:0,1 (v/v) eingesetzt. Als elutionssenkendes Lösungsmittel wird n-Hexan verwendet. Die ersten 5 Entwicklungsläufe werden jeweils bis zu einer Laufstrecke von 12 mm ausgeführt, während die folgenden 21 Entwicklungsschritte äquidistant um 3,4 mm verlängert werden. Die gesamte Laufstrecke liegt bei 80 mm, und die insgesamte Entwicklungszeit beträgt 4,5 Stunden.



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▲ *25-stufiger AMD2-Gradient, basierend auf Dichlormethan*

Derivatisierung

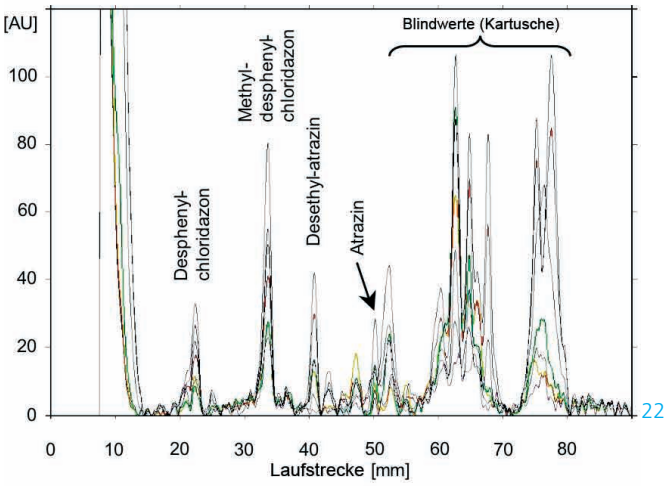
Zur postchromatographischen Derivatisierung wird die HPTLC-Platte zunächst salzsauer bedampft, dann mit NO_X zur Bildung des Diazoniumkations begast und zur Azokupplung anschliessend in eine Lösung aus 0,2 g Bratton-Marshall-Reagenz (N[1-Naphthyl]-ethylendiamindihydrochlorid) in 100 mL Methanol – Dichlormethan 1:4 (v/v) mittels der Tauchvorrichtung III getaucht (3 cm/s, 3 s). Es entstehen aus den primären aromatischen Aminen farbige Azoverbindungen, d.h. rotviolette Zonen auf farblosem Untergrund. Zur Stabilisierung des Azofarbstoffes empfiehlt sich ein anschliessendes Bedampfen mit Ammoniak.

Densitometrische Auswertung

Absorptionsmessung (Mehrwellenlängen-Scan) im UV-Bereich zwischen 190 und 300 nm bzw. im sichtbaren Bereich bei 350 und 540/550 nm (nach postchromatographischer Derivatisierung) mit dem TLC-Scanner 3 und winCATS Software. Ebenfalls können mit dem TLC-Scanner 3 UV/VIS-Spektren der Analyten zum Vergleich mit den Spektren der Standardsubstanzen aufgenommen werden.

Ergebnisse und Diskussion

Bei den Untersuchungen der Rohwässer des Zweckverbandes Landeswasserversorgung konnten nach Festphasen-Extraktion und unter Einsatz der HPTLC/AMD neben Atrazin und Desethyl-atrazin zwei Kontaminanten mit jeweils den ungewöhnlichen Zweitmaxima bei 280/300 nm (neben dem Hauptmaximum bei 220 nm) mit Laufstrecken von 22,0 und 33,5 mm erkannt werden. Diese Substanzen konnten zunächst anhand ihrer UV-Spektren keinen bisher bekannten Pestiziden oder sonstigen Stoffen zugeordnet werden.



