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Ein hoch aktuelles Thema: HPTLC zur Untersuchung von Lebensmitteln auf unerlaubte Farbzusätze

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HPTLC-Bestimmung verbotener Farbstoffe in Chili, Paprika und Curry

1

Von links nach rechts: Matthias Bleisch und Dr. Helmut Kandler

Als offizielle Lebensmittelkontrollbehörde ist das Kantonale Labor Zürich interessiert an neuen und/oder optimierten, zuverlässigen analytischen Methoden. Dr. Helmut Kandler, Abteilungsleiter Lebensmittelanalytik, und seine Mitarbeiter setzen seit mehreren Jahren HPTLC als wertvolle Ergänzung zu anderen Techniken ein. In Zusammenarbeit mit Dr. Eike Reich und Valeria Widmer vom CAMAG Labor in Muttenz wurde nun eine schnelle und zuverlässige HPTLC-Methode zur Bestimmung verbotener Farbstoffe in Gewürzen entwickelt und validiert.

Einleitung

Verschiedene Länder der Europäischen Union haben in den letzten Jahren in Proben von Chilipulver die Azofarbstoffe Sudan I–IV nachgewiesen. Diese synthetischen orangen und roten Farbstoffe sind für die Verwendung in Lebensmitteln nicht zugelassen, werden aber verbotenerweise eingesetzt, um die natürliche Farbe der Gewürze künstlich zu verstärken und damit mangelhafte Qualität zu kaschieren.

Die vorgestellte validierte RP-HPTLC Methode wird seit 2007 vom Kantonalen Labor Zürich erfolgreich in der Routine eingesetzt. Vor allem Chili-, Paprika-, Currypulver und Gewürzmischungen wurden auf die verbotenen Farbstoffe Sudan I, II, III, IV, Sudanrot B, Sudanrot 7B, Sudanrot G, Pararot, FD&C Orange 2, Buttergelb, Citrusrot 2, Toluidinrot und Dispers Orange 11 visuell gescreent. Eine zusätzliche Bestätigung und/oder Quantifizierung erfolgte im Falle von positiven Proben densitometrisch unter Anwendung einer Matrixkalibration. Verfälschte Gewürzprodukte zeigten typischerweise Kontaminationen über 100 mg/kg.

Standardlösungen

Je 20 mg Pararot, Sudan III, Sudan IV, Toluidinrot und Sudanrot 7B und je 20 mg Sudan I, Sudan II, Citrusrot 2, Buttergelb, Sudanrot B, FD&C Orange 2, Sudanrot G und Dispers Orange 11 wurden in Aceton bzw. Acetonitril gelöst und auf 100 mL aufgefüllt (200 µg/mL). Je 5 mL der Lösungen der entsprechenden Mischung wurden zur Trockne eingedampft (50 °C, 120 hPa) und in 10 mL Acetonitril aufgenommen (Farbstoff je 100 µg/mL in Mix 1- bzw. Mix 2-Standardlösung).

Probenvorbereitung

5 g homogenisierte Probe wurden mit 50 mL Acetonitril unter Rühren 10 min extrahiert und danach filtriert. Zu 10 mL Filtrat wurde tropfenweise Eisen(III)-chloridlösung (5 mg/mL in Acetonitril) bis zum Farbumschlag von rot nach grün (ca. 0.3–0.8 mL) zugegeben. Die Lösung wurde zur Trockne eingedampft, der Rückstand in 1 mL alkalischem Dichlormethan (250 mL Methylenchlorid mit 10 mL Ammoniak 25% ausgeschüttelt und abgetrennt) aufgenommen und über eine Festphasenextraktionssäule mit Kieselgel gereinigt. Das Eluat wurde zur Trockne eingedampft und der Rückstand in 1 mL Acetonitril aufgenommen.

Aufgestockte Proben

In einem 100 mL Erlenmeyerkolben wurden je 5 g einer unkontaminierten Blindprobe mit 0.5, 1.5, 3, 4.5 und 6 mL der Mix 1- oder Mix 2-Standardlösung in Doppelbestimmung dotiert, mit Acetonitril auf 50 mL aufgefüllt und wie oben beschrieben extrahiert (Konzentration der dotierten Probe: 10–120 mg/kg).

Schicht

HPTLC-Platten Kieselgel RP18 F_{254s} 10 x 10 cm und 20 x 10 cm, Merck

Probenauftragung

Bandförmig mit Linomat 5, Bandlänge 8 mm, unterer Randabstand 8 mm, seitlicher Randabstand mind. 15 mm, Bahnabstand mind. 10 mm, Auftragevolumina 10 µL für Proben und 1–12 µL für Standardlösungen bzw. 3–15 µL nach 1:10 Verdünnung

Chromatographie

In der Automatischen Entwicklungskammer ADC2 mit Acetonitril – 25 % Ammoniak 19:1, Laufstrecke (vom unteren Plattenrand) 60 mm

Dokumentation

Mit DigiStore 2 oder TLC Visualizer unter Weisslichtbeleuchtung

Mix 1	R_F	R_{rel}			R_{rel}	R_F	Mix 2
Pararot	0.60	1.22			1.38	0.66	Disp. Orange 11
Citrusrot 2	0.54	1.10			1.23	0.59	Buttergelb
Sudan I*	0.49	1.00			1.10	0.53	Toluidinrot
Sudan II	0.33	0.67			1.00	0.48	Sudanrot G*
Sudan III	0.23	0.47			0.83	0.40	FD&C Orange 2
Sudan IV	0.16	0.33			0.44	0.21	Sudanrot 7B
-	-	-			0.31	0.15	Sudanrot B

2

*Willkürlich gewählt als Bezugspunkt für den relativen R_f-Wert

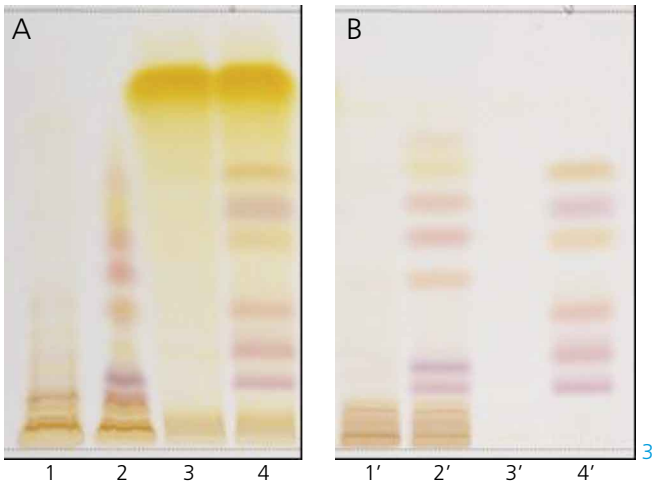
Densitometrie

Mehrwellenlängenscan mit dem TLC-Scanner 3 und winCATS Software beim jeweiligen Absorptionsmaximum der einzelnen Farbstoffe:

Farbstoff	λ_{max} (nm)	Farbstoff	λ_{max} (nm)
Sudan I	495	Sudanrot G	514
Sudan II	508	Sudanrot B	533
Sudan III	523	FD&C Orange 2	502
Sudan IV	534	Buttergelb	453
Pararot	498	Toluidinrot	522
Citrusrot 2	529	Dispers Orange 11	488
Sudanrot 7B	551		

Ergebnisse und Diskussion

Die Probenvorbereitung erwies sich als entscheidender Schritt der Analyse. Selektive Extraktion der synthetischen Farbstoffe wurde durch Zugeben einer Eisen(III)-chloridlösung erreicht, wodurch die natürlichen Gewürzpigmente zu farblosen Derivaten oxidiert wurden. Mit einer zusätzlichen Reinigung über Festphasenextraktion konnte der Einfluss der Probenmatrix weiter reduziert werden.

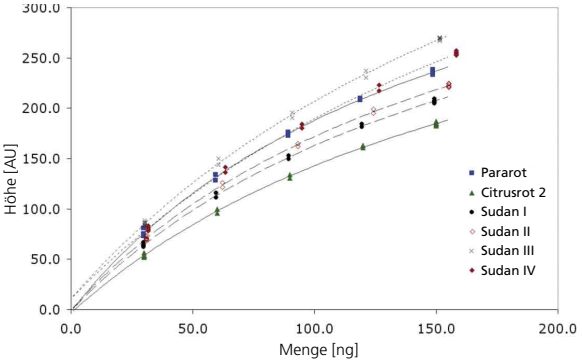


3

Einfluss des Oxidationsschrittes auf die Chromatographie: A: ohne Oxidation; B: Oxidation mit Eisen(III)-chlorid 1, 1': Paprika; 2, 2': Paprika dotiert mit 50 mg/kg der Standardlösung Mix 2; 3, 3': Curry; 4, 4': Curry dotiert mit 50 mg/kg der Standardlösung Mix 1

Die in der Routine angewendete Methode wurde bezüglich Güte der Kalibrierfunktion, Matrixkalibrierfunktion, Methodenpräzision und Nachweisgrenzen validiert. Zunächst wurden Kalibrierkurven im unteren (30–150 ng/Zone) und oberen (100–1200 ng/Zone) Konzentrationsbereich erstellt und mit einer Michaelis-Menten 2-Funktion ausgewertet.

4 *Chromatogramm von Mix 1: Die tiefste und die höchste Konzentration wurden jeweils 5-mal aufgetragen (Mitte der Platte) sowie doppelt der gesamte Konzentrationsbereich (links und rechts).*

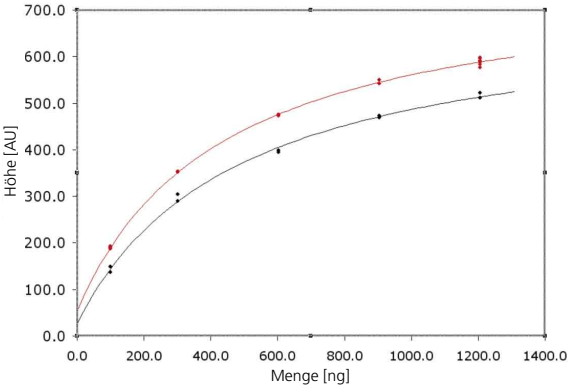


5 *Kalibrationskurven der Mix 1-Farbstoffe im unteren Konzentrationsbereich*

Kalibration von 13 Farbstoffen (Mix1, Mix2)	*%RSD* bei tiefster Konzentration	*%RSD* bei höchster Konzentration	sdv* (%)
Unterer Konzentrationsbereich	2.3–6.7	0.5–2.9	1.2–3.8
Oberer Konzentrationsbereich	0.7–6.4	0.3–4.3	0.5–4.8

sdv= Relative Reststandardabweichung der Kalibierkurven

Die Matrixkalibration wurde mit aufgestockten Paprika- und Curryproben im Bereich von 10–120 mg/kg geprüft. Die erhaltenen Matrixkalibrationskurven waren vergleichbar mit direkter Kalibration der Standards, die etwas tieferen Werte lassen sich durch einen gewissen Probenverlust während der Aufarbeitung erklären. In einzelnen Fällen wurden aufgrund schwacher Matrixinterferenzen etwas höhere Werte gemessen (Bsp. Sudan I in Paprikapulver).



6 *Vergleich der Kalibrationskurven: Standardlösung Sudanrot B (rote Kurve) und Sudanrot B in Currypulver (schwarze Kurve)*

Aufgrund der festgestellten Matrixeffekte lassen sich Wiederfindungen nur bedingt bestimmen. Eine detaillierte Prüfung und Bestimmung der einzelnen Wiederfindungen wurde daher nicht durchgeführt.

Zur Bestimmung der Präzision der Methode (0.4–7.0 %) wurden jeweils 6 Proben Paprika und Curry mit 50 mg/kg der Standardlösungen Mix 1 bzw. Mix 2 aufgestockt und unter Anwendung einer Matrixkalibration quantifiziert.

Die realen Nachweisgrenzen wurden aus einer Matrixkalibration (Bereich: 1–17 mg/kg) aufgestockter Paprika und Curryproben abgeschätzt. Mit densitometrischer Auswertung wurde im Vergleich zur visuellen Beurteilung eine um den Faktor 2 tiefere Nachweisgrenze für Paprika und Curry erreicht.

	Nachweisgrenze (visuell)	Nachweisgrenze (densitometrisch)
Curry	ca. 3 mg/kg (5 mg/kg Sudan I,7 mg/kg Buttergelb, 13 mg/kg Dispers Orange)	1–3 mg/kg (7 mg/kg Dispers Orange)
Paprika	ca. 3 mg/kg (5 mg/kg Sudan I und Buttergelb, 12 mg/kg Dispers Orange)	1–3 mg/kg

Bei der Untersuchung auf dem Markt erhältlicher Produkte wurde festgestellt, dass mit verbotenen Farbstoffen verfälschte Gewürze typischerweise mit mehr als 100 mg/kg belastet sind. Eine Kontamination im tieferen mg/kg-Bereich war nicht Gegenstand dieser Untersuchungen, da in diesen Mengen die gewünschte Farbverstärkung nicht erreicht wird. Die vorgestellte Methode erwies sich damit als geeignet für den schnellen, empfindlichen und reproduzierbaren Nachweis einer Verfälschung von Gewürzen mit verbotenen Farbstoffen.

Weitere Informationen sind bei den Autoren auf Anfrage erhältlich.

[1] H. Kandler, M. Bleisch, V. Widmer, E. Reich, J. Liq. Chromatogr. Related Technol. 32 (2009) 1273

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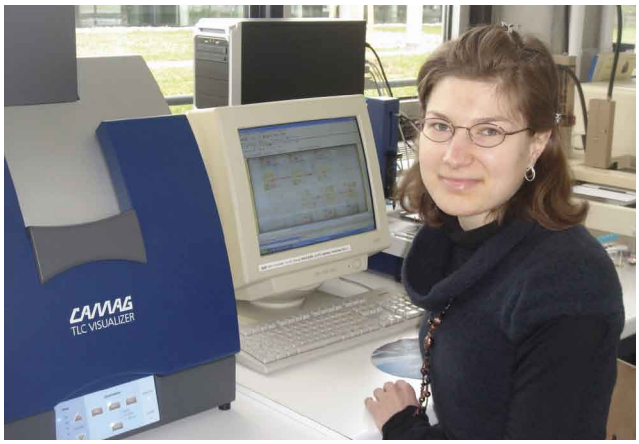
Analyse wasserlöslicher Farbstoffe in Lebensmitteln



7 *Claudia Oellig*

Am Institut für Lebensmittelchemie der Universität Hohenheim in Stuttgart wird die Planar-Chromatographie aufgrund ihrer Vorteile in der Lebensmittelanalytik eingesetzt. Claudia Oellig nutzte diese Technik im Rahmen ihrer Wissenschaftlichen Abschlussarbeit.

Einleitung

In den vergangenen Jahren wurde die Anzahl zugelassener Lebensmittel-Farbstoffe aus Gründen der Lebensmittelsicherheit deutlich reduziert. Durch die EG-Verordnung 94/36 werden die etwa 40 zugelassenen Lebensmittel-Farbstoffe hinsichtlich ihrer Einsatzbereiche und ihrer Höchstmengen geregelt. Zur Sicherung des Verbraucherschutzes sind schnelle und effiziente quantitative Bestimmungsmethoden notwendig, da manche Farbstoffe ein kanzerogenes Gefährdungspotential besitzen. Bisherige DC/HPTLC-Methoden waren für die Trennung von 9–12 Lebensmittel-Farbstoffen ausgerichtet. Ziel war es, eine Methode zu entwickeln, bei der die wichtigsten wasserlöslichen Lebensmittel-Farbstoffe quantifiziert werden können.

Im Vergleich zu vorhandenen Methoden zur Farbstoffanalytik ist die neue HPTLC-Methode eine zuverlässige, schnelle und kosteneffektive quantitative Alternative [1–3]. Sie ermöglicht einen Durchsatz von 1000 Proben/Tag mit geringen laufenden Kosten. Für eine Probe berechnet sich die gesamte Analysenzeit auf 1.5 min mit einem Lösungsmittelverbrauch von 200 µL. Der analytische Aufwand kann graduell nach Notwendigkeit gewählt werden – von visueller Auswertung über die spektrale Korrelation der Absorptionsspektren bis hin zur Aufnahme von Massenspektren.

Probenvorbereitung

Kommerziell erhältliche Lebensmittel wurden entsprechend mit Methanol – Ammoniumacetat-Puffer (pH 6.8) 1:1 verdünnt und bei Bedarf entgast.

Standardlösungen

Die Farbstoffe wurden in Methanol – Ammoniumacetat-Puffer (pH 6.8) 1:1 in folgenden Konzentrationen gelöst:

Mix 1			Mix 2			Mix 3		
Farbstoffe	Konzentr. [ng/µL]	hR_F	Farbstoffe	Konzentr. [ng/µL]	hR_F	Farbstoffe	Konzentr. [ng/µL]	hR_F
E 100	30	93	E 103	50	86	E101	30	72
E 101b	45	5	E 104	100	55	E102	20	19
E 110	20	57	E 120	70	0	E 105	25	53
E 122	20	71	E 121	125	93	E 129	15	60
E 124	15	27	E 123	8	25	E 133	8	26
E 126	30	10	E 125	60	72	E 141Na	860	86
E 127	10	93	E 151	15	15	E 141Cu	200	97
E 131	10	40				E 163	300	0
E 132	200	0						
E 142	8	23						

Schicht

HPTLC-Platten Kieselgel 60 F_{254} (Merck), 20 × 10 cm, vorgewaschen durch Chromatographie mit Methanol – Wasser 4:1

Probenauftragung

Bandförmig mit DC-Probenautomat 4, 18 Bahnen, Bandlänge 7,5 mm, Bahnabstand 9 mm, seitlicher Randabstand 24 mm, unterer Randabstand 8 mm (bei beidseitiger Auftragung 5 mm), Auftragevolumen 2 µL (Proben) bzw. 1–4 µL (Standardgemische)

Chromatographie

In der Doppeltrogkammer 20×10 cm mit 8 mL Ethylacetat – Methanol – Wasser – Essigsäure 65:23:11:1, Laufstrecke max. 50 mm, Laufzeit 12 min; alternativ kann auch die ADC2 oder HDC eingesetzt werden, insbesondere, wenn ein hoher Probendurchsatz gefordert wird.

Dokumentation

Mit TLC-Visualizer bei UV 254, UV 366 nm und Weisslicht-Beleuchtung

Densitometrische Auswertung

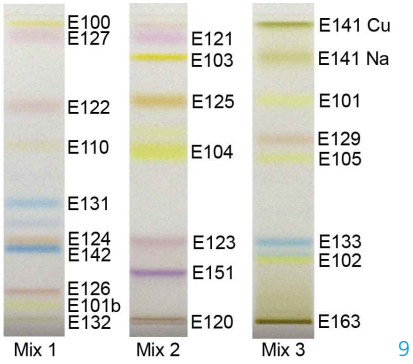
- Digitale Bildauswertung mit VideoScan-Software (Savitsky Golay-Filterweite: meist 7 oder 9, Basislinienkorrektur: geringste Steigung, Einsatz unterschiedlicher elektronischer Filter) oder
- Auswertung mit TLC-Scanner 3 und winCATS-Software, Absorptionsmessung über den Mehrwellenlängen-Scan bei 11 verschiedenen Wellenlängen [1]

Spektrenaufnahme (Vis, MS)

- Aufnahme der Vis-Spektren (400–800 nm) und Berechnung der Spektrenkorrelationen (Probe versus Standard) mit TLC-Scanner 3 und winCATS-Software und/oder
- Aufnahme der HPTLC/ESI-Massenspektren mit einem Prototyp des TLC-MS-Interface (Extraktionsmittel Methanol, Flussrate 0,2 mL/min)

Ergebnisse und Diskussion

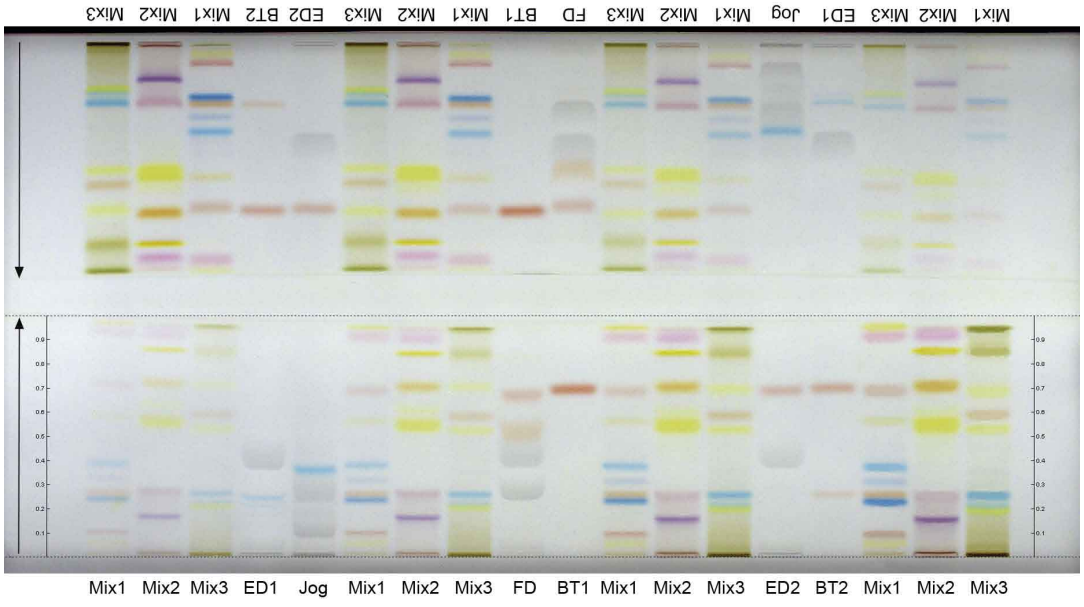
Die Trennung auf Kieselgel-Platten war mit Ethylacetat – Methanol – Wasser – Essigsäure 65:23:11:1 als Fliessmittel optimal. Der Säuregehalt von 1% erwies sich für die Fokussierung der Zonen als entscheidend. Wie in der Literatur üblich, wurden die Lebensmittelfarbstoffe zur besseren Quantifizierung in Farbstoffgemische aufgeteilt.



9 *Trennung von 25 wasserlöslichen Lebensmittelfarbstoffen, aufgeteilt in 3 Farbstoff-Mischungen (Farbstoffe zum Teil nur zu 50 bzw. 85 % rein)*

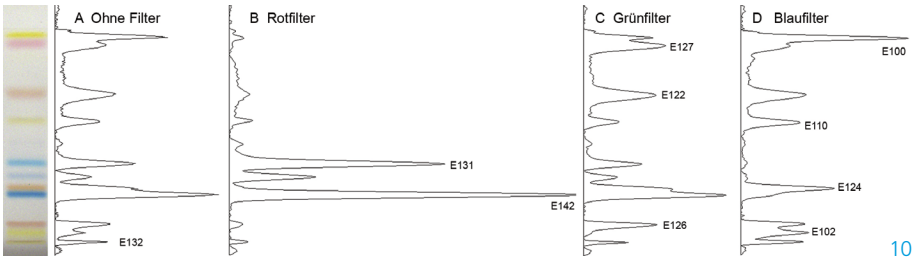
Bei einer Laufstrecke von max. 50 mm betrug die benötigte Trennzeit 12 min. Bei Entwicklung in der HDC wurden 36 Läufe simultan unter identischen Bedingungen entwickelt. Pro Probe berechneten sich die Chromatographiezeit auf 20 s und der Lösemittelverbrauch auf 220 µL; die Entsorgungskosten lagen deutlich unter 0,01 Cent. Die gesamte Analysenzeit (inkl. Probenvorbereitung, Auftragung und digitaler Bildauswertung) betrug 1,5 min – im Zeitalter ultraschneller Chromatographie-Methoden ein Spitzenwert.

Bei hoher Matrixbelastung waren die Flächenauftragung und/oder ein Verdünnen der Probe samt einer guten digitalen Bildauswertung conditio sine qua non. Im Falle von Proben mit Farbstoffen vom hR_F-Wert 0 (E120, E132, E163) wurde die Platte mit einem elutionsstärkeren Fliessmittel, z.B. im Verhältnis 45:35:18:2 zweimal auf 1 cm entwickelt (je 6 s).

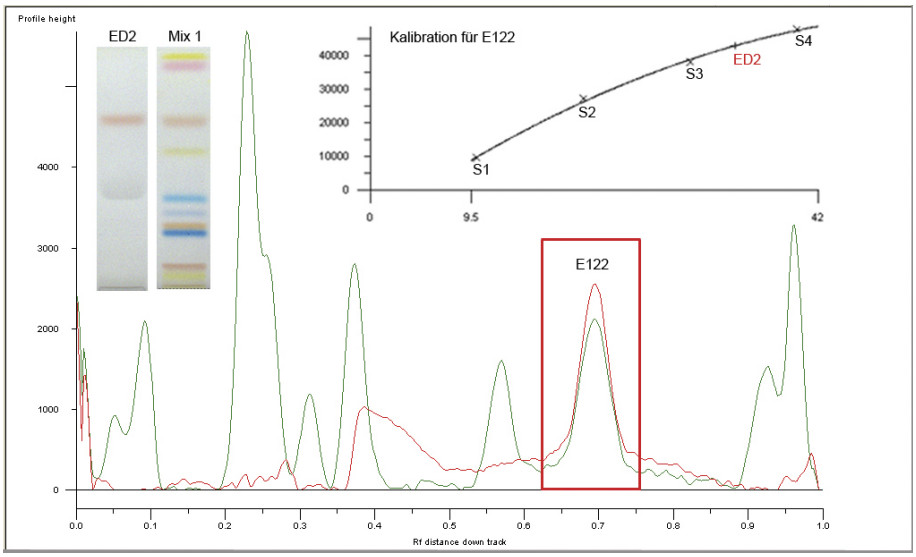


8 *Analytik von 12 Lebensmittelproben (Energydrink (ED), Joghurt (Jog), Fruchtgetränk (FD), Bäckereitinten-Formulierung (BT)) auf 25 wasserlösliche Lebensmittelfarbstoffe durch anti-parallele Entwicklung in 12 min*

Die Quantifizierung erfolgte mittels Mehrwellenlängen-Scan durch Absorptionsmessung im ultravioletten und sichtbaren Bereich oder über die digitale Quantifizierung des Plattenfotos.



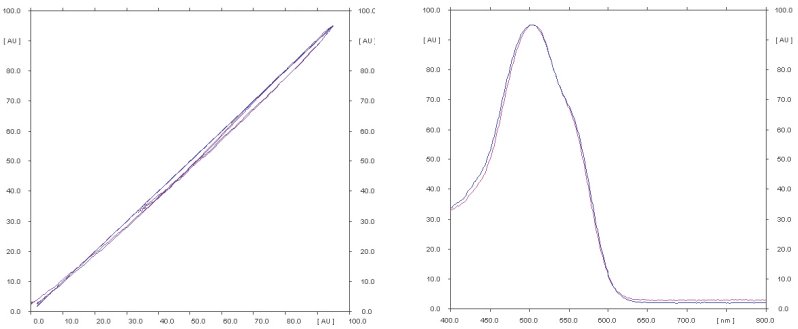
10 *Digitale Bildauswertung unter Einsatz von elektronischen Filtern (B–D) zur verbesserten post-chromatographischen Auflösung von Farbzonen (Mix 1)*



11 *Digitale Bildauswertung (ohne Filter) der verdünnten Energydrink-Probe 2 (ED2), die den roten Farbstoff E122 (Mix 1) enthält; Überlagerung der Analogkurven von Probe (rot) und Mix 1 (grün) sowie polynome Kalibrierfunktion (Peakfläche)*

Exemplarisch die Auswertung für einige Lebensmittel, wobei die Identität über die Spektrenkorrelation und das Massensignal abgesichert wurde.

Bezeichnung	Ermittelte Farbstoffe	Bestimmte Konzentration	*%RSD* (n = 2)	Identität	
				Spektrenkorrelation (400 – 800 nm) von Standard und Probe	Massensignal(e) (full scan, *m/z* 100 – 900)
Bäckerei-Tinte	122	66,4 g/L	0,0	≥ 0,99996	228 $[M\text{-}2Na]^{2-}$
	124	13,3 g/L	2,1	≥ 0,99957	279 $[M\text{-}2Na]^{2-}$
					178 $[M\text{-}3Na]^{3-}$
Energydrink 1	133	9,1 mg/L	0,1	≥ 0,99964	373 $[M\text{-}2Na]^{2-}$
Energydrink 2	122	76,2 mg/L	3,6	≥ 0,99958	228 $[M\text{-}2Na]^{2-}$



12 *Korrelation der Vis-Spektren von Standardzone E122 und der entsprechenden Zone in Energydrink 2*

Fortsetzung auf Seite 9

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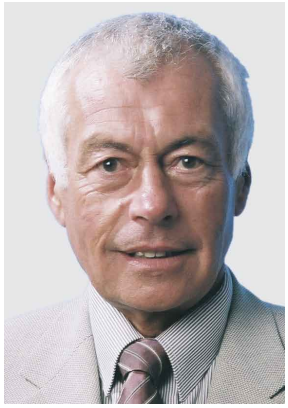
13

Wechsel im Bereich Finanzen



14

Herr Christian Gfeller ist seit 1966 bei CAMAG. 42 Jahre lang hat er als Leiter des Bereichs Finanzen (CFO) die Geschicke des Unternehmens in guten und in schwierigen Zeiten massgebend mit bestimmt. Seit 1992 ist er Mitglied des Verwaltungsrates und seit 2003 dessen Präsident. Mitte 2008 gab er seine Funktion als CFO an Frau Sabine Bührer weiter.

Wir wünschen uns, dass Herr Christian Gfeller noch lange Zeit als Präsident des Verwaltungsrats der CAMAG zur Verfügung steht.

15

Frau Sabine Bührer ist seit 1. Dezember 2007 bei CAMAG. Sie besitzt das Handelsdiplom der École Supérieure de Commerce in Neuchâtel seit 1987 und absolvierte in den darauf folgenden Jahren Weiterbildungen in den Bereichen Betriebswirtschaft, Personalführung, Controlling und Unternehmensführung. Fachliche Erfahrungen erwarb sie sich bei Schweizer und internationalen Unternehmen als Leiterin Administration, Finanzen und Personal.

Seit ihrem Eintritt bei CAMAG zeichnete sie sich aus durch Kompetenz, schnelle Auffassungsgabe in unserem sehr speziellen technischen Arbeitsgebiet sowie vor allem durch ihre absolute Vertrauenswürdigkeit. So kann Herr Gfeller guten Gewissens den Stab an Frau Sabine Bührer übergeben.

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Liebe Freunde

Die Weichen für das nächste International Symposium on High Performance Thin-Layer Chromatography sind gestellt: Es wird stattfinden vom 6. bis 8. Juli 2011, und zwar in Basel.

Damit wird die Reihe der Veranstaltungen fortgesetzt, die mit dem Symposium 1980 in Bad Dürkheim ins Leben gerufen wurde und deren bisher letzte 2006 in Berlin und 2008 in Helsinki stattfanden. Bitte reservieren Sie sich den Termin und überlegen Sie, ob Sie einen Vortrag oder ein Poster anmelden wollen. Das erste Zirkular mit Call for Papers wird Ende dieses Jahres herausgehen und im nächsten CBS wiederholt werden.

Die Beiträge der weissen Seiten des vorliegenden CBS-Hefts, die die Analyse von Farbstoffzusätzen in Lebensmitteln thematisieren, belegen wiederum eindrucksvoll, wie man mit der HPTLC schnell und kostengünstig anspruchsvolle, analytische Aufgaben lösen kann. U.a. werden für den Nachweis unzulässiger Farbstoffe in Gewürzen alternativ zwei Methoden dargestellt, die sich durch Probenvorbereitung und das chromatographische System unterscheiden, jedoch gleichermassen die analytische Fragestellung lösen.

Der Beitrag Schwack/Pellissier schliesst die rasche Identifizierung durch UV/Vis-Spektrensuche und HPTLC-MS-Spektren mit ein. Dabei wird eine interessante MS-Software angesprochen, mit der die Massengenauigkeit verbessert und die Trefferquote für die richtige Summenformel erhöht werden.

Sie sehen, die Planar-Chromatographie bewegt sich auf wissenschaftlich anspruchsvollem Gebiet.

Mit freundlichen Grüssen

Gerda Morlock

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Dear friends

Preparations for the next International Symposium on High Performance Thin-Layer Chromatography are already in progress. It will be held in Basel, 6^{th}–8^{th} of July 2011. It will continue the series of international conferences that began with the symposium in Bad Dürkheim, Germany in 1980, the latest two being held in Berlin 2006 and Helsinki 2008. Please reserve the date and start planning your participation, whether it will be a paper or a poster presentation or just attending. The first circular with a call for papers will go out at the end of this year and will also be included in the spring edition of CBS.

The contributions on the white pages of this CBS focusing on the analysis of illegal dyes in food prove once more the suitability of HPTLC for solving demanding analytical tasks rapidly and cost effectively. Two applications describe the identification of unauthorized dyes in spices by two alternative methods, each employing different ways of sample cleanup und different chromatographic systems. Both are solving the analytical task.

The contribution of Schwack/Pellissier includes the rapid identification by HPTLC-MS spectra and by UV/Vis spectra search. In this paper an interesting MS software is employed which significantly improves mass accuracy, thus greatly enhancing the score rate for a correct sum formula.

You see, planar chromatography is well established in scientifically demanding fields.

Sincerely,

Gerda Morlock

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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, herbal and traditional medicines
    f) Clinico-chemical applications and profiling body fluids
33. **Inorganic substances**
    a) Cations
    b) Anions
34. **Radioactive and other isotopic compounds**
35. **Other technical products and complex mixtures**
    a) Surfactants
    b) Antioxidants and preservatives
    c) Various specific technical products
    d) Complex mixtures and non-identified compounds
36. **Thin-layer electrophoresis**
37. **Environmental analysis**
    a) General papers
    b) Air pollution
    c) Water pollution
    d) Soil pollution
38. **Chiral separations**

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

1. Reviews and books

103 001 J. SHERMA (Department of Chemistry, Lafayette College, Easton, Pennsylvania 18042, USA; shermaj@lafayette.edu): Planar chromatography. Anal. Chem. 80, 4253-4267 (2008). This review covers the literature of TLC and HPTLC found by computer-assisted searching in Chem. Abstr. and the ISI Web of Science from November 1 2005 to November 1 2007 augmented by Analytical Abstracts and the following journals: J. Chromatogr. A and B, J. Chromatogr. Sci., Chromatographia, Anal. Chem., J. Liq. Chromatogr. Relat. Technol., J. AOAC Int., J. Planar Chromatogr., Acta Chromatographica, books, and reviews with the following chapters (number of citations): history (22), theory and fundamental studies (19); chromatographic systems (28); apparatus and techniques (32); detection and identification (46): chemical detection, biological detection, TLC/mass spectrometry, TLC coupled with other spectrometric methods; quantitative analysis (56): technique and instruments as well as applications; preparative layer chromatography (4); radiochromatography (6).

review 1

103 002 L.R. SNYDER (LC Resources, 26 Silverwood Ct, Orinda, CA 94563, USA; snyder0036@comcast.net): Solvent selectivity in normal-phase TLC. J. Planar Chromatogr. 21, 315-323 (2008). The role of the mobile phase in controlling selectivity for adsorption chromatography - with either thin-layer plates or columns - is reviewed and expanded. The use of different solvent mixtures of varying selectivity in normal-phase chromatography is now on a firm theoretical and practical basis. The choice of a more polar component (B-solvent) of a binary solvent mixture (A/B) largely determines relative retention and resolution. Maximum differences in selectivity are achieved by the use of two mobile phases where the B-solvent is either very polar (requiring a lower % B for desirable values of k) or relatively nonpolar (requiring a higher % B).

review 1, 2e

2. Fundamentals, theory and general

103 003 V.G. BEREZKIN (A. V. Topchiev Insitute of Petrochemical Synthesis, Russian Academy of Sciences, Leninskii pr. 29, Moskow, 117912 Russia; berezkin@ips.ac.ru): Development of nontraditional planar chromatographic methods. J. Planar Chromatogr. 21, 325-329 (2008). Development and systematization of nontraditional TLC methods, in which the chromatographic process occurs in a closed absorption layer on a standard TLC plate. The advantages and limitations of the methods were assessed and the expediency of their further development and wider use in practical TLC was proved.

review 2a

103 004 T. DZIDO*, P. PLOCHARZ, P. SLAZAK, Aneta HALKA (*Department of Physical Chemistry, Medical University, Staszica 6, 20-081 Lublin, Poland, tadeusz.dzido@am.lublin.pl): Progress in planar electrochromatography. Anal. Bioanal. Chem. 391, 2111-2118 (2008). Review of developments in planar electrochromatography in open (PEC) and closed (PPEC) systems regarding the progress in chamber construction for planar electrochromatography, separating system performance, equilibration of the PPEC process, separation time and selectivity, and the general advantages, disadvantages and prospects of this separation mode. PPEC of 4-cholesten-3-one, 4-androsten-17alpha-ol-3-one acetate, 17R-acetoxyprogesterone, androstenedione, 4-pregen-11R-ol-3,20-dione, benzanilide, o-nitroaniline, hydrocortisone alcohol, and benzamide on RP-18 with 55 % aqueous acetonitrile containing 5 mM acetate buffer (pH 4.7) at 9 kV and under a pressure of 63 atm.

review, planar electrochromatography 2a

103 005 J.D. FAIR*, CH.M. KORMOS (*Department of Chemistry, University of Connecticut, 55 North Eagleville Road, Unit 3060, Storrs, CT 06269-3060, USA): Flash column chromatograms estimated from thin-layer chromatography data. J. Chromatogr. A 1211 (1-2), 49-54 (2008). Presen-

tation of a spreadsheet that provides information on the amount of silica gel needed, the optimal fraction size, and the degree of separation to be expected before flash chromatography is attempted. Information is based on sample mass and TLC data given. This is the first utility of its kind to accurately estimate the retention volume and band volume of analytes, as well as the fraction numbers expected to contain each analyte, and the resolution between adjacent peaks. The information allows users to select optimal parameters for preparative-scale separations before the flash column itself is attempted; ensuring a successful first separation.

preparative TLC, flash chromatography 2d

103 006 Jolanta FLIEGER*, R. SWIEBODA, M. TATARCZAK (*Department of Inorganic and Analytical Chemistry, Medical University of Lublin, 20-081 Lublin, Staszica 6, Poland): Chemometric analysis of retention data from salting-out thin-layer chromatography in relation to structural parameters and biological activity of chosen sulphonamides. J. Chromatogr. B 846 (1-2), 334-340 (2007). Salting-out TLC of sulphonamides on silica gel with aqueous solutions of salts (sulphates, chlorides, nitrates, phosphates, acetates, and thiocyanates) showed that the applied salts have different effects on the retention of sulphonamides according to Hofmeister's classification (e.g. kosmotropes, chaotropes and neutral). Parameters of the linear regression analysis were compared with QSAR data of dependences between the RM values and salt concentration. Chromatographic data obtained by salting-out TLC showed not only the physico-chemical properties of the examined compounds but also information about their activity. The method was suitable for prediction and classification of sulphonamide drugs by localization of other structurally similar compounds with antagonistic activity towards sulphonamides.

doping, HPTLC, quantitative analysis, qualitative identification 2c

103 007 L. KOMSTA (Department of Medicinal Chemistry, Skubiszewski Medical University, Jaczewskiego 4, 20-090 Lublin, Poland): Prediction of the retention in thin layer chromatography screening systems by atomic contributions. Anal. Chim. Acta 593 (2), 224-237 (2007). Presentation of a novel atomic-contribution system for predicting RM values by TLC on silica gel with 13 screening systems where the large experimental datasets (198-761 RM values) are available. RM values could be predicted with less than 0.5 % error in the majority of solutes (besides several outliers), which corresponds to an hRf value difference of 28 in the worst case. Validation of the system by dividing the data into training and validation datasets, proving its accuracy. The main reason for outliers were large conjugated heterocycles, quarternary ammonium cations, large amount of polar atoms or very simple but unique molecules. The method invloves easy manual calculation and no need for a software. It was applied to predict retention of new compounds in existing chromatographic screening systems.

cosmetics, HPTLC, qualitative identification 2c

103 008 S. NYIREDY (Research Institute for Medicinal Plants, Budakalász, Hungary): Multidimensional planar chromatography. LC-GC Europe Applications 16, 2-9 (2003). Overview of various multidimensional planar chromatography (MD-PC) techniques: Comprehensive two-dimensional PC (PC x PC), targeted or selective two-dimensional PC (PC x PC), modulated two-dimensional PC (nPC), coupled-layer PC (PC-PC), combined MD-PC methods (cMD-PC).

comparison of methods, review 2a

103 009 J. SHERMA (Department of Chemistry, Lafayette College, Easton, Pennsylvania 18042, USA): Planar chromatography. Anal. Chem. 76, 3251-3262 (2004). This review covers the literature of TLC/HPTLC found in Chemical Abstracts and ICI Web of Science from November 1, 2001 to November 1, 2003. Review Contents: 1. History, Student Experiments, Books, and Reviews; 2. Theory and Fundamental Studies; 3. Chromatographic Systems (Stationary and Mobile Phases); 4. Apparatus and Techniques; 5. Detection and Identification of Separated Zones; 6. Quantitative Analysis; 7. Preparative-Layer Chromatography and Thin-Layer Radiochromatography 8. Literature Cited:

review, preparative TLC, quantitative analysis, qualitative identification, postchromatographic derivatization 2a

103 010 J. SHERMA (Department of Chemistry, Lafayette, Collegte, Easton, Pennsylvania 18042, USA): Planar chromatography. Anal. Chem. 74, 2653-2662 (2002). This review covers the literature of TLC/HPTLC found in Chemical Abstracts and ICI Web of Science from November 1, 1999 to November 1, 2001. Review Contents: 1. History, Student Experiments, Books, and Reviews; 2. Theory and Fundamental Studies; 3. Chromatographic Systems (Stationary and Mobile Phases); 4. Apparatus and Techniques; 5. Detection and Identification of Separated Zones; 6. Quantitative Analysis; 7. Preparative-Layer Chromatography and Thin-Layer Radiochromatography 8. Literature Cited.

review, preparative TLC, quantitative analysis, qualitative identification, postchromatographic derivatization 2a

103 002 L.R. SNYDER, see section 1

3. General techniques

103 011 Virginia COMAN*, S. KREIBIK. M. VLASSA (*"Babes-Bolyai" University, „Raluca Ripan" Institute for Research in Chemistry, Fântânele 30, 400294 Cluj-Napoca, Romania; coman_virginia@yahoo.com): Planar dielectrochromatography in a vertical chamber. J. Planar Chromatogr. 21, 373-378 (2008). TLC of a lipophilic test dye mixture (indophenol blue, Sudan red G, 4-dimethylaminoazobenzene) on aluminum oxide in a vertical planar dielectrochromatography chamber with toluene and benzene. Evaluation by densitometric absorbance measurement at 506 nm. In classical TLC the mobile phase is drawn by capillary forces therefore its flow velocity is inversely related to the distance migrated by the solvent front. For this reason classical TLC may be time-consuming. To improve the separation selectivity suitable transverse alternating electric fields were used in planar electrochromatography to modify the mobile phase front velocity and the migration distance of solutes. Resolution was improved in the countercurrent arrangement of the instrument.

densitometry 3d

103 012 Y. LI (Li Yulan)*, L. LI (Li Li), J. XU (Xu Jianan) (*Zhejiang Parm. Coll., Ningbo, Zhejiang 311100, China): (Study on temperature controlled apparatus and its application in pharmaceutical analysis) (Chinese). Chinese J. Modern App. Pharm. 25 (4), 348-350 (2008). Description of an apparatus for the temperature control of the TLC separation process. The operational temperature below room temperature is adjusted by using semi-conductor refrigeration. Temperatures above room temperature are controlled by hot thread heating. Good reproducibility of hRf values is obtained with the apparatus.

temperature controlled development 3d

103 124 O. MORINAGA et al., see section 32e

103 013 D. NUROK *, J. KOERS, A. NOVOTNY, M. CARMICHAEL, J. KOSIBA, R. SANTINI, G. HAWKINS, R. REPLOGLE (*Department of Chemistry, Indiana University-Purdue University, 402 N. Blackford Street, Indianapolis, Indiana 46202, USA): Apparatus and initial results for pressurized planar electrochromatography. Anal. Chem. 76, 1690-1695 (2004). Pressurized planar electrochromatography (PPEC) is a new planar chromatographic technique in which the mobile phase is driven by electroosmotic flow, while the sorbent layer is pressurized in a manner that allows heat to flow from the layer through an electrically insulating, thermally conducting sheet of aluminum nitride ceramic. Separation in a PPEC prototype apparatus is faster than by conventional TLC, and an example is presented of a 24-fold enhancement in the speed of separation. PPEC was performed on TLC and HPTLC RP-18 phases which required conditioning at elevated temperature before use. Solute migration velocity increases with temperature. The flow rate increases in a linear manner with increasing voltage and diminishes in a nonlinear manner with increasing pressure. Both electrical current and Joule heating diminish with increasing pressure,

and the diminution of flow at high pressure can be compensated by an increase in voltage. PPEC is more efficient than classical TLC. Theoretical plate heights diminish with increasing hRf and are in the range 29-21 and 55-27 µm for the HPTLC and TLC, respectively. PPEC retains the advantages of classical TLC, but has the ability to separate a substantially higher number of samples. An example is presented on the separation of nine samples in 1 min.

HPTLC, pressurized planar electrochromatography 3d

103 014 T. TANG (Tang Tie-Xin)*, H. WU (Wu Hong) (*Center for Medicinal Plants Research, South China Agricultural University, 510642 Guangzhou, China): Research on color channel selection, three-dimensional visualization, and acquisition time of computerized image analysis for one-dimensional planar separation. Chromatographia 70 (1-2), 305-308 (2009). Description of color channel selection, three-dimensional visualization, and acquisition time of computerized image analysis for one-dimensional planar separation, known as computerized image analysis or video densitometry. This is an efficient, low-cost technique for quantitative and qualitative analysis of planar separations, e.g. planar chromatography and gel electrophoresis. The image of the TLC plate is captured in black and white, then the proper color channel of the image is selected in order to enhance the signal-to-noise ratio. To facilitate image evaluation a three-dimensional visualization of the planar image was applied by use of OpenGL technology. It was found that the sensitivity is increased by use of longer acquisition times whereas linearity of quantitative analysis is reduced.

doping, pharmaceutical research, HPTLC, quantitative analysis, densitometry, qualitative identification 3f

103 015 E. TYIHAK*, Á. M. MORICZ, P. G. OTT (*Plant Protection Institute, Hungarian Academy of Sciences, Herman Ottó Str. 15, P. O. B. 102, 1525 Budapest, Hungary; etyih@nki.hu): Use of the BioArena system for indirect detection of endogenous ozone in spots after TLC or OPLC separation. J. Planar Chromatogr. 21, 77-82 (2008). Ozone can be detected indirectly in TLC or OPLC zones by use of the bioautographic BioArena system and ozone-eliminating molecules (e.g. d-limonene and indigo carmine) in the culture medium. TLC and OPLC of test substances (trans-resveratrol, aflatoxin B1) on silica gel with a variety of mobile phases (e. g. chloroform - methanol 10:1). The developed and dried TLC plates were immersed in a bacterial suspension of Pseudomonas savastanoi for 20 s. Visualization of the chromatograms with MTT was performed either after a a short draining period or after overnight incubation.

qualitative identification, bioautography 3e

103 016 R. ZAKRZEWSKI*, W. CIESIELSKI, A. ULANOWSKA, R. MARTINEZ (*Department of Instrumental Analysis, University of Lodz, Lodz, Poland, robzak@chemul.uni.lodz.pl): 2,4,6-triphenylpyrylium cations as derivatization reagents for sulfide ions detection in TLC. Phosphorus, Sulfur Silicon Relat. Elem. 184, 1139-1148 (2009). The new TLC system for sulfide ions detection is based on the use of 2,4,6-triphenylpyrylium salts as pre-chromatographic derivatization reagents. The cations L1 (2,4,6-triphenylpyrylium) or LN1 (4-[p-(N,N-dimethylamino) phenyl]-2,6-diphenylpyrylium) were used in the derivatization reactions in a tube or directly on the TLC plate before the developing step. TLC of L1 on silica gel with methanol - dichloromethane 1:5. TLC of LN1 on cellulose with phosphoric buffer (pH 6.0) - acetonitrile - 1,4-dioxane 4:2:1. The detection procedure allows selective and sensitive detection for sulfide anions at several dozen pmol/spot.

environmental, quantitative analysis 3e

4. Special techniques

103 017 W. CHAI*, Christine LETEUX, A. LAWSON, M. STOLL (*MRC Glycosciences Laboratory, Imperial College School of Medicine, Northwick Park Hospital, Watford Road, Harrow, Middlesex HA1 3UJ, U.K., w.chai@ic.ac.uk): On-line overpressure thin-layer chromatographic separation and electrospray mass spectrometric detection of glycolipids. Anal. Chem. 75, 118-125

(2003). Online TLC separation and electrospray mass spectrometry (TLC/ESI-MS) by direct linking of a commercial overpressure TLC instrument, OPLC 50, and a Q-TOF mass spectrometer. Separation on silica gel with dichlormethane - methanol - water 60:35:8. A sensitivity of 5 pmol of glycosphingolipid was readily demonstrated for TLC/ESI-MS and 20 pmol for TLC/ESI-MS/MS production scanning to derive the saccharide sequence and long chain base/fatty acid composition of the ceramide. Initial preconditioning of TLC plates is necessary to achieve high sensitivity detection by reducing chemical background noise. Plates can be used repeatedly (at least 10 times) for analysis, although this may result in a minor reduction in TLC resolution. Following solvent development, separated components on the TLC plates can be detected in the conventional way by nondestructive staining or UV absorption or fluorescence and can be stored for on-line TLC/ESI-MS analysis at a later stage without reduction in mass spectrometric detection sensitivity and chromatographic resolution. Aspects for further improvement of OPLC instrumentation include use of narrower TLC plate dimensions and refined design of the eluate exit system.

qualitative identification, HPTLC, OPLC 4e, 11e

103 018 Irena CHOMA (Department of Chromatographic Methods, M. Curie-Sklodowska University, Lublin, Poland): The use of thin-layer chromatography with direct bioautography for antimicrobial analysis. LC-GC Europe 18, 482-488 (2005). Contact Bioautography: Antimicrobials diffusion from a TLC plate to an inoculated agar plate.The chromatogram is placed face down onto the inoculated agar layer and left for some minutes or hours for diffusion. After removing the plate the inhibition zones are observed on the agar surface in the places where the spots of antimicrobials are stuck to the agar. The method resembles a disk assay. Immersion Bioautography: The chromatogram is covered with a molten, seeded agar medium. After solidification, incubation and staining (usually with tetrazolium dye) the inhibition or growth bands are visualized. Direct Bioautography: A developed plate is dipped in the suspension of microorganisms growing in a suitable broth or this suspension is sprayed onto the plate. The plate is incubated and microorganisms grow directly on it. It can be performed with Photobacterium phosphoreum (Vibrio fischeri) suspension. Bioautography systems and coupling possibilities are presented.

food, analysis, qualitative identification, comparison of methods, bioautography 4e

103 019 Anna CRECELIUS*, M. CLENCH, D. RICHARDS (*Biomedical Research Centre, Sheffield Hallam University, Sheffield, UK): TLC-MALDI in pharmaceutical analysis. LC-GC Europe 16, 2-5 (2003). Overview of utility of the technique for the identification and quantification of pharmaceutical compounds and related substances such as UK-137,457 (C31H31NO5) and UK-124,912 ($C_{27}H_{25}NO_3$). Several of the issues that have arisen in the development of TLC-MALDI-MS methods for the successful analysis of pharmaceuticals were adressed in this article. Electrospray deposition method, which was found to be superior to other methods studied and was successfully applied to a range of compounds presented, concerns a method for the deposition of the MALDI matrix onto the TLC plate. Post-source decay analysis can be performed directly on the TLC spots, to aid in structural evaluation of the analyzed compounds. The generation of quantitative data using a structural analogue as internal standard and incorporation into the mobile phase has also been demonstrated. Outlook for next step development of TLC-MALDI-MS in pharmaceutical analysis for more widespread use in industry: enhance sensitivity, mass resolution and reproducibility, availability of commercial instruments that allow the scanning of whole TLC plates rapidly with data-imaging software.

pharmaceutical research, quantitative analysis, qualitative identification, TLC-MALDI-MS 4e

103 020 K. DREISEWERD*, J. MUTHING, A. ROHLFING, Iris MEISEN, Zeljka VUKELIC, Jasna PETER-KATALINIC, F. HILLENKAMP, S. BERKENKAMP (*Institute of Medical Physics and Biophysics, Westfälische Wilhelms-Universität Münster, 48149 Münster, Germany, Klaus.Dreisewerd@uni-muenster.de): Analysis of gangliosides directly from thin-layer chromatography plates by infrared matrix-assisted laser desorption/ionization orthogonal time-of-flight mass spectrometry with a glycerol matrix. Anal. Chem. 77, 4098-4107 (2005). Novel method for direct

coupling of HPTLC with matrix-assisted laser desorption/ionization mass spectrometry (MALDI-MS) for the analysis of biomolecules. HPTLC of gangliosides on silica gel with chloroform - methanol - water 24:17:4 containing 2 mM calcium chloride, with chamber saturation for 20 min. Detection by dipping for 10 s in 0.3 % (w/v) orcinol in 3 M sulfuric acid, followed by heating at 110 °C. Use of a glycerol as matrix, which provides a homogeneous wetting of the silica gel and a simple and fast preparation protocol. Use of an Er:YAG infrared laser, which ablates layers of 10 µm thickness of analyte-loaded silica gel and provides a soft desorption/ionization of even very labile analyte molecules. The orthogonal time-of-flight mass spectrometer employed in this study, finally provides a high accuracy of the mass determination, which is independent of any irregularity of the silica gel surface. The method is demonstrated by the compositional mapping of a native GM3 (II3-r- Neu5Ac-LacCer) ganglioside mixture from cultured Chinese hamster ovary cells. The analysis is characterized by a high relative sensitivity, allowing the simultaneous detection of various major and minor GM3 species directly from analyte bands. The lateral resolution of the direct HPTLC-MALDI-MS analysis is defined by the laser focus diameter of currently 200 µm. This allows one to determine mobility profiles of individual species with a higher resolution than by reading off the chromatogram by optical absorption.

HPTLC, MALDI-MS 4e, 11e

103 021 Beate FUCHS, J. SCHILLER*, Rosmarie SÜSZ, M. SCHÜRENBERG, D. SUCKAU (*Faculty of Medicine, Institute of Medical Physics and Biophysics, University of Leipzig, Härtelstr. 16-18, 04107 Leipzig, Germany, juergen.schiller@medizin.uni-leipzig.de): A direct and simple method of coupling matrix-assisted laser desorption and ionization time-of-flight mass spectrometry (MALDI-TOF MS) to thin-layer chromatography (TLC) for the analysis of phospholipids from egg yolk. Anal. Bioanal. Chem. 389, 827-834 (2007). A total extract of hen egg yolk is used as phospolipid mixture to demonstrate the capabilities. TLC of phosphatidylcholine, lysophosphatidylcholine, sphingomyelin, phosphatidylethanolamine, lysophosphatidylethanolamine and phosphatidylinositol on silica gel with chloroform - ethanol - water - triethylamine 5:5:1:5. Detection under UV 366 nm after spraying with primuline (Direct Yellow 59) reagent. Direct coupling MALDI-TOF MS and TLC can be easily implemented on commercially available MALDI-TOF devices. Roughly clean spectra without major contributions from fragmentation products and matrix peaks were obtained. This approach was sensitive enough to detect the presence of phospholipids at levels of less than 1 % of the total extract.

clinical chemistry research, food, analysis, qualitative identification, coupling TLC/MALDI-TOF MS 4e, 11c

103 022 Beate FUCHS, J. SCHILLER*, Rosmarie SÜSZ, M. ZSCHARNACK, A. BADER, P. MÜLLER, M. SCHÜRENBERG, M. BECKER, D. SUCKAU (*Faculty of Medicine, Institute of Medical Physics and Biophysics, University of Leipzig, Härtelstrasse 16-18, 04107 Leipzig, Germany, juergen.schiller@medizin.uni-leipzig.de): Analysis of stem cell lipids by offline HPTLC-MALDI-TOF MS. Anal. Bioanal. Chem. 392, 849-860 (2008). HPTLC of cell lipids on silica gel with chloroform - ethanol - water - triethylamine 5:5:1:5. Detection under UV 366 nm after spraying with primuline (Direct Yellow) reagent. Coupling with MALDI-TOF-MS which is traditionally used for proteomics, but is also a useful tool for lipid analysis. Depending on the applied matrix, however, some lipid classes are more sensitively detected than others and this may even lead to suppression effects if complex mixtures are analyzed. Therefore, a previous separation into the individual lipid classes is necessary. Using artificial lipid mixtures or easily available tissue extracts, it has already been shown that lipids can be conveniently analyzed by MALDI-TOF MS directly on the TLC plate. An initial TLC-MALDI-TOF-MS study of the lipid composition of ovine mesenchymal stem cells is presented. Due to the complex composition of these cells, data are also compared to lipids extracted from human erythrocytes. Even very minor lipid classes can be easily detected and with much higher sensitivity than by common staining protocols. clinical chemistry research, HPTLC, qualitative identification, MALDI-TOF MS

4e, 11c

103 055 R. HADDAD et al., see section 17

103 023 F.L. HSU, C.H. CHEN, C.H. YUAN, J. SHIEA* (*Department of Chemistry, National Sun Yat-Sen University, Kaohsiung, Taiwan, jetea@mail.nsysu.edu.tw): Interfaces to connect thin-layer chromatography with electrospray ionization mass spectrometry. Anal. Chem. 75, 2493-2498 (2003). Development of two interfaces to connect small-size thin-layer chromatography with electrospray ionization mass spectrometry (ESI-MS) for the continuous analysis of organic mixtures. The interfaces were 1) two bound optical fibers inserted into the RP-18 particles at the exit of a small TLC channel; 2) a small commercial TLC strip with a sharpened tip. A reservoir continuously supplied a makeup solution to the tip of the TLC channel. The high voltage required for electrospray ionization was introduced into the makeup solution or mobile phase through a Pt wire, and electrospray was generated at the tip of the bonded optical fibers and at the sharp end of the TLC strip. Since small-size TLC channels were used, the elution time was short and less than 0.2 µL of sample solution and 200 µL of solvent were required.

HPTLC, qualitative identification, ESI-MS 4e

103 101 W. HUANG et al., see section 32e

103 024 Vera IVLEVA, Y. ELKIN, B. BUDNIK, Susanne MOYER, P. O'CONNOR, Catherine COSTELLO* (*Mass Spectrometry Resource, Cardiovascular Proteomics Center, and Department of Biochemistry, Boston University, School of Medicine, 715 Albany Street R-806, Boston, Massachusetts 02118-2526, USA, cecmsms@bu.edu): Coupling thin-layer chromatography with vibrational cooling matrix-assisted laser desorption/ionization fourier transform mass spectrometry for the analysis of ganglioside mixtures. Anal. Chem. 76, 6484-6491 (2004). TLC on silica gel with chloroform - methanol - 0.2 % calcium chloride 11:9:2 or chloroform - 2-propanol - 50 mM potassium chloride 10:67:23 for separation of glycolipids, oligosaccharides, lipids, and compounds of environmental and pharmaceutical interest with coupling to an external ion source MALDI-Fourier transform (FT) MS instrument without compromising mass accuracy and resolution of the spectra. Furthermore, when the FTMS, with >70 000 resolving power, has a vibrationally cooled MALDI ion source, fragile glycolipids can be desorbed from TLC plates without fragmentation, even to the point that desorption of intact molecules from „hot" matrixes such as a-cyano-4-hydroxycinnamic acid is possible. TLC of whole brain gangliosides derivatized with orcinol-sulfuric acid reagent. The positions of the fractions on a parallel developed plate were determined; the TLC plates were attached directly to the MALDI target using double-sided adhesive tape or the strip cut from TLC plate. Different matrices were tested.

MALDI-MS 4e, 11e

103 168 J.P. LAFLEUR et al., see section 33a

103 025 M. LOPPACHER*, R. ROLLI (*CAMAG, Sonnenmattstr. 11, 4132 Muttenz, Switzerland, matthias. loppacher@camag.com): The new TLC-MS interface. CBS 102, 2-3 (2009). A semi-automatic interface for hyphenation of TLC with mass spectrometry, which was based on the ChromeXtractor by Luftmann, is introduced. The interface is connected to a HPLC pump and the MS. By means of an extraction piston any target zone can be eluted from a TLC/HPTLC plate directly into the MS. Example: for identification of the zone at hRf 15 in a standard mixture of caffeine, paracetamol, and acetylsalicylic acid the mass spectrum of the zone is recorded. After subtraction of a background spectrum a mass spectrum free from system peaks is obtained, which shows mainly substance signals - here the mass signal m/z 195 [M+H]+ for caffeine.

HPTLC 4e

103 073 Iris MEISEN et al., see section 28b

103 026 Gertrud MORLOCK*, Ute JAUTZ (*University of Hohenheim, Institute of Food Chemistry, Garbenstrasse 28, 70599 Stuttgart, Germany; gmorlock@uni-hohenheim.de): Comparison of two

different plunger geometries for HPTLC-MS coupling via an extractor-based interface. J. Planar Chromatogr. 21, 367-371 (2008). HPTLC of harmane (1-methyl-9H-pyrido[3,4-b]indole) and Glu-P-1 (2-amino-6-methyldipyrido[1,2-a:3‘,2‘-d]imidazole) on silica gel (prewashed with methanol) in a flat-bottom chamber with diethyl ether - methanol 49:1 at a relative humidity of 42 % and a temperature of 26 °C. Before development, the activity and pH of the silica gel were adjusted in a saturated twin trough chamber with 20 % aqueous ammonia (25 %) for 15 min. Quantitative determination by fluorescence measurement at UV 366/>400 nm. HPTLC-MS coupling via an extractor-based interface using a circular and an oval plunger for extraction of the adjacent heterocyclic amines. The circular plunger was easier to position, however the oval plunger was shown to be optimal (more selective) for adjacent bands.

qualitative identification, quantitative analysis, HPTLC 4e

103 027 T. MROCZEK*, Karine NDJOKO-IOSET, K. GLOWNIAK, Agnieszka MIETKIEWICZ-CAPALA, K. HOSTETTMANN (*Department of Pharmacognosy with Medicinal Plants Laboratory, Medical University, 1 Chodzki St., 20-093 Lublin, Poland): Investigation of Symphytum cordatum alkaloids by liquid-liquid partitioning, thin-layer chromatography and liquid chromatography - ion trap mass spectrometry. Anal. Chim. Acta 566 (2), 157-166 (2006). Extraction of pyrrolizidine alkaloids from the alkalised crude extract of Symphytum cordatum (L.) W.K. roots as free tertiary bases and polar N-oxides by one-step liquid-liquid partitioning and pre-fractionation by preparative multiple-development TLC on silica gel with chloroform - methanol - 25 % ammonia 50:5:1. Three alkaloid fractions of different polarities were obtained which showed different retention on silica gel: the most polar N-oxides had the highest retention, the tertiary bases had medium retention, and diesterified N-oxides had the lowest retention. Purification of the former fraction, which was reduced into free bases by sodium hydrosulfite, by liquid-liquid partitioning on Extrelut-NT3 cartridge. Further analysis by HPLC - ion-trap MS.

preparative TLC 4d, 22

103 028 G. VAN BERKEL*, J. LLAVE, M. DE APADOCA, M. FORD (*Organic and Biological Mass Spectrometry Group, Chemical Sciences Division, Oak Ridge National Laboratory, Oak Ridge, Tennessee 37831-6131, USA, vanberkelgj@ornl.gov): Rotation planar chromatography coupled on-line with atmospheric pressure chemical ionization mass spectrometry. Anal. Chem. 76, 479-482 (2004). Coupling of a rotation preparative layer chromatography system on-line with mass spectrometry using a simple plumbing scheme and a self-aspirating heated nebulizer probe of a corona discharge atmospheric pressure chemical ionization (APCI) source. The self-aspiration of the heated nebulizer delivers approx. 20 µL/min of the 3.0 mL/min eluate stream to the mass spectrometer, eliminating the need for an external pump in the system. The viability of the coupling is demonstrated with a three-dye mixture composed of fat red 7B, solvent green 3, and solvent blue 35 separated and eluted from a silica gel-coated rotor using toluene. The real-time characterization of the dyes eluting from the rotor is illustrated in positive ion full-scan mode. Other self-aspirating ion source systems including atmospheric pressure photoionization, electrospray ionization, and inductively coupled plasma ionization, for example, might be configured and used in a similar manner coupled to the chromatograph to expand the types of analytes that could be ionized, detected, and characterized effectively.

HPTLC, preparative TLC, APCI-MS 4e

103 029 G. VAN BERKEL*, M. FORD, M. DEIBEL (*Organic and Biological Mass Spectrometry Group, Chemical Sciences Division, Oak Ridge National Laboratory, Oak Ridge, Tennessee 37831-6131, USA, vanberkelgj@ornl.gov): Thin-layer chromatography and mass spectrometry coupled using desorption electrospray ionization. Anal. Chem. 77, 1207-1215 (2005). Desorption electrospray ionization (DESI) was demonstrated as a means to couple TLC with mass spectrometry. HPTLC of rhodamines on RP-2 and RP-8 with methanol - water 4:1 containing 200 mM ammonium acetate. HPTLC of FD&C dyes on RP-18 with water - acetone 7:3 containing 500 mM ammonium acetate. HPTLC of aspirin, acetaminophen and caffeine on silica gel with acetate - acetic acid 99:1. Tracks were scanned by moving the plate under computer control while directing the stationary DESI emitter charged droplet plume at the plate surface. Positioning of the DESI emitter,

plate surface, and the atmospheric sampling orifice of the mass spectrometer were found to be crucial for obtaining maximum analyte signal levels. Desorption ionization from all TLC phases was not equivalent.

HPTLC, DESI-MS 4e

103 064 G. VAN BERKEL et al., see section 22

103 161 G. XU et al., see section 32e

5. Hydrocarbons and halogen derivatives

103 030 Irena MALINOWSKA*, M. STUDZINSKI, H. MALINOWSKI (*Faculty of Chemistry, Department of Planar Chromatography, M. Curie-Sklodowska University, M. Curie-Sklodowska Sq. 3, 20-031 Lublin, Poland; irena.malinowska@poczta.umcs.lublin.pl): The effect of a magnetic field on the retention of polyaromatic hydrocarbons in planar chromatography. J. Planar Chromatogr. 21, 379-385 (2008). Magnetic fields can affect the retention and shape of the chromatographic bands of the solutes investigated. The effect depends on the type of mobile phase, the properties of the adsorbent layer and the mode of development of the chromatogram (development distance). TLC and HPTLC of diphenyl, pyrene, benzo[a]pyrene, phenanthrene, fluoranthene, and chrysene on silica gel with n-hexane, n-octane, carbon tetrachloride, cyclohexane, benzene, and toluene, and n-hexane - benzene and n-hexane - toluene binary mobile phases. Evaluation under UV light.

qualitative identification 5b

7. Phenols

103 031 T.H. DZIDO*, P.W. PLOCHARZ, A. KLIMEK-TUREK, A. TORBICZ, B. BUSZEWSKI (*Department of Physical Chemistry, Medical University, Lublin, Poland; tadeusz.dzido@am.lublin.pl): Pressurized planar electrochromatography as the mode for determination of solvent composition - retention relationships in reversed-phase systems. J. Planar Chromatogr. 21, 295-298 (2008). HPTLC of a test compound [1-(4-hydroxyphenylazo)-2-napthol] on RP-18 (prewashed with methanol) with acetonitrile - water - buffer (pH 4.8) in the desired volume ratio in a horizontal developing chamber saturated for 15 min. Quantitative determination by absorbance measurement with a diode array TLC scanner. The retention-composition relationship obtained with TLC is similar to those of pressurized planar electrochromatography and HPLC in the modifier concentration range 60-80 % but deviates substantially at higher modifier concentrations.

comparison of methods, qualitative identification, HPTLC, physicochemical, organic chemistry 7

103 032 Vesna GLAVNIK*, Breda SIMONOVSKA, Irena VOVK (*National Institute of Chemistry, Laboratory for Food Chemistry, Hajdrihova 19, 1000 Ljubljana, Slovenia): Densitometric determination of (+)-catechin and (-)-epicatechin by 4-dimethylaminocinnamaldehyde reagent. J. Chromatogr. A 1216(20), 4485-4491 (2009). TLC of (+)-catechin and (-)-epicatechin on cellulose with water. Detection with 4-dimethylaminocinnamaldehyde in HCl produced blue bands. Detection with vanillin reagent produced quickly fading red spots. Quantitative determination by absorbance measurement at 655 nm. Linearity was between 2 to 12 ng/zone and a polynomial regression fit from 2 to 30 ng/zone. The repeatability of the separation of 20 ng/zone was 3.5 % (% RSD, n = 6). The visible limit of detection of both standards was 1 ng/zone, the densitometric limit of detection was 0.2 ng/zone. The optimized 4-dimethylaminocinnamaldehyde reagent is superior to the more frequently used vanillin reagent and is applicable also for determination of mixtures containing other catechins, such as (-)-catechin, (-)-epicatechin gallate, (-)-epigallocatechin gallate, procyanidin A2, procyanidin B1 and procyanidin B2.

herbal, HPTLC, densitometry 7

103 033 Monika WAKSMUNDZKA-HAJNOS*, H. SMOLARZ, R. NOWAK (*Department of Inorganic and Analytical Chemistry, Medical Academy, Staszica 6, 20-081 Lublin, Poland: Chromatographic separations of phenolic acids by normal-phase-TLC. Retention behaviour on polar adsorbents (silica gel, aluminia, polyamide) with non-aqueous mobile phase. Acta Chromatographica 9, 38-54 (1999). TLC of salicylic acid, 3-hydroxybenzoic acid, 4-hydroxybenzoic acid, gentisic acid, protocatechuic acid, alpha-resorcylic acid, beta-resorcylic acid, gamma-resorcylic acid, gallic acid, vanillic acid, syringic acid, 3,5-dimethoxybenzoic acid, homoprotocatechuic acid, homogentisic acid, 4-coumaric acid, 2-coumaric acid, 3-coumaric acid, caffeic acid, ferulic acid, 3,4-dimethoxycinnamic acid, 2,5-dimethoxycinnamic acid, phloretic acid, hydrocaffeic acid, cinnamic acid on silica gel, alumina and polyamide layers with non-aqueous ternary mobile phases comprising a non-polar diluent (n-heptane), a polar modifier (2-propanol, dioxane, tetrahydrofuran or ethyl acetate) and 2 % glacial acetic acid to suppress the ionization of the solutes after preconditioning for 15 min in the mobile phase vapor. Detection at UV 254 nm and after derivatization by spraying with diazotized sulphanilic acid in 10 % sodium bicarbonate solution, or with a 2 % solution of iron chloride. The separation selectivity for benzoic acid derivatives was best on alumina for all the mobile phases investigated, especially for hydroxy derivatives. When methoxy derivatives of benzoic acid or cinnamic acid derivatives are separated the best selectivity was usually obtained on polyamide layers.

pharmaceutical research, herbal 7

8. Substances containing heterocyclic oxygen

103 034 Ildiko BROS*, M.L. SORAN, R.D. BRICIU, S.C. COBZAC (*National Institute for Research and Development of Isotopic and Molecular Technologies, 65-103 Donath Street, 400293 Cluj-Napoca, Romania; ildikobros@yahoo.com): HPTLC quantification of some flavonoids in extracts of Satureja hortensis L. obtained by use of different techniques. J. Planar Chromatogr. 22, 25-28 (2009). HPTLC of flavonoids (rosmarinic acid and luteoline) on silica gel with ethyl acetate - formic acid - water 136:5:6, ethyl acetate - methanol - water 77:13:10, ethyl acetate - diethyl ether 4:1, n-hexane - ethyl acetate - formic acid 60:40:3, chloroform - methanol - formic acid 882:60:47, and chloroform - acetone - formic acid 19:4:2. The mobile phase chosen for quantification was chloroform - ethyl acetate - formic acid 60:40:3. Detection by spraying with natural products reagent, followed by polyethylene glycol 400 reagent. Quantitative determination by densitometry at 328 nm (rosmarinic acid) and 349 nm (luteoline).

HPTLC, quantitative analysis, densitometry 8a

103 035 A. GUERRINI, R. BRUNI, S. MAIETTI, F. POLI, D. ROSSI, G. PAGANETTO, M. MUZZOLI, L. SCALVENZI, G. SACCHETTI* (*Department of Biology and Evolution, AgriUnife Center, University of Ferrara, C.so Ercole I d'Este 32, 44100 Ferrara, Italy, scg@unife.it): Ecuadorian stingless bee (Meliponinae) honey: A chemical and functional profile of an ancient health product. Food Chem. 114, 1413-1420 (2009). HPTLC of methanolic fractions of stingless bee honey samples and commercial samples from Apis mellifera (European honey bee) on silica gel with a five step development with two different mobile phases: ethyl acetate - formic acid - acetic acid - water 100:11:11:27 and toluene - ethyl acetate - acetic acid 10:9:1. Detection by fluorescence measurement at 400 nm and absorbance measurement at 240 nm, after fluorescence induction at 365 nm with a mercury vapor lamp. Detection of flavonoids by spraying with an aqueous solution of 4 % aluminium sulphate. Flavonoids and coumarins were identified by comparison with commercial standards.

food analysis, HPTLC, quantitative analysis, qualitative identification 8a

103 036 M. LIGOR, O. KORNYSOVA, A. MARUSKA, B. BUSZEWSKI* (*Chair of Environmental Chemistry and Bioanalysis, Faculty of Chemistry, Nicolaus Copernicus University, 7 Gagarin St, 87 100 Torun, Poland; bbusz@chem.uni.torun.pl): Determination of flavonoids in tea and Rooibos extracts by TLC and HPLC. J. Planar Chromatogr. 21, 355-360 (2008). TLC of flavonoids (myrecetin, rutin, catechin, quercetin, and kaempferol) on silica gel in a horizontal chamber with acetone - chloroform - water 8:2:1. Detection by dipping in natural products reagent followed by dipping in

PEG reagent (polyethylene glycol 400 in ethanol). Evaluation under UV 254 and 366 nm.

food analysis, qualitative identification 8a

103 037 K. WANG (Wang Kunbo), Z. LIU* (Liu Zhonghua), J. HUANG (Huang Jianan), D. FU (Fu Donghe), F. LIU (Liu Fang), Y.GONG (Gong Yushun), X. WU (Wu Xiasong) (*Laboratory of Tea Science of the Ministry of Education, Hunan Agricultural University, Furong District, Changsha, Hunan, 410128, China; zhonhualiu163@163.com): TLC separation of catechins and theaflavins on polyamide plates. J. Planar Chromatogr. 22, 97-100 (2009). TLC of (+)-catechin, (-)-epicatechin, (-)-epigallocatechin, (-)-epicatechin gallate, (-)-epigallocatechin gallate, theaflavin, theaflavin 3-gallate, theaflavin 3‘-gallate, and theaflavin 3,3‘-digallate on polyamide phase in a horizontal chamber (saturated for 15 min) by twofold development with chloroform - methanol 2:3 or n-butanol - acetone - acetic acid 5:5:3. Separation of the flavonols myricetin, quercetin, kaempferol, rutin and the phenolic acids gallic acid, chlorogenic acid, and caffeic acid was achieved by twofold development with chloroform - methanol 2:3. Detection by spraying with iron(III) chloride solution and evaluation under daylight. Quantitative determination by absorbance measurement at 600 nm.

food analysis, quantitative analysis, qualitative identification, densitometry 8a

10. Carbohydrates

103 038 D. CLINE, B. FRIED, J. SHERMA* (*Department of Chemistry, 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 determine the effect of starvation on the amounts of these carbohydrates. Acta Chromatographica 9, 79-86 (1999). HPTLC (TLC) of glucose, maltose, sucrose and trehalose on silica gel with acetonitrile - water 17:3 and ethyl acetate - acetic acid - methanol - water 12:3:3:2 for three times with chamber saturation; on amino plates with ethyl acetate - pyridine - water - acetic acid 12:6:2:1 and on cellulose plates with ethyl acetate - pyridine - water 2:1:2, both in a non-equilibrated chamber; on RP-18 with tetrahydrofuran - water 44:6 in a pre-equilibrated chamber. All plates were prewashed, e.g. by chromatography with dichloromethane - methanol 1:1. Detection by 4-aminobenzoic acid reagent, 1-naphthol-sulfuric acid reagent or aniline-DPA reagent, each followed by heating at 110 °C for 10 min. The combined results from comparison of hRf values for two different mobile phases on silica gel, on cellulose, amino phase and C18-bonded layers with diverse separation mechanisms, spiking experiments on silica gel, detection with selective reagents, and GC-MS analysis definitely proved the presence of maltose and glucose and the absence of trehalose in digestive gland-gonad complex and hemolymph samples from B. glabrata. Earlier papers reporting the presence of trehalose were undoubtedly in error.

HPTLC, densitometry, quantitative analysis, qualitative identification, postchromatographic derivatization 10a

103 039 Gertrud MORLOCK*, Shashi PRABHA (*Institute of Food Chemistry, University of Hohenheim, Garbenstrasse 28, 70599 Stuttgart, Germany, gmorlock@uni-hohenheim.de): Analysis and stability of sucralose in a milk-based confection by a simple planar chromatographic method. J. Agric. Food Chem. 55, 7217-7223 (2007). HPTLC of sucralose in burfi, a milk-based confection, on amino phase with acetonitrile - water 4:1 in a horizontal chamber up to a migration distance of 70 mm. Detection by thermal in situ derivatization, drying the plate for at least 5 min, followed by heating at 190 °C for 20 min. Quantitative determination by fluorescence measurement at UV 366/>400 nm. The within-run precision of sucralose determination in the matrix was 4.2 % and the mean recovery rate was 88 %. The limit of detection was 6 ng/band for standard solutions and 1 mg/kg for milk-based matrix. It was demostrated that over 300 runs can be performed within a day of labor with cost-effective instrumentation.

food analysis, HPTLC, quantitative analysis 10a

103 040 Katarina REIFFOVA*, Radomira NEMCOVA (*University of P.J. Safarik, Faculty of Natural Sci-

ences, Institute of Chemistry Sciences, Moyzesova 11, 040 01 Kosice, Slovak Republic): Thin-layer chromatography analysis of fructooligosaccharides in biological samples. J. Chromatogr. A 1110 (1-2), 214-221 (2006). Presentation of a rapid, simple and inexpensive method for the analysis of fructooligosaccharides as feed additives (prebiotics) in complicated biological samples with minimal pre-treatment. TLC of fructooligosaccharides in dietetic products and in samples from intestinal tract of monogastric animals on silica gel (impregnated with sodium acetate) with butanol - ethanol - water 5:3:2 with chamber saturation. Detection by spraying with diphenylamine - aniline - phosphoric acid in acetone. Quantitative determination by absorbance measurement at 370 nm.

quality control, clinical chemistry research, HPTLC, densitometry, qualitative identification, quantitative analysis, fructooligosaccharides 10

103 041 J. SHERMA*, D. ZULICK (*Department of Chemistry, Lafayette College, Easton, PA 18042-1782, USA): Determination of fructose, glucose and sucrose in beverages by high-performance thin-layer chromatography. Acta Chromatographica 6, 7-14 (1996). HPTLC of fructose, glucose and sucrose on a channeled silica gel plate with concentration zone with acetonitrile - water 17:3 for three times (freshly prepared each time, taking 15 min per run) with chamber saturation for 10-15 min. Before, just the silica gel layer was impregnated by spraying with 0.10 M sodium bisulfate solution, and subsequently with citrate buffer (1:10 dilution of citrate buffer with water, pH 4.8), each followed by drying. Detection by spraying with 1-naphthol-sulfuric acid reagent, followed by heating at 100-110 °C for 5-10 min. Quantitative determination by absorbance measurement at 515 nm. The hRf values of fructose, glucose and sucrose were 47, 43, and 28, respectively, and selectivity regarding matrix was given. No zones other than the sugars were detected in sample chromatograms because of the selectivity of the detection reagent and the retention of the beverage components in the preadsorbent. Correlation coefficients (r) for linear regression of the calibration curves typically ranged from 0.90-0.99, with an average of 0.97. The precision in matrix was 2.5 % (n = 5). The mean reproducibility of the twofold sample analyses was 4 %, ranged between 0.45 % and 7.5 %. The accuracy of the method was 94.6 %, 97.0 % and 90.8 % for sucrose, glucose and fructose, respectively. Sugar concentrations in the samples ranged from 18.4-34.3 mg/mL.

food analysis, HPTLC, densitometry, quantitative analysis, qualitative identification, postchromatographic derivatization 10a

103 042 Elena TAMBURINI*, T. BERNARDI, E. BIANCHINI, P. PEDRINI (*Agro-tech and Pharmaceutical Research Laboratory, Department of Evolutive Biology, University of Ferrara, Ferrara, Italy; tme@unife.it): Fermentation monitoring based on HPTLC-OPLC. The effect of a complex biological matrix on quantitative performance. J. Planar Chromatogr. 22, 9-14 (2009). HPTLC-OPLC of fructose, glucose, galactose, sucrose, lactose, 1-kestose, raffinose, nystose, and fructosil-nystose on silica gel with acetonitrile - water 17:3. After development and drying, the separated bands were derivatized by a thermal in-situ reaction on a hot plate. The plates were immersed in lead(IV) acetate-dichlorofluorescein reagent for 9 s and heated at 105 °C for 3 min. Quantitative determination by fluorescence measurement at 313 nm.

food analysis, quality control, densitometry, HPTLC, quantitative analysis 10a

11. Organic acids and lipids

103 017 W. CHAI et al., see section 4e

103 043 U. DISTLER, M. HÜLSEWIG, J. SOUADY, K. DREISEWERD, J. HAIER, N. SENNINGER, A. W. FRIEDRICH, H. KARCH, F. HILLENKAMP, S. BERKENKAMP, J. PETER-KATALINIC, J. MÜTHING* (*Institute of Medical Physics and Biophysics, University of Münster, 48149 Münster, Germany; jm@uni-muenster.de): Matching IR-MALDI-o-TOF mass spectrometry with the TLC overlay binding assay and its clinical application for tracing tumor-associated glycosphingolipids in hepatocellular and pancreatic cancer. Anal. Chem. 80, 1835-1846 (2008). HPT-

LC of neutral glycosphingolipids on silica gel with chloroform - methanol - water 120:70:17 and 120:85:20, both supplemented with 2 mM calciumchloride solution. Detection by treatment with orcinol. Quantitative determination by absorbance measurement at 544 nm (orcinol) and 630 nm (indolylphosphate). A TLC overlay assay was performed, the TLC immunodetection procedure used glycosphingolipid antibodies and toxins in conjunction with antitoxin antibodies.

clinical chemistry research, HPTLC quantitative analysis, qualitative identification 11e

103 020 K. DREISEWERD et al., see section 4e

103 044 B. FUCHS, J. SCHILLER*, R. SÜSS, A. NIMPTSCH, M. SCHÜRENBERG, D. SUCKAU (*Universität Leipzig, Medizinische Fakultät, Institut für Medizinische Physik und Biophysik, Härtelstr. 16-18, 04107 Leipzig, Germany; juergen.schiller@medisin.uni-leipzig.de): Capabilities and disadvantages of combined matrix-assisted laser-desorption/ionization time-of-flight mass spectrometry (MALDI-TOF MS) and high-performance thin-layer chromatography (HPTLC): Analysis of egg yolk lipids. J. Planar Chromatogr. 22, 35-42 (2009). HPTLC of phospholipids (lyso-phosphatidylcholine, sphingomyelin, phosphatidylcholin, lyso-phosphatidylethanolamine, phosphatidylinositol, phosphatidylethanolamine, phosphatidylserine, phosphatidic acid, and triacylglycerol) on silica gel with chloroform - ethanol - water - triethylamine 5:5:1:5. Detection by spraying with a solution of primulin (Direct Yellow 59). Evaluation under UV 366 nm. Detection by MALDI-TOF MS directly on the plates.

pharmaceutical research, clinical chemistry research, HPTLC, qualitative identification 11c

103 021 Beate FUCHS et al., see section 4e

103 022 Beate FUCHS et al., see section 4e

103 024 Vera IVLEVA et al., see section 4e

103 045 D.R. MASSA, J.D. VASTA, B. FRIED, J. SHERMA* (*Department of Chemistry, Lafayette College. Easton, PA, USA; shermaj@lafayette.edu): High-performance thin-layer chromatographic analysis of the polar lipid content of human urine and urine from BALB/c mice experimentally infected with Echinostoma caproni. J. Planar Chromatogr. 21, 337-341 (2008). HPTLC of phospholipids (cholesterol, phosphatidylethanolamine, phosphatidylcholine, and lysophosphatidylcholine) on silica gel plates with a concentration zone (prewashed with dichloromethane - methanol 1:1) with chloroform - methanol - water 65:25:4 in a saturated twin trough chamber. Detection by spraying with aqueous copper sulfate reagent followed by heating. Quantitative determination by absorbance measurement at 370 nm. Ninhydrin spray reagent was used to confirm the presence of phosphatidylethanolamine. The limit of quantification was 250 ng/spot.

HPTLC, quantitative analysis, densitometry 11c

03 046 A. ROHLFING, J. MÜTHING, G. POHLENTZ, U. DISTLER, J. PETER-KATALINIC, S. BERKENKAMP, K. DREISEWERD* (*Institute of Medical Physics and Biophysics, Westfälische Wilhelms-Universität Münster, Robert-Koch-Strasse 31, 48149 Münster, Germany; Klaus.Dreisewerd@uni-muenster.de): IR-MALDI-MS analysis of HPTLC-separated phospholipid mixtures directly from the TLC plate. Anal. Chem. 79, 5793-5808 (2007). HPTLC of phospholipids (phosphatidylcholine, phosphatidylethanolamine, phosphatidylglycerol, phosphatidic acid, cardiolipin, sphingomyelin) on silica gel in a saturated chamber with chloroform-methanol - 2-propanol - triethylamine - 0.25 % potassium chloride solution 30:9:23:18:6. Detection by dipping in molybdenum blue reagent for 1 s and quantitative evaluation by densitometry. The sensitivity was in the range of 10 to 150 pmol/spot (depending on phospholipid acidity, hRf value, and ion polarity).

HPTLC, quantitative analysis 11c

103 047 P.K. ZARZYCKI*, M.A. BARTOSZUK (*Toxicology and Bioanalysis Section, Department of Environmental Biology, Koszalin University of Technology, Sniadeckich 2, 75-453 Koszalin, Poland; pkzarz@wp.pl or pawel_k_z@hotmail.com): Improved TLC detection of prostaglandins by post-run derivatization with phosphomolybdic acid. J. Planar Chromatogr. 21, 387-390 (2008). TLC of prostaglandin PGE2 and PGF2a on silica gel and RP-18 in a saturated chamber with methanol - dichloromethane 1:9 and 100 % acetonitrile, respectively. Detection by spraying with phosphomolybdic acid (10 % in methanol) and heating at temperatures ranging from 80 to 100 °C (for silica gel) and below 80 °C (for RP-18). Densitometric evaluation; chromatographic spot quantification, however, was performed manually using the peak height above baseline method.

pharmaceutical research, densitometry, qualitative identification 11b

13. Steroids

103 048 W. KRZYCZKOWSKI*, E. MALINOWSKA, P. SUCHOCKI, J. KLEPS, M. OLEJNIK, F. HEROLD (*Department of Drug Technology, Faculty of Pharmacy, The Medical University of Warsaw, Warsaw, Poland, wkrzyczkowski@wum.edu.pl): Isolation and quantitative determination of ergosterol peroxide in various edible mushroom species. Food Chem. 113, 351-355 (2009). HPTLC of ergosterol peroxide from the mycelia of Hericium erinaceum (lion‘s mane mushroom), Laetiporus sulfureus (chicken mushroom), and Morchella esculenta (common morel), as well as in the fruiting bodies of Boletus edulis (king bolete), Suillus bovinus (Jersey cow mushroom), and B. badius (bay bolete) on silica gel with n-hexane - ethyl acetate 1:1. Detection by spraying with an alcoholic solution of phosphotungstic acid, followed by heating at 100 °C for 5 min. Quantitative determination by absorbance measurement at 515 nm. The hRf value was between 30 and 32. Selectivity regarding matrix was given. Linearity was between 0.125 and 1.00 µg/spot. The limit of detection was 50 ng/spot.

pharmaceutical research, food analysis, HPTLC, quantitative analysis 13c

103 049 P. ZARZYCKI*, Magdalena ZARZYCKA (*Section of Toxicology and Bioanalytics, Department of Environmental Biology, Koszalin University of Technology, Sniadeckich 2, 75-453 Koszalin, Poland, pkzarz@wp.pl): Application of temperature-controlled micro planar chromatography for separation and quantification of testosterone and its derivatives. Anal. Bioanal. Chem. 391, 2219-2225 (2008). HPTLC/TLC of testosterone and derivates (methyltestosterone, testosterone propionate, testosterone isobutyrate, testosterone phenylpropionate, testosterone isocaproate, testosterone enanthate, testosterone caprate on silica gel, RP-18W and aluminium oxide with a whole range of binary mixtures such as methanol/water, acetonitrile/water, methanol/dichloromethane and acetone/hexane 0 - 100 % on the effect of temperature ranging from -20 to +60 °C under saturated and unsaturated chamber conditions. Detection by dipping in 10 % methanolic phosphomolybdic acid, followed by heating at 100 °C for 10 min. Evaluation under UV 366 and 254 nm. The best separation was observed on RP-18W with methanol/water mobile phases.

quantitative analysis, HPTLC 13e

14. Steroid glycosides, saponins and other terpenoid glycosides

103 050 C. CHAICHAROENPONG*, A. PETSOM (*Institute of Biotechnology and Genetic Engineering, Chulalongkorn University, Bangkok 10330, Thailand, Chanya.C@Chula.ac.th): Quantitative thin layer chromatographic analysis of the saponins in tea seed meal. Phytochem. Anal. 20, 253-255 (2009). HPTLC of saponins in the tea seed meal of Camellia oleifera on silica gel with ethyl acetate - methanol - water 4:2:1. Quantitative determination by absorbance measurement at 214 nm. The hRf value of tea saponin was 40 and selectivity regarding matrix was given. The correlation coefficient was 0.9978 and the relative standard deviation 1.6 %. Linearity was between 0.9 and 22.2 µg/spot. The limit of detection and quantification was 870 ng and 2900 ng/spot, respectively.

HPTLC, agricultural, quantitative analysis, densitometry 14

15. Terpenes and other volatile plant ingredients

103 051 M. CHANAMA, T. WUNNAKUP, W. DE-EKNAMKUL, S. CHANAMA* (*Department of Biochemistry, Faculty of Science, Chulalongkorn University, Bangkok 10330, Thailand; suchart.c@chula.ac.th): Improvement of thin-layer chromatography for enzyme assay of geranylgeraniol 18-hydroxylase from Croton stellatopilosus Ohba. J. Planar Chromatogr. 22, 49-53 (2009). TLC of geranylgeraniol and plaunotol on silica gel with chloroform - n-propanol 24:1 or 48:1, or with ethyl acetate, in saturated chambers. Quantitative determination by absorbance measurement at 210 nm. Detection of plaunotol by exposure to iodine vapor for 10 min. The acyclic diterpenoid plaunotol present in Croton stellatopilosus leaves is a hydroxylation product, catalyzed by the enzyme geranylgeraniol-18-hydroxylase. The activity of the enzyme in cell-free extracts of C. stellatopilosus leaves was previously reported. In this study a new mobile phase (ethyl acetate) was used for determination of geranylgeraniol-18-hydroxylase in the 20,000 g and 100,000 g precipitates of the crude extracts. In addition ethyl acetate successfully separated plaunotol from various cytochrome P-450 inhibitors (ancymidol, metyrapone, and miconazole) frequently used for biochemical characterization of the hydroxylase enzymes.

pharmaceutical research, herbal, densitometry, qualitative identification 15a

103 052 P. RINTHONG, A. JINDAPRASERT, W. DE-EKNAMKUL* (*Department of Pharmacognosy, Faculty of Pharmaceutical Sciences, Chulalongkorn University, Bangkok 10330, Thailand; dwanchai@chula.ac.th): Simple densitometric TLC analysis of plaunotol for screening of high-plaunotol-containing plants of Croton stellatopilosus Ohba. J. Planar Chromatogr. 22, 55-58 (2009). TLC of plaunotol in leaves of Croton stellatopilosus Ohba, on silica gel with chloroform - n-propanol 24:1 in a twin trough chamber saturated for 30 min. Quantitative determination by absorbance measurement at 220 nm. Recovery was 98.7 %. The plaunotol content of the leaves of C. stellatopilosus was highly variable (0.14 to 0.79 % w/w of dry weight).

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

16. Nitro and nitroso compounds

103 053 T. WIDLA*, M. SLIWIOK (*Faculty of Law and Administration, Department of Criminalistics, Silesian University, 40-006 Katowice, Bankowa Street, Poland): Detection and determination of trotyl by HPTLC. Acta Chromatographica 6, 1-4 (1996). TLC of TNT (trotyl) on silica gel with hexane-benzene 1:1. Detection by spraying with phenol red, bromophenol blue, thymol blue, and bromothymol blue, separately, followed by heating at 100 °C for 10 min. These solutions were prepared immediately before spraying of the plates. The hRf value was 50 (± 2). Linearity was between 2.5 and 10 µg/zone. The correlation coefficient was 0.931. LOD (in average, n = 6) was 1.0 µg/zone (phenol red), 1.0 µg/zone (bromophenol blue), 1.5 µg/zone (thymol blue) and 1.5 µg/zone (bromothymol blue).

HPTLC, postchromatographic derivatization, explosives, TNT 16

17. Amines, amides and related nitrogen compounds

103 054 Zlatka BAJC*, K.S. GACNIK (*Institute for Food Hygiene and Bromatology, Veterinary Faculty, University of Ljubljana, Gerbiceva 60, 1000 Ljubljana, Slovenia; zlatka.bajc@vf.uni-lj.si): Densitometric TLC analysis of histamine in fish and fishery products. J. Planar Chromatogr. 22, 15-17 (2009). HPTLC of biogenic amines (spermidine, putrescine, cadaverine, histamine, and tyramine) on silica gel (with concentration zone) in a twin trough chamber saturated for at least 1 h with acetone - ammonia 19:1. Detection by heating at 75 °C for 2 min, followed by spraying with ninhydrin reagent (300 mg ninhydrin in 100 mL n-butanol - glacial acetic acid 97:3) and again heating at 75 °C for 2 min. Quantitative determination by absorbance measurement at 410 nm.

food analysis, densitometry, quantitative analysis, HPTLC 17a

103 055 R. HADDAD, H.M.S. MILAGRE, R.R. CATHARINO, M.N. EBERLIN* (*Thomson Mass Spectrometry Laboratory, Institute of Chemistry, State University of Campinas, 13084-971,

Campinas SP, Brazil; eberlin@iqm.unicamp.br): Easy ambient sonic-spray ionization mass spectrometry combined with thin-layer chromatography. Anal. Chem. 80, 2744-2750 (2008). TLC of mixtures of semipolar nitrogenated compounds as well as pharmaceutical drugs (allyl phenylamine, phenylamine, ethylpyridine, propanolol hydrochloride, and amlodipine besylate) on silica gel with ethyl acetate - hexane 1:4; detection under UV 254 nm. On-spot detection and analyte characterization via easy ambient desorption and sonic-spray ionization (EASI) and (tandem) mass spectrometry detection.

quality control, qualitative identification 17, 4e

103 056 S. LIN (Lin Shuyao), M. HUANG (Huang Minzong), H. CHANG (Chang Huichiu), J. SHIEA (Shiea Jentaie)* (*Department of Chemistry, National Sun Yat-Sen University, Kaohsiung Medical University Joint Research Center, Kaohsiung, 80424 Taiwan; jetea@mail.nsysn.edu.tw): Using electrospray-assisted laser desorption/ionization mass spectrometry to characterize organic compounds separated on thin-layer chromatography plates. Anal. Chem. 79, 8789-8795 (2007). TLC of dyes, amines, extracts of drug tablets (erythoxine B, erioglaucine, fast green FCF, 2,2‘-diaminodiethylamine, 3-quinolinamine, 2-acetylaniline; tablets containing DL-methylephedrin hydrochloride, caffeine, ethoxybenzamide, chlorpheniramine maleate, noscapine, acetaminophen) on RP-18 with 500 mM ammonium acetate - acetone 7:3. Separation of amines on silica gel with ethyl acetate - acetic acid - dichloromethane 98:1:1. The detection limit of TLC/ELDI/MS is 1 µM. toxicology,

quantitative analysis, qualitative identification 17

18. Amino acids and peptides, chemical structure of proteins

103 057 Rania BAKRY*, G.K. BONN, D. MAIR, F. SVEC (*Institute of Analytical Chemistry and Radiochemistry, Leopold Franzens University, 6020 Innsbruck, Austria; Rania.Bakry@uibk.ac.at): Monolithic porous polymer layer for the separation of peptides and proteins using thin-layer chromatography coupled with MALDI-TOF-MS. Anal. Chem. 79, 486-493 (2007). TLC of methylene blue and methyl red on monolithic phase with ethyl acetate - ethanol -water 6:4:3 and 3:2:1 with chamber saturation for 30 min. After development the plates were dried and scanned with MALDI. TLC separation of fluorescamine labeled proteins (insulin, cytochrome c, lysozyme, and myoglobin) with 40 or 55 % aqueous acetonitrile and 0.1 % trifluoro acetic acid with chamber saturation for 30 min. Detection under UV 366 nm. Preparation of the monolithic layer: The polymerization mixture consisted of butyl methacrylate, ethylene dimethacrylate, 1-decanol, cyclohexanol, and 2,2-dimethoxy-2-phenyl-acetophenone.

qualitative identification, postchromatographic derivatization 18b

103 058 Irena BARANOWSKA*, P. MARKOWSKI, A. WILCZEK, M. SZOSTEK, M. STADNICZUK (*Department of Analytical and General Chemistry, Faculty of Chemistry, Silesian University of Technology, M. Strzody Street 7, 44-100 Gliwice, Poland; irena.baranowska@polsl.pl): Normal and reversed-phase thin-layer chromatography in the analysis of L-arginine, its metabolites, and selected drugs. J. Planar Chromatogr. 22, 89-96 (2009). TLC of L-arginine and its metabolites on silica gel with methanol - 50 % acetic acid 3:1 and on RP-18 with 5 % acetic acid - methanol - acetonitrile 50:36:15. TLC of selected drugs (dexamethasone, prednisolone, furosemide, vancomycin, amikacin, fluconazole, digoxin, captopril, dipyrone, metoprolol, and sildenafil) on silica gel with acetonitrile - water 2:3 with chamber saturation for 1 h. Detection of L-arginine and its metabolites by spraying with a 1 % ethanolic solution of ninhydrin, followed by heating at 60 °C for 15 min. Additional detection by exposure to iodine vapor.

pharmaceutical research, qualitative identification 18a

103 059 A. MOHAMMAD*, A. ZEHRA (*Analytical Research Laboratory, Department of Applied Chemistry, Faculty of Engineering and Technology, Aligarh Muslim University, Aligarh, India; mohammadali4u@rediffmail.com): Separation of coexisting tryptophan, alanine, and phenylalanine or tyrosine by silver ion high-performance thin-layer chromatography. J. Planar Chromatogr. 21,

299-304 (2008). HPTLC of 21 amino acids on silica gel impregnated with 5 % Ag ion with borate phosphate buffer pH 2.3. After drying at 50 °C detection by spraying with ninhydrin. With this system separation of alanine, phenylalanine and tryptophane could be achieved, as well as separation of alanine, tyrosine and tryptophan. Separation of these amino acids was neither achieved on conventional silical gel (without Ag impregnation) nor by addtion of buffered 5 % silver nitrate solution (pH 2.3) to the mobile phase.

HPTLC, qualitative identification 18a

103 060 H.W. RAVN (Aarhus University, National Environmental Research Institute, Department of Terrestrial Ecology, PhytoChemLab, Vejlsoevej 25, P. O. Box 314, 8600 Silkeborg, Denmark, her@dmu.dk): Two new methods for early detection of the effects of herbicides in plants using biomarkers. J. Planar Chromatogr. 22, 65-71 (2009). Presentation of two simple and rapid HPTLC methods for early detection of the effects of herbicides using two different groups of plant biomarkers, which were developed as field tests (Herbicide Weed Response test - HWR-Test). Phytochemical changes can be detected before any morphological changes are visible on the plants. These changes are defined as biomarkers and can be detected by HPTLC-screening. After overall identification of the phytochemical biomarker pattern, two different biomarker groups, carbohydrates and amino acids, were detected using modified reagents for color reactions. Evaluation under daylight and videodensitometric analysis of digital images by VideoScan software. The screening method was previously described [H. W. Ravn, M. Hjorth, L. Lauridsen, P. Kudsk, S. K. Mathiassen, L. Mondolot, Bull. Environ. Contam. Toxicol. 75, 236-245 (2005)].

environmental, agricultural, HPTLC, qualitative identification 18a, 29d

21. Purines, pyrimidines, nucleic acids and their constituents

103 061 E. MINCSOVICS, K. PÁPAI, K. LUDÁNYI, Á. Z. DÁVID, M. BUDAI, I. ANTAL, I. KLEBOVICH* (*Semmelweis University, Department of Pharmaceutics, Högyes Endre Street 7, 1092 Budapest, Hungary; klebovich@gyok.sote.hu): Fully on-line hyphenation of an experimental OPLC separation unit with diode-array detection and mass spectrometry (OPLC-DAD-MS) for analysis of xanthine compounds. J. Planar Chromatogr. 21, 361-366 (2008). OPLC of xanthine standards (caffeine, theophylline, theobromine) and green tea extract on silica gel (prewashed with 15 mL acetonitrile - water 17:3) with chloroform - trifluoroacetic acid - acetonitrile - methanol 760:40:67:133. Quantitative determination by absorbance measurement at 280 nm. Xanthine standards were used as model compounds to test the connected systems OPLC-UV and OPLC-DAD-ESI-MS. Sensitivity of OPLC-UV or OPLC-DAD was increased by hyphenation with ESI-MS coupled in series. After background subtraction the extracted ion chromatogram (m / z = 181.1 Da) yielded well measurable peaks for theophylline and theobromine in tea leaf extract. Analysis time was 10 min only.

densitometry, quantitative analysis 21a

22. Alkaloids

103 062 S. BERKOV*, J. BASTIDA, M. NIKOLOVA, F. VILADOMAT, C. CODINA (*Departament de Productes Naturals, Biologia Vegetal i Edafologia, Facultat de Farmàcia, Universitat de Barcelona. Av. Joan XXIII s/n, 08028 Barcelona, Catalonia, Spain; berkov@ub.edu): Rapid TLC/GC-MS identification of acetylcholinesterase inhibitors in alkaloid extracts. Phytochem. Anal. 19, 411-419 (2008). TLC of alkaloid extracts on silica gel with ethyl acetate - methanol - 25 % ammonia 30:10:1 and n-hexane - ethyl acetate - methanol - 25 % ammonia 30:30:10:1. Detection of the separated compounds with acetylcholinesterase inhibitory activity and by spraying with Dragendorff reagent.

herbal, qualitative identification, preparative TLC, bioautography 22

103 063 J. MÄDER*, W. FISCHER, T. SCHNICK, L. W. KROH (*Berlin University of Technology, Institute of Food Technology and Food Chemistry, Department of Food Analysis, Gustav-Meyer-Allee

25, 13355 Berlin, Germany; J.Maeder@TU-Berlin.de): Changes in glycoalkaloid composition during potato processing: Simple and reliable quality control by HPTLC. J. Planar Chromatogr. 22, 43-47 (2009). HPTLC of alpha-solanine and alpha-chaconine on silica gel with dichloromethane - methanol - 2.5 % ammonia 175: 75:11 in a horizontal chamber saturated for 15 min. After drying and heating at 90 °C for 25 min detection by dipping twice in modified Carr-Price reagent (antimony(III) chloride in acetic acid - dichloromethane), followed by heating at 110 °C for 3 min. Quantitative determination by absorbance measurement at 560 nm. Linearity was between 30 and 700 ng. The limit of detection was 5-20 ng/zone depending on the sample matrix, the limit of quantification was 30 ng/zone. The ratio of alpha-chaconine and alpha-solanine was between 4:1 to 2:1 for all analyzed samples.

food analysis, quality control, densitometry, quantitative analysis, HPTLC 22

103 027 T. MROCZEK et al., see section 4d

103 064 G. VAN BERKEL*, B.A. TOMKINS, V. KERTESZ (*Organic and Biological Mass Spectrometry Group, Chemical Sciences Division, Oak Ridge National Laboratory, Oak Ridge, Tennessee 37831-6131, USA; vanberkel@ornl.gov): Thin-layer chromatography/desorption electrospray ionization mass spectrometry: Investigation of Goldenseal alkaloids. Anal. Chem. 79, 2778-2789 (2007). TLC of alkaloids (berberine chloride, palmatine chloride, hydrastine, tetrahydroberberine, hydrastinine hydrochloride and jatrorrhizine) on silica gel with ethyl acetate - methanol - formic acid - water 50:10:6:3. Detection under UV 254 nm. Detection levels were 5 ng/zone each or 14-28 pmol. Desorption electrospray ionization mass spectrometry was investigated as a means to qualitatively identify and to quantify analytes directly from developed normal-phase TLC plates.

food analysis, herbal, qualitative identification, quantitative analysis 22, 4e

23. Other substances containing heterocyclic nitrogen

103 065 B.B. DAUNDKAR. M.K. MALVE, R. KRISHNAMURTHY* (*Directorate of Forensic Science Laboratories, Home Dept., State of Maharashtra, Hans Bhugra Marg, Kalina, Vidyanagari, Santa Cruz (E), Mumbai 400098, India; dfsl@gmail.com): A specific chromogenic reagent for detection of diazepam among other benzodiazepines from biological and nonbiological samples after HPTLC. J. Planar Chromatogr. 21, 249-250 (2008). HPTLC of diazepam, oxazepam, nitrazepam, lorazepam, chlordiazepoxide, and flurazepam on silica gel in a saturated twin trough chamber with chloroform - methanol 9:1. Detection by spraying with 5 % sodium hydroxide solution followed by 1 % m-dinitrobenzene in dimethyl sulfoxide. Violet bands were obtained for diazepam, the other compounds did not react. The sensitivity of this reagent for diazepam is approx. 5 µg/spot.

toxicology, qualitative identification, HPTLC 23e

103 066 D. DI GREGORIO, H. HARNETT, J. SHERMA* (*Department of Chemistry, 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 Chromatographica 9, 72-78 (1999). HPTLC of dextromethorphan hydrobromide (DH) and clemastine fumarate (CF) on silica gel (prewashed by chromatography with dichloromethane - methanol 1:1) with ethyl acetate - methanol - ammonia 17:1:2 for DH, and dichloromethane - methanol - ammonia 90:10:1 for CF, both with chamber saturation. Quantitative determination by absorbance measurement at 225 nm (DH) and 216 nm (CF). The hRf value of DH was 55, and of CF 62. Selectivity regarding matrix was given. Linear correlation coefficient values were in the range of 0.990-0.999. The amount of DH in the analyzed caplets and gelcaps ranged from 100 to 114 % of the label values, precision (% RSD) ranged from 1.2 to 1.9 %, and recovery of spiked samples averaged 99.4 %. For CF, tablets assayed at 99.0-103 % relative to the label value, precision (% RSD) was 2.2 %, and recovery from a spiked sample was 97.9 %.

pharmaceutical research, HPTLC, densitometry, quantitative analysis 23e

103 067 J. HABDAS, Marzena PODGÓRNA* (*Insitute of Chemistry, University of Silesia, 9, Szkolna St., 40-006 Katowice, Poland; marzenapodgorna@wp.pl): Assessment of the lipophilic properties of selected porphyrins by thin-layer chromatography. J. Planar Chromatogr. 21, 259-261 (2008). TLC of 6 porphyrins (5,10,15,20-tetra-(4N-pyridyl)porphyrin, 5-(4-acetamidophenyl)-10,15,20-tri-(4N-pyridyl)porphyrin, 5,10-di-(4-acetamidophenyl)-15,20-di-(4N-pyridyl)porphyrin, 5,15-di-(4-acetamidophenyl)-10,20-di-(4N-pyridyl)porphyrin, 5,10,15-tri-(4-acetamidophenyl)-20-(4N-pyridyl)porphyrin, and 5,10,15,20-tetra-(acetamidophenyl)porphyrin) on RP-18 with methanol - chloroform 7:3. Visual detection. The lipophilic properties of the porphyrins were expressed by use of log P Recker, HLB (hydrophilic-lipophilic balance), and HK (determines the percentage contribution to hydrophilicity of one or more functional amido groups (-NH-CO-CH3))

physicochemial organic chemistry 23a

103 068 Ute JAUTZ, M. GIBIS, Gertrud MORLOCK* (*Institute of Food Chemistry, University of Hohenheim, Garbenstrasse 28, D-70599 Stuttgart, Germany, gmorlock@uni-hohenheim.de): Quantification of heterocyclic aromatic amines in fried meat by HPTLC/UV-FLD and HPLC/UV-FLD: a comparison of two methods. J. Agric. Food Chem. 56, 4311-4319 (2008). HPTLC of the heterocyclic aromatic amines (HAA) most frequently found in fried meat: 2-amino-1-methyl-6-phenylimidazol[4,5-b]pyridine (PhIP), 2-amino-3,8-dimethylimidazol[4,5-f]quinoxaline (MeIQx), 2-amino-3,4,8-trimethylimidazo[4,5-f]quinoxaline (4,8-DiMeIQx), 9H-pyrido[3,4-b]indole (norharman), and 1-methyl-9H-pyrido[3,4-b]indole (harman) using a previously validated method. Concentrations obtained by HPTLC were in a similar range as such obtained by HPLC with correlations of both methods between 0.8875 and 0.9751. The precision in meat matrix was between 7 and 49 % (HPTLC) and between 5 and 38 % (HPLC) at the very low µg/kg-levels formed during heating. Cost and time comparison showed that HPTLC is 4 times faster and 3 times less expensive than the HPLC reference method.

food analysis, HPTLC, quantitative analysis, comparison of methods 23e

103 069 P.K. SALO, S. VILMUNEN, H. SALOMIES, R.A. KETOLA, R. KOSTIAINEN* (*Faculty of Pharmacy, Division of Pharmaceutical Chemistry, and Drug Discovery and Development Technology Center, University of Helsinki, Helsinki, Finland; risto.kostiainen@helsinki.fi): Two-dimensional ultra-thin-layer chromatography and atmospheric pressure matrix-assisted laser desorption/ionization mass spectrometry in bioanalysis. Anal. Chem. 79, 2101-2108 (2007). TLC of benzodiazepines (midazolam, diazepam, lorazepam, oxazepam, N-desalkyl-flurazepam, triazolam, nitrazepam) on monolithic UTLC phase (prewashed with methanol) with dichloromethane - acetone 93:7 for one-dimensional and toluene - acetone - ethanol - 25 % ammonia 70:20:3:1 for two-dimensional separation in a saturated chamber. Developing distance 2 cm; separation time for 1-D chromatography 2-6 min, for 2-D 4-12 min. Quantitative determination by absorbance measurement at 254 nm; for visual detection the analytes were derivatized by spraying with Dragendorff reagent. Identification of the separated compounds by AP-MALDI-MS. The limits of detection were in the picomole range and thus low enough for bioanalysis.

clinical chemistry research, qualitative identification, postchromatographic derivatization 23e

103 070 F. TELLIER*, R. FRITZ, L. KERHOAS, P. DUCROT, J. EINHORN, A. SINCLAIR, P. LEROUX (*Department of Chemistry, Versailles Saint-Quentin-en-Yvelines University, 78001 Versailles, France, ftellier@versailles.inra.fr): Characterization of metabolites of fungicidal cymoxanil in a sensitive strain of Botrytis cinerea. J. Agric. Food Chem. 56, 8050-8057 (2008). HPTLC of [2-14C]-cymoxanil in the cultures (medium and mycelium) and in the cell-free extracts of Botrytis cinerea on silica gel with hexane - ethyl acetate - acetic acid 70:30:1. The most polar products were separated on RP-18 (impregnated with a methanolic solution of tetrabutylammonium bromide 70 mM and dried at 80 °C for 5 min) with phosphate buffer (0.01 M, pH 6) - methanol 11:9. Detection by autoradiography. The hRf value of cymoxanil was 35 on silica gel. Different metabolites were detected using both stationary phases.

agricultural, HPTLC, autoradiography, radioscanning, postchromatographic derivatization 23e

27. Vitamins and various growth regulators

103 071 I. BARANOWSKA*, A. KADZIOLKA (*Department of Analytical and General Chemistry, Silesian Technical University, Gliwice, Poland): RPTLC and Derivative Spectrometry for the Analysis of Selected Vitamins. Acta Chromatographica 6, 1-4 (1996). TLC of 5 water-soluble vitamins (vitamin C, nicotinic acid, nicotinamide, vitamin B1, rutin) on RP-18 with with water-methanol 5:4 and water-acetic acid 7:1, and of 4 fat-soluble vitamins (A-acetate, E, E-acetate and D3) on RP-18 with acetonitrile - benzene - chloroform 10:10:1. Detection of vitamin C, nicotinic acid, nicotinamide and vitamin B1 with a solution of potassium hexaiodoplatinate(IV) (10 % potassium iodide with 5 % hexachloroplatinic acid in the ratio 9:1). Detection of rutin with 25 % lead(II)acetate. Detection of fat-soluble vitamins with a 10 % solution of antimony chloride. The hRf value of water-soluble vitamins (water - methanol 5:4; water - acetic acid 7:1) was 94 and 91 (vitamin C), 76 and 57 (nicotinic acid), 71 and 40 (nicotinamide), 0 and 68 (vitamin B1), and 28 and 0 (rutin). The hRf value of fat-soluble vitamins was 80 (vitamin E and vitamin E-acetate), 62 (vitamin D3), and 86 (vitamin A-acetate). Derivative spectrophotometry (up to fifth-order spectra) was applied to the determination of vitamins B1, B6 and A-acetate in mixtures with other vitamins.

pharmaceutical research, food analysis 27

103 072 Alina PYKA (Department of Analytical Chemistry, Faculty of Pharmacy, Medical University of Silesia, Jagiellonska 4, 41-200 Sosnowiec, Poland; alinapyka@wp.pl; apyka@sum.edu.pl): Evaluation of the lipophilicity of fat-soluble vitamins. J. Planar Chromatogr. 22, 211-215 (2009). TLC of fat soluble vitamins (ergocalciferol, cholecalciferol, (+/-)-alpha-tocopherol, tocopherol acetate, retinol, retinol acetate, retinol palmitate, menadione, and phytonadione) on RP-8 and RP-18 (prewashed with methanol) with methanol - water in different volume proportions, with chamber saturation for 20 min at ambient temperature. Determination of hRf values by densitometry. Linear relationships were obtained between the RM values of the fat-soluble vitamins and the volume fraction of methanol in the mobile phase.

pharmaceutical research, qualitative identification, physicochemial organic chemistry 27

28. Antibiotics, Mycotoxins

103 073 Iris MEISEN*, U. DISTLER, J. MÜTHING, S. BERKENKAMP, K. DREISEWERD, W. MATHYS, H. KARCH, M. MORMANN (*Institute for Hygiene, University of Muenster, Robert-Koch-Str. 41, 48149 Muenster, Germany; meisen@uni-muenster.de): Direct coupling of high-performance thin-layer chromatography with UV spectroscopy and IR-MALDI orthogonal TOF MS for the analysis of cyanobacterial toxins. Anal. Chem. 81, 3858-3866 (2009). HPTLC of microcystin LR and nodularin on silica gel with 1-propanol - ethyl acetate - water 3:5:2 with 5 % acetic acid. Detection and quantification by UV spectroscopy at 232 nm and direct identification of separated analytes on the HPTLC plate by IR-MALDI-o-TOF MS. The detection limit was 3-5 ng/zone. For detection of peptides, plates were cut and sprayed with a solution of p-anisaldehyde, followed by heating at 105 °C until purple-blue peptide spots appeared.

toxicology, agricultural, HPTLC, quantitative analysis 28b, 4e

29. Pesticides and other agrochemicals

103 074 M. GÖCER, K. HOFERER, J. ZIPFEL, B. SPANGENBERG* (*University of Offenburg, Institute of Process Engineering, Badstrasse 24, 77652 Offenburg, Germany; Spangenberg@FH-Offenburg.de): A new TLC method for quantification of paraquat, diquat, difenzoquat, mepiquat, and chloromequat in water. J. Planar Chromatogr. 22, 59-63 (2009). HPTLC of paraquat, diquat, difenzoquat, mepiquat, and chloromequat on LiChrospher silica gel with 1-propanol - methanol - 2.5 M aqueous sodium chloride 1:1:3. Detection by immersion for 2 s in a solution of 50 mg sodium tetraphenyl borate in 50 mL of water containing 50 µL concentrated hydrochloric acid. The wet plate was illuminated for 5 min with UV light at 254 nm which resulted in the formation of fluorescing spots corresponding to mepiquat, chloromequat, and difenzoquat. Immediately after illuminating at 254 nm all spots were illuminated for 10 min with UV light at 365 nm, which

converted all compounds into fluorescent derivatives. Then the dry plate was dipped into ethylene glycol - methanol 1:1 for 2 s, which enhanced the fluorescence by a factor of two. Detection by fluorescence measurement and averaged densitograms were obtained in the emission wavelength range from 440 to 480 nm.

environmental, toxicology, food analysis, HPTLC, quantitative analysis, densitometry 29d

103 075 Malgorzata JANICKA*, E. TYIHÁK, À. M. MÓRICZ, B. OSZIK-MENDYK (*Faculty of Chemistry, Maria-Sklodowska University, M. Curie-Sklodowska Sq. 3, 20-031 Lublin, Poland; mjanicka@hermes.umes.lublin.pl): Crucial role of formaldehyde and its reaction products in the antiproliferative activity of some potential pesticides. J. Planar Chromatogr. 21, 161-166 (2008). TLC of 13 newly synthesized potential herbicides (N-aryltrichloroacetamides or 2-(chlorophenoxy)acyl derivatives) on silica gel or RP-18 in saturated sandwich chambers at 20 °C with hexane - 1,2-dichloroethane 2:3. Detection in UV light at 254 nm. The lipophilicity of the substances was described by retention factors in water, log k w, both calculated from experimental RP-TLC data, and by log P values calculated by use of software. Biological activity was determined with the BioArena system. Antibacterial action against Pseudomonas savastanoi pv. phaseolicola bacterial cells and the role of formaldehyde were investigated. Additionally the effect of formaldehyde capturers (L -arginine and reduced glutathione) and Cu 2+ ions was evaluated in regard of bioactivity and toxicity. Formaldehyde and its reaction products are crucial in the action mechanism of these substances.

agricultural, qualitative identification 29a

103 076 K.V. KULKARNI*, D.B. SHINDE, D.V. MANE, M.V. GARAD (*Regional Forensic Science Laboratory, Dindori Road, Panchvati, Nasik 431004, India; krishnakulkarni96@yahoo.com): A new chromogenic spray reagent for detection and identification of monocrotophos. J. Planar Chromatogr. 22, 133-135 (2009). HPTLC of monocrotophos on silica gel with chloroform - acetone 7:3 with chamber saturation. Detection by spraying with 20 % sodium carbonate followed by acidic methanolic iron chloride reagent. Evaluation of purple spots, while organophosphorus, organochlorine, carbamate, and pyrethroid insecticides were not detected as purple spots. Alkaline hydrolysis of monocrotophos yields one molecule each of O,O-dimethylphosphoric acid and N-methylacetoacetamide. After acidiification N-methylacetoacetamide yields the enol form of monomethylamide which reacts with ferric ions to a purple complex. The detection limit for monocrotophos is 0.5 µg/zone.

agricultural, toxicology, HPTLC, qualitative identification 29b

103 060 H.W. RAVN, see section 18a

103 077 U. TAMRAKAR, V.K. GUPTA, A.K. PILLAI* (*Chemistry Department, Government V.Y.T. P.G. Autonomous College, Durg (Chhattisgarh) 491001, India; drajaipillai@gmail.com): Spectrophotometric analysis of carbamate pesticides after thermal gradient separation. J. Planar Chromatogr. 22, 77-82 (2009). Detection of carbamates (carbaryl, propoxur, and carbosulfan) on silica gel activated at 110 °C for 5 min in the oven. After application the pesticide solutions were hydrolyzed by addition of 2 µL of 2 M sodium hydroxide solution. After 5 min, 5 µL diazotized p-aminoacetanilid was applied onto the spots to form the colored derivatives - red, yellow, and yellow-orange for carbaryl, propoxur, and carbosulfan, respectively. The plates were then placed horizontally in an oven at 110 °C and the colored derivatives migrated as concentric rings under the influence of the temperature gradient.

agricultural, qualitative identification 29b

30. Synthetic and natural dyes

103 078 C.L. BROSSEAU, A. GAMBARDELLA F. CASADIO, C.M. GRZYWACZ, J. WOUTERS, R.P. VAN DUYNE* (*Department of Chemistry, Northwestern University, 2145 Sheridan Road,

Evanston, Illinois 60208, USA; vanduyne@northwestern.edu): Ad-hoc surface-enhanced raman spectroscopy methodologies for the detection of artist dyestuffs: thin-layer chromatography-surfance enhanced raman spectroscopy and in situ on the fiber analysis. Anal. Chem. 81, 3056-3062 (2009). TLC of organic dyes and pigments (alizarin, purpurin, carminic acid, lac dye, crocin, Cape jasmin) on silica gel with toluene - acetic acid - methanol 9:2:2. After sample application a centrifuged citrate-reduced silver colloid was spotted on top of the applied zones. SER spectra were recorded soon after application of the colloids.

environmental 30a, 4e

103 079 J.E. CLARK, Susan V. OLESIK* (*Department of Chemistry, The Ohio State University, 100 West 18th Ave, Columbus, Ohio 43210, USA; olesik@osu.edu): Technique for ultra thin layer chromatography using an electrospun, nanofibrous stationary phase. Anal. Chem. 81, 4121-4129 (2009). The polymer used for electrospinning the new stationary phase was polyacrylonitrile with a molecular weight of ca.150.000 in dimethylformamide as solvent; the substrate for the fibers was aluminium foil. The new material showed fiber diameters that are 400 nm. The most efficient layer thickness was 25 µm. The eletrospun plate, which was a cut, rectangular plate roughly sized 3 × 6 cm, was compared to (1) TLC on silica gel with acetonitrile - methanol 3:2 for mixtures of laser dyes (sulforhodamine 640, pyrromethene 597, rhodamine 610 perchlorate, rhodamine 610 chloride, rhodamine 590 chloride, rhodamine 101, and kiton red), and (2) TLC on cyano phase with water - acetone 7:3 for steroidal compounds (androsterone, cholesterol, and cortisone), both with chamber saturation for 10 min. The development time was comparatively faster. Detection under UV light and by spraying with molybdenum phosphoric acid and heating at 120 °C for 10 min. Plate heights (Note: referred to the migration distance of the solvent front instead of substance zone) of max. 120.000 per meter resulted and were comparatively better. Two-dimensional TLC had a similar efficiency compared to one-dimensional separation (no major peak broadening was observed).

UTLC 30a, 3d

103 080 B.H. HAN, I. MANNERS, M.A. WINNIK* (*Department of Chemistry, University of Toronto, 80 St. George Street, Toronto, ON M5S 3H6, Canada): Phosphorescence quenching of dyes adsorbed to silica thin-layer chromatography plates. Anal. Chem. 77 (24), 8075-8085 (2006). Description of photoluminescence quenching experiments by oxygen for a series of transition metal dyes adsorbed on silica gel. The quenching kinetics showed differences regarding the behavior of the same dyes adsorbed to the well-defined surfaces of SBA-15 mesoporous silica particles and adsorbed to a thin layer-by-layer polymer film. Pore size and pore size distribution are larger for TLC silica than for mesoporous silica. On TLC silica the dye photoluminescence decay profiles show smaller deviations from an exponential form, and larger differences between the intensity and lifetime Stern-Volmer plots. Dyes adsorbed to TLC silica gel are three times more sensitive to quenching by oxygen.

photoluminescence quenching 30

103 081 Maria-Loredana SORAN*, I. BROS, E. SURDUCAN, V. SURDUCAN (*National Institute of Research and Development for Isotopic and Molecular Technology, 72-103 Donath Street, 400293 Cluj-Napoca, Romania; loredana_soran@yahoo.com): Microwave assisted thin-layer chromatography - an improved separation technique. J. Planar Chromatogr. 21, 243-248 (2008) TLC plates were placed perpendicular or parallel to 2.45 +/- 0.5 GHz electromagnetic waves (microwaves) to improve chromatographic resolution. Perpendicular arrangements led to the greatest improvement. Compared with separation under normal conditions in microwave assisted TLC the migration distance of target compounds was increased, the migration distance of the mobile phase was decreased and the spot size was reduced. TLC of a lipophilic test dye mixture (indophenol blue, Sudan red G, 4-dimethylaminoazobenzene), a hydrophilic test mixture (brilliant black BN, amaranth S 75, fast yellow, chryosine) and a pesticide mixture (cymoxanil, kelthane, triadimefon, and trifluralin) on silica gel and cellulose with toluene, n-propanol - water in different ratios, and n-hexane - acetone 2:1. Detection of pesticides under UV light at 254 nm.

microwave assisted thin-layer chromatography 30a

32. Pharmaceutical and biomedical applications

103 082 E.A. ABOURASHED*, M.M. HEFNAWY, H.I. EL-SUBBAGH (*ElSohly Laboratories, 5 Industrial Park Drive, Oxford, MS 38655, USA; eabourashed@elsohly.com): HPTLC analysis of a new ultra-short-acting thiazolodiazepine hypnotic (HIE-124) in spiked human plasma. J. Planar Chromatogr. 22, 183-186 (2009). HPTLC of ethyl 8-oxo-5,6,7,8-tetrahydrothiazolo[3,2-a][1,3]diazepin-3-carboxylate (HIE-124) in plasma (with diazepam as internal standard) on silica gel with chloroform - ethyl acetate 4:1 with chamber saturation for 30 min. Quantitative determination by absorbance measurement at 265 nm. The limit of detection and quantification was 20 µg/L (40 ng/band) and 40 µg/L (80 ng/band), respectively.

pharmaceutical research, clinical chemistry research, HPTLC, quantitative analysis, densitometry 32a

103 083 R. AN (An Rui)*, S. ZHOU (Zhou Sili), L. YOU (You Lisha), X. WANG (Wang Xinhong) (*Shanghai Univ. TCM, Shanghai 201203, China): (Study of the quality standard for compound Shenyi granules) (Chinese). J. Chinese Trad. Patent Med. 30 (12), 1781-1785 (2008). TLC of Shenyi granule extracts on silica gel with 1) dichloromethane - ethyl acetate - petroleum ether (60-90 ºC) - methanol 20:14:6:1; 2) ethyl acetate - formic acid - glacial acetic acid - water 30:3:4:3; 3) toluene - ethyl acetate - formic acid 8:6:1; 4) chloroform - methanol - water 26:10:3. Detection 1) by spraying with 10 % sulfuric acid in ethanol and heating; 2) by spraying with iron(III) chloride solution; 3) by spraying with vanillin reagent. Identification by comparison with the standards glycyrrhizic acid and glycyrrhetinic acid.

pharmaceutical research, traditional medicine, quality control, HPTLC, quantitative analysis, qualitative identification 32e

103 084 S. CHOPRA*, F.J. AHMAD, R.K. KHAR, S.K. MOTWANI, S. MAHDI, Z. IQBAL, S. TALEGAONKAR (*Department of Pharmaceutics, Faculty of Pharmacy, Jamia Hamdard, Hamdard Nagar, New Delhi 110062, India): Validated high-performance thin-layer chromatography method for determination of trigonelline in herbal extract and pharmaceutical dosage form. Anal. Chim. Acta 577 (1), 46-51 (2006). HPTLC of trigonelline in herbal extracts and in pharmaceutical dosage forms on silica gel with n-propanol - methanol - water 4:1:4. Quantitative determination by absorbance measurement at 269 nm. Linearity was between 100 - 1200 ng/spot (via peak area) and the correlation coefficient was 0.9991. The trigonelline content of herbal extracts quantified and estimated from the formulation was found to be well within limits (± 5 %) of the labeled content of the formulations. The method is reproducible and selective for the estimation of trigonelline by statistical analysis of the data.

pharmaceutical research, traditional medicine, quality control, HPTLC, densitometry, quantitative analysis, qualitative identification 32e

103 085 V.P. CHOUDHARI*, Anna P. NIKALJE (*Maharashtra Institute of Pharmacy, MIT Campus, Paud Road, Kothrud, Pune, 411038, Maharashtra, India): Stability-indicating TLC method for the determination of dutasteride in pharmaceutical dosage forms. Chromatographia 70 (1-2), 309-313 (2009). TLC on silica gel with acetonitrile - methanol - dichloromethane 2:1:2. The hRf value of dutasteride was 64. Separation of dutasteride from its degradation products (produced by acid and alkali hydrolysis, oxidation, photo degradation, dry and wet heat treatment) was good. Quantitative determination by absorbance measurement at 244 nm. Linearity was in the range of 100 - 600 ng/band and the correlation coefficient was 0.9943 (via peak area) The limit of detection and quantitation was 7 and 23 ng/band.

quantitative analysis, qualitative identification, comparison of methods 32c

103 086 L. CIESLA, A. PETRUCZYNIK, M. HAJNOS, A. BOGUCKA-KOCKA, Monika WAKSMUNDZKA-HAJNOS* (*Department of Inorganic Chemistry, Medical University, 20-081 Lublin, Poland; monika.hajnos@am.lublin.pl): Two-dimensional thin-layer chromatography of structural analogs. Part I: Graft TLC of selected coumarins. J. Planar Chromatogr. 21, 237-241

(2008). HPTLC of coumarins (present in extracts of Archangelica officinalis, Pastinaca sativa and Heracleum sphondylium fruits) on cyano phase with acetonitrile - water 3:7 (triple development) in the first dimension, and on silica gel with ethyl acetate - n-heptane 7:13 (triple development) in the second dimension. Good selectivity differences were also obtained on silica gel with ethyl acetate - n-heptane 7:13 (triple development) in the first dimension and on RP-18 phase with methanol - water 11:9 (triple development) in the second dimension. Detection and quantitative determination by fluorescence measurement at 366 nm.

food analysis, HPTLC, qualitative identification, densitometry 32e

103 087 S. CORAN*, M. ALBERTI, V. GIANNELLINI, A. BALDI, G. PICCHIONI, F. PAOLI (*Dept. of Science Pharmaceutical, University of Firenze, Via G. Capponi 9, 50121 Firenze, Italy): Development of a densitometric method for the determination of cephalexin as an alternative to the standard HPLC procedure. J. Pharm. Biomed. Anal. 18, 271-274 (1998). HPTLC of cephalexin on silica gel (prewashed with the mobile phase) with ethyl acetate - acetic acid - water 7:2:1. Quantitative determination by absorbance measurement at 263 nm. The method was linear in the range of 200-1600 ng/spot, average recovery was 101 %. The analytical results obtained by HPTLC were comparable with the HPLC method of USP XXIII. The HPTLC method was suggested as alternative to the USP method considering the high throughput.

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

103 088 S. DATTA*, D. GUPTA, Pinki DATTA (*Dept. of Pharmacy, Bharat Institute of Technology, Bypass road, Meerut 250103, India): Pharmacognostical studies on the leaves of Viola odorata. Abstract No. 9147, IHCB (2009). HPTLC of quercitin in methanolic leaf extracts of Viola odorata on silica gel with ethyl acetate - formic acid - glacial acetic acid - water 100:11:11:26. Quantitative determination by absorbance measurement at 366/>400 nm for quantification. The extract contained 0.36 % quercetin.

pharmaceutical research, herbal, HPTLC, densitometry, quantitative analysis 32e

103 089 Namita DESAI*, Purnima AMIN (*Pharmaceutical Sciences & Technology Div., University Institute of Chemical Technology University of Mumbai, Mantunga, Mumbai 400019, India): Stability indicating HPTLC determination of meloxicam. Ind. J. Pharm. Sci. 70(5), 644-647 (2008). HPTLC of meloxicam on silica gel with ethyl acetate - cyclohexane - glacial acetic acid 325:175:1 with chamber saturation for 45 min. Quantitative determination by absorbance measurement at 353 nm. The method was linear in the range of 100-500 ng/zone, recovery was 100.3 %. The method was evaluated for stability (acid, base, thermal, oxidative, photodegradation) and degradation products were well separated from the main drug.

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

103 090 S.R. DHANESHWAR*, N.G. PATRE*, M.V. MAHADIK (*Bharati Vidyapeeth University, Poona College of Pharmacy, Department of Pharmaceutical Chemistry, Pune 411038, India): Stability-indicating HPTLC method for quantitation of quetiapine fumarate in the pharmaceutical dosage form. Acta Chromatographica 21 (1), 83-93 (2009). HPTLC of quetiapine fumarate on silica gel with toluene - methanol 4:1. The hRf value of quetiapine fumarate was 37. Quantitative determination by absorbance measurement at 254 nm. There was no chromatographic interference from tablet excipients. The drug is susceptible to treatment with acid and alkaline hydrolysis, oxidation, and photodegradation. The method was able to separate the degradation products from the pure drug, it can be used for stability tests.

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

103 091 Virginie ESTERS*, L. ANGENOT, Viviane BRANDT, M. FRÉDÉRICH, Monique TITS, CH.

VAN NERUM, J.N. WAUTERS, P. HUBERT (*Laboratory of Pharmacognosy, Department of Pharmacy, University of Liège, CHU, B36, Avenue de l'Hôpital 1, 4000 Liège, Belgium): Validation of a high-performance thin-layer chromatography/densitometry method for the quantitative determination of glucosamine in a herbal dietary supplement. J. Chromatogr. A 1112 (1-2), 156-164 (2006). HPTLC of glucosamine in a dietary supplement containing dried extracts of the main plants traditionally used for rheumatic disorders, on silica gel with a saturated mixture of 2-propanol - ethyl acetate - ammonia (8 %) 1:1:1. Detection by dipping into a modified anisaldehyde reagent and heating at 120 °C for 30 min in a drying oven. Quantitative determination by absorbance measurement at 415 nm. Validation of the method by applying the novel validation protocol proposed by a commission of the Société Française des Sciences et Techniques Pharmaceutiques. Relative standard deviations for repeatability and intermediate precision were between 4.9 and 8.6 %, accuracy was good, the two-sided 95 % beta-expectation tolerance interval was within the acceptance limits of 85 and 115 % on the whole analytical range (800 - 1200 ng of glucosamine).

pharmaceutical research, quality control, traditional medicine, HPTLC, quantitative analysis, qualitative identification, densitometry, glucosamine 32e

103 092 A.R. FAKHARI*, A.R. KHORRAMI, M. SHAMSIPUR (*Department of Chemistry, Shahid Beheshti University, Tehran, Iran): Stability-indicating high-performance thin-layer chromatographic determination of levonorgestrel and ethinyloestradiol in bulk drug and in low-dosage oral contraceptives. Anal. Chim. Acta, 572 (2), 237-242 (2006). Presentation of a stability-indicating method for simultaneous determination of the steroidal hormones levonorgestrel and ethinyloestradiol both in bulk drug and in low-dosage oral contraceptives by HPTLC on silica gel with hexane - chloroform - methanol 4:12:1. The hRf value of levonorgestrel was 65 and of ethinyloestradiol 43. The compounds were well separated from their degradation products. Quantitative determination by absorbance measurement at 225 nm. Linearity of levonorgestrel and ethinyloestradiol was 200-800 and 40-160 ng/spot, respectively.

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

103 093 Izabela FECKA (Department of Pharmacognosy, Wroclaw Medical University, pl. Nankiera 1, 50-140 Wroclaw, Poland; izabela@farmgn.am.wroc.pl): Qualitative and quantitative determination of hydrolysable tannins and other polyphenols in herbal products from meadowsweet and dog rose. Phytochem. Anal. 20, 177-190 (2009). TLC and HPTLC of polyphenols (tellimagrandins I and II, rugosins A, B, D and E and other) in commercially available products of meadowsweet (Filipendula ulmaria) and dog rose (Rosa canina) on silica gel, LiChrospher Si 60, RP-18 and amino phase with tetrahydrofuran - acetonitrile - water 3:1:1, of tannins on amino phase with acetone - formic acid 3:1, of flavonols with acetone - formic acid 17:3, of tannins with diisopropyl ether - acetone - formic acid - water 4:4:1:1, and of flavonols with diisopropyl ether - acetone - formic acid - water 5:3:1:1. Detection of polyphenolic compounds under UV light at 254 and 366 nm before and after spraying with natural products reagent followed by polyethylene glycol reagent. Detection in white light after spraying with 1 % iron(III) chloride (for tannins and phenolic acids), vanillin-hydrochloric acid (flavonols) or bis-diazotised sulfanilamide (for all polyphenols). Quantitative determination of polyphenol contents by HPLC with UV detection. Meadowsweet flowers and rose hips with seeds yielded 55.5-124.8 and 0.4-1.3 mg/g of ellagitannins, respectively. The sum of detected polyphenols was 83.9-165.7 mg/g for Filipendulae ulmariae flos and 1.2-2.7 mg/g for Rosae pseudo-fructus cum fructibus.

herbal, traditional medicine, food, analysis, HPTLC, qualitative identification 32e

103 094 Jolanta FLIEGER*, P. PANETH, K. GIELZAK-KOCWIN, M. TATARCZAK (*Department of Inorganic and Analytical Chemistry, Medical University of Lublin, 20-081 Lublin, Staszica 6, Poland; j.flieger@am.lublin.pl): Micropreparative isolation of Cu(II) complexes of isoniazid and ethambutol and determination of their structures. J. Planar Chromatogr. 22, 83-88 (2009). TLC of isoniazid, pyrazinamide, ethambutol, and aminosalicylic acid on RP-18 in a horizontal chamber at 20 °C with acetonitrile - water 3:7. The mobile phase was modified by adding copper(II) chlo-

ride to the mixture at a constant concentration of 0.05 M. Detection under UV light at 254 nm. Quantitative determination by absorbance measurement in the range 200 - 700 nm with a TLC scanner equipped with a diode-array detector.

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

103 095 M. GANDHIMATHI*, T. K. RAVI (*Department of Pharmaceutical Analysis, College of Pharmacy, Sri Ramakrishna Institute of Paramedical Sciences, Coimbatore 641 044, India; gands72@yahoo.co.in): Simultaneous densitometric analysis of tramadol hydrochloride and chlorzoxazone by high-performance thin-layer chromatography. J. Planar Chromatogr. 21, 305-307 (2008). HPTLC of tramadol hydrochloride and chlorzoxazone on silica gel, prewashed with methanol, in a twin trough chamber, saturated for 10 min, with ethyl acetate - toluene - ammonia 35:15:1. Quantitative determination by absorbance measurement at 273 nm.

quality control, HPTLC, densitometry, quantitative analysis 32a

103 096 M. GANDHIMATHI*, T. K. RAVI (*Dept. of Pharmaceutical Analysis, College of Pharmacy, Sri Ramakrishna Institute of Para Medical Sciences, Coimbatore 641044, India, gands72@yahoo.co.in): HPTLC method for the estimation of hydrochlorthiazide from its combined dosage forms. Indian Drugs 46(2), 150-153 (2009). HPTLC of hydrochlorthiazide (HZ) in combination with quinapril (QP) or candensartan (CD) on silica gel with toluene - ethyl acetate - glacial acetic acid 2:12:1 (for HZ and QP), or 20:50:1 (for HZ and CD), with chamber saturation for 30 min at room temperature. Quantitative determination by absorbance measurement at 273 nm. The method was linear in the range of 480-960 (HZ), 400-800 (QP) and 50-250 ng/spot (CD).

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

103 097 C. GIAGINIS, Anna TSANTILI-KAKOULIDOU* (*Department of Pharmaceutical Chemistry, School of Pharmacy, University of Athens, Panepistimiopolis, Zografou, Athens 157 71, Greece; tsantili@pharm.uoa.gr): RPTLC retention indices of basic and neutral drugs as surrogates of octanol-water distribution coefficients. Effect of buffer constituents and pH. J. Planar Chromatogr. 22, 217-224 (2009). TLC of 26 structurally diverse basic and neutral drugs on RP-18 with phosphate buffer, or phosphate-buffered saline, or morpholinepropansulfonic acid at pH 7.4, or phosphate buffer at pH 11.0, or pure water, and different proportions of methanol. The use of n-octanol as mobile phase additive was also investigated. Detection under UV light at 254 nm.

pharmaceutical research, qualitative identification 32a

103 098 Rina GOKANI*, Mamta SHAH (*L.M. College of Pharmacy, Dept. of Pharmacognosy, Navrangpura, Ahmedabad 380009, India): Isolation and estimation of clerodendrin A in Clerodendrum phlomidis and Ptemna integrifolia root. J. Pharm. Res. 8(1), 9-11 (2009). HPTLC of clerodendrin A in Clerodendrum phlomidis and Ptemna integrifolia root on silica gel with n-hexane - ethyl formate 7:3. Detection by spraying with a 5 % aqueous solution of H2SO4, followed by heating at 110 °C. Quantitative determination by absorbance measurement at 396 nm. Clerodendrum phlomidis roots contained more clerodendrin A (0.07 %) than roots of Ptemna integrifolia (0.04 %).

pharmaceutical research, herbal, HPTLC, densitometry, quantitative analysis 32c

103 099 C. GREEN*, S. HIBBERT, Y. SHAW, L. WILLIAMS, S. MITCHELL, E. GARRAWAY (*Natural Products Unit, Product Research and Development Division, Scientific Research Council, Hope Gardens, Kingston, Jamaica, cherylg@src-jamaica.org): Extraction, processing, and storage effects on curcuminoids and oleoresin yields from Curcuma longa L. grown in Jamaica. J. Agric. Food Chem. 56, 3664-3670 (2008). HPTLC of curcumin from the rhizomes of Curcuma longa on silica gel with chloroform - ethyl acetate 19:1. Detection by spraying with ammonium molybdate 5 % in aqueous sulphuric acid and examination under UV 254 nm. The hRf value of curcumin was 57.

herbal, HPTLC, qualitative identification 32e

103 100 A. HAWRYL, D. CICHOCKI, Monika WAKSMUNDSKA-HAJNOS* (*Department of Inorganic Chemistry, Faculty of Pharmacy, Medical University, 6 Staszica, 20-081 Lublin, Poland; monika.hajnos@am. lublin.pl): Determination of the lipophilicity of some psychotropic drugs by RP-TLC. J. Planar Chromatogr. 21, 343-348 (2008). HPTLC of 12 psychotropic drugs (chloropromazine, perazine, flupentixol, haloperidol, risperidon, alprazolam, midazolam, clomipramine, amitryptyline, doxepin, moclobemide, carbamazepine) on RP-18 in a horizontal chamber. The 10 mobile phases were prepared by mixing different amounts of water and the polar modifiers methanol, dioxane, acetone, acetonitrile, or tetrahydrofuran. Ammonia solution, acetate buffer (pH 4.75), or dodecyl sulfate was added to each of these mixtures. Detection under UV 254 nm.

pharmaceutical research, HPTLC, qualitative identification 32a

103 101 W. HUANG (Huang Wenhua)*, B. ZHAO (Zhao Bin), CH. XIE (Xie Chaoliang), J. LI (Li Jianli) (*Sichuan Inst. TCM, Chengdu, Sichuan 610031, China): (Study of the quality standard for Hongjinchan granules) (Chinese). J. Chinese Trad. Patent Med. 30 (12), 1997-1800 (2008). TLC of Hongjinchan granule extracts on silica gel with 1) n-butanol - glacial acetic acid - water 7:1:2; and 2) chloroform - ethyl acetate 7:3. Detection 1) under UV 365 nm; 2) by spraying with 2 % iron(III) chloride in ethanol; 3) by exposure to ammonia vapor for approx. 15 min. Identification by comparison with the standards of the component drugs. Quantification of scutellarin by HPLC.

quality control, traditional medicine, pharmaceutical research, quantitative analysis, qualitative identification 32e, 4d

103 102 T.W. INGLOT*, K. DABROWSKA, A. GUMIENICZEK (*Department of Medicinal Chemistry, Skubiszewski Medical University, Jaczewskiego 4, 20-090 Lublin, Poland; t.inglot@am.lublin.pl): The reversed-phase retention behavior of some angiotensin-II receptor antagonists. J. Planar Chromatogr. 22, 145-155 (2009). TLC of candesartan, eprosartan, telmisartan, losartan, and valsartan on RP-8 and RP-18 with mobile phases comprising 3:7 mixtures of phosphate buffer of different pH (2-8) and one of three modifiers, acetonitrile, methanol, or tetrahydrofuran, in horizontal chambers at ambient temperature. Detection under UV 254 nm. Quantitative determination by absorbance measurement. Separation of the five sartans was also achieved by TLC on RP-18 with dimethyl sulfoxide - phosphate buffer (pH 5.0) 4:1.

pharmaceutical research, densitometry, qualitative identification 32a

103 103 N. JAIN*, G.K. JAIN, F.J. AHMAD, R.K. KHAR (*Department of Pharmaceutics, Faculty of Pharmacy, Hamdard University, Hamdard Nagar, New Delhi 110062, India): Validated stability-indicating densitometric thin-layer chromatography: Application to stress degradation studies of minocycline. Anal. Chim. Acta 599 (2), 302-309 (2007). HPTLC of minocycline on silica gel (impregnated with a 10 % (w/v) solution of disodium ethylene diaminetetraacetic acid (EDTA) with a pH of 9.0) with methanol - acetonitrile - isopropyl alcohol - water 10:8:1:1. Quantitative determination by absorbance measurement at 345 nm. The limit of detection was 3.7 ng/spot, recovery was 99.2 - 100.2 %, and precision was good with a % RSD of 0.4 %. The method was able to separate all degradation products (produced by acidic and basic degradation, oxidation and photodegradation) from the pure drug.

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

103 104 A. JAMSHIDI*, A, SHARIFI (*Iran Polymer and Petrochemical Institute, Novel Drug Delivery Systems Department, P. O. Box 14185/458 Tehran, Iran; a.jamshidi@ippi.ac.ir): HPTLC analysis of tamoxifen citrate in drug-release media during development of an in-situ-cross-linking delivery system. J. Planar Chromatogr. 22, 187-189 (2009). HPTLC of tamoxifen citrate ((Z)-2-[4-(1,2-diphenylbut-1-enyl)phenoxy]ethyldimethylamine citrate) on silica gel (prewashed with methanol - chloroform 1:1) by automated multiple development (AMD 2) using single-step isocratic development with toluene - methanol - glacial acetic acid 57:38:5. Detection under UV 254 nm. Quantitative determination by absorbance measurement at 256 nm. The limit of detection and

quantitation was 25 and 52 ng/band, respectively.

pharmaceutical research, HPTLC, densitometry, quantitative analysis, AMD 32a

103 105 W. JIANG (Jiang Weike)*, T. ZHOU (Zhou Tao), P. GUO (Guo Peinan), Z. CHEN (Chen Zaifa) (*Guiyang Coll. TCM, Guiyang, Guizhou 550002, China): (Study of the quality standard for Yinao pills) (Chinese). J. Chinese Trad. Patent Med. 30 (11), 1642-1646 (2008). TLC of Yinao pill extracts on silica gel with 1) toluene - ethyl acetate 9:1; 2) benzene - ethyl acetate 3:2; 3) toluene - ethyl acetate - formic acid 28:8:1. Detection 1) by spraying with 10 % phosphomolybdic acid in ethanol and heating at 105 ºC; 2) by spraying with 10 % sulfuric acid in ethanol and heating at 105 ºC.

quality control, traditional medicine, pharmaceutical research, quantitative analysis, qualitative identification, Chinese traditional medicine, 32e

103 106 R. KAKDE*, N. BAWANE (*Department of Pharmaceutical Sciences, RTM Nagpur University, Nagpur 440 033, Maharashtra, India; drkakde@yahoo.com; nilesh.bawane2000@gmail.com): High-performance thin-layer chromatographic method for simultaneous analysis of metoprolol succinate and amlodipine besylate in pharmaceutical preparations. J. Planar Chromatogr. 22, 115-119 (2009). HPTLC of metoprolol succinate and amlodipine besylate on silica gel (prewashed with methanol) with methanol - ethyl acetate - water - toluene - 25 % ammonia 15:50:3:30:3 in a twin trough chamber saturated for 20 min at 25 +/- 2 °C. Quantitative determination by absorbance measurement at 236 nm.

quality control, HPTLC, densitometry, quantitative analysis 32a

103 107 B. KALYANI, K.S. LADDHA* (*Medicinal Natural Product Laboratory, Pharmaceutical Division, University Institute of Chemical Technology, Matunga, Mumbai 400 019, India; ksladdha@udct.org, ksladdha@yahoo.co.in): Discriminating features of safed musli and shatavari. J. Planar Chromatogr. 22, 157-161 (2009). Safed musli is the common name for Chlorophytum borivilianum, whereas shatavari is Asparagus