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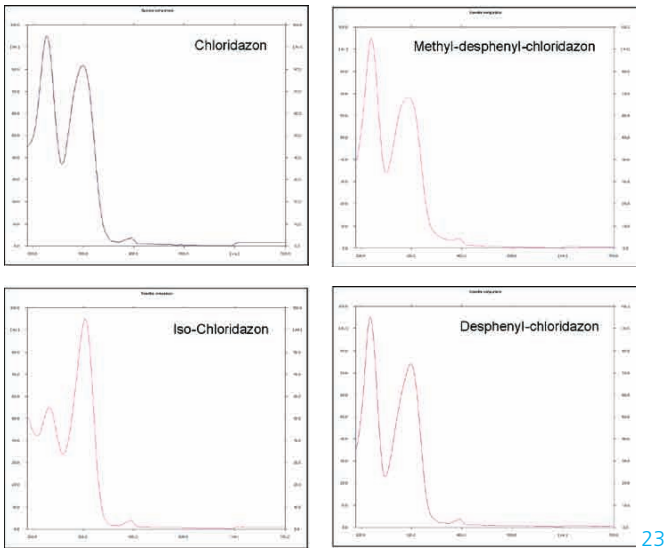
▲ *HPTLC/AMD-UV/VIS-Chromatogramm eines Grundwasserextraktes, aufgenommen im Mehrwellenlängenscan*



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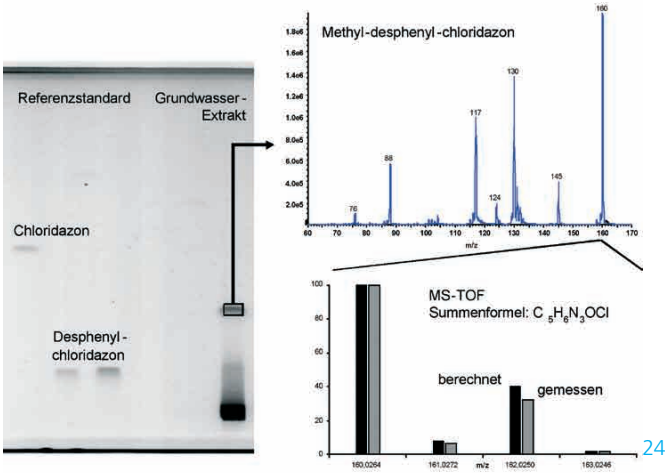
▲ *UV/VIS-Spektren von Chloridazon, Iso-Chloridazon, Desphenyl-chloridazon und Methyl-desphenyl-chloridazon (190 bis 700 nm)*

Zur Strukturaufklärung wurden die beiden unbekannten Substanzen mittels ChromeXtract® von der HPTLC-Platte in ein Flugzeit-Massenspektrometer überführt. Es konnten die molaren Massen von 145,0042 g/mol und 159,0199 g/mol bestimmt werden. Die Massendifferenz von 14 Einheiten lässt auf eine Methylgruppe schliessen. Zudem deutete das jeweilige Isotopenmuster auf monochlorierte Verbindungen hin. Weiterhin war es möglich, auch mit Hilfe des Nachweises der primären Amino-Gruppe die Metabolite Desphenyl-chloridazon bzw. Methyl-desphenyl-chloridazon zu identifizieren.



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▲ *Massenspektrum der zweiten unbekannten Substanz »Methyl-desphenyl-chloridazon« nach Transfer von der HPTLC-Platte (vor Anfärbung mittels Bratton-Marshall-Reagenz)*

Weiterhin wurden Laborversuche zur Entfernung von Chloridazon und Desphenyl-chloridazon mittels Ozonbehandlung durchgeführt. Dabei kamen wässrige ungepufferte Lösungen der Testsubstanzen mit den Konzentrationen 10 µg/L bzw. 0,5 µg/L zum Einsatz. Eine Ozon-Luftmischung wurde kontinuierlich unter Rühren in einen Semi-Batch-Reaktor dosiert, so dass sich eine Ozonkonzentration von 3 mg/L einstellte. Die Untersuchung der Ozonung ergab, dass Desphenyl-chloridazon im Vergleich zu Chloridazon geringfügig langsamer oxidiert wurde. Bereits nach 6 min Reaktionszeit konnte unter den gegebenen Bedingungen kein Chloridazon mehr nachgewiesen werden, wohingegen erst nach 8 min das Desphenyl-chloridazon vollständig abgebaut worden war. Vermutlich ist der sterisch ungehinderte Angriff des Ozons am Phenylrest des Chloridazons im Vergleich zu seinem Metaboliten

für den beobachteten Geschwindigkeitsunterschied bei der Ozonung verantwortlich.

Die aktuellen Untersuchungen im Rahmen eines Monitoring-Programms lassen vermuten, dass der Herbizid-Metabolit Desphenyl-chloridazon nicht nur in Bayern und Baden-Württemberg relativ häufig und in verhältnismässig hohen Konzentrationen nachgewiesen werden kann, sondern möglicherweise auch in vielen Oberflächen- und Grundwässern des gesamten Bundesgebietes und darüber hinaus zu erwarten ist.

Weitere Information sind bei den Autoren auf Anfrage erhältlich.

* Dr. W. Weber, Zweckverband Landeswasserversorgung, Betriebs- und Forschungslaboratorium, Am Spitzigen Berg 1, D-89129 Langenau, weber.w@lw-online.de

[1] W. Weber, W. Seitz, W. Schulz, H.-A. Wagener, Vom Wasser 105 (1) (2007)

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CAMAG AMD 2 System
(Automatisierte Mehrfachentwicklung)

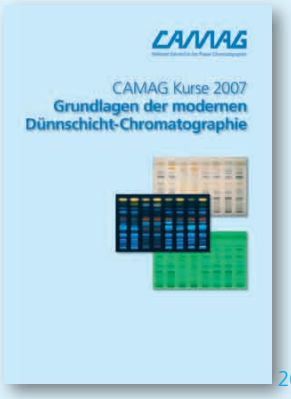
Wieder einmal war es das AMD-System, das bei der routinemässigen Ressourcenüberwachung im Rahmen des »multidimensionalen Screenings« zwei bis dato nicht bekannte Kontaminanten entdeckte.

Neben Bayer Industry Services (siehe CBS 96, S. 2–5) nutzt auch das Betriebs- und Forschungslaboratorium des Zweckverbandes Landeswasserversorgung die mit AMD verbesserte Trennleistung in Verbindung mit dem Mehrwellenlängen-Scan der winCATS-Software, um umfassender screenen zu können.

Durch den hohen Automatisierungsgrad erweist sich die planar-chromatographische Methode als durchaus wettbewerbsfähig. AMD wird eingesetzt, um die gewünschte Trennleistung auf der zur Verfügung stehenden Trennstrecke zu erreichen. Bei grossem Polaritätsbereich der zu trennenden Komponenten, bei hoher und unterschiedlicher Matrixbelastung sowie generell bei Vielkomponenten-Gemischen ist dies eine bevorzugt eingesetzte Trenntechnik. Durch die Mehrfach- und Gradientenentwicklung wird eine Fokussierung der Zonen erreicht und die Peakschärfe verbessert. Dies führt oft zu einer besseren Nachweisempfindlichkeit.

Aus- und Weiterbildung in der Planar-Chromatographie

CAMAG ist auch Ihr Partner, wenn es um Aus- und Weiterbildung auf dem Gebiet der Planar-Chromatographie geht. In Muttenz (Schweiz) bieten wir regelmässig Kurse an.



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Grundlagen der modernen Dünnschicht-Chromatographie

- Prinzipielle theoretische Zusammenhänge der DC/HPTLC,
- Vorteile der Technik,
- Einstieg in die quantitative Auswertung,
- Auswahl des Trennsystems,
- Gerätesysteme und Software.

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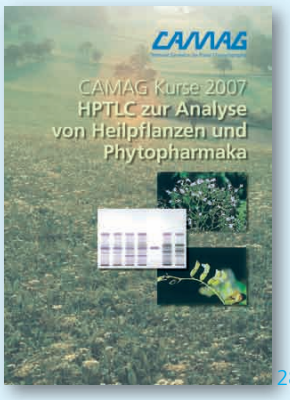
Methodenentwicklung in der Dünnschicht-Chromatographie

- Prinzipien und praktische Anwendung der Methoden-entwicklung und –validierung,
- Beurteilung des Trennproblems und die Auswahl eines geeigneten Trennsystems,
- Weiterentwicklung und Optimierung bestehender Methoden.



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HPTLC zur Analyse von Heilpflanzen und Phytopharmaka

- Einsatzmöglichkeiten der HPTLC für die Analyse von Heilpflanzen,
- Fingerprint-Analyse,
- Bilderfassungstechnologie zur Dokumentation und Auswertung,
- Stabilitätsuntersuchungen,
- Validierung quantitativer und qualitativer Methoden.

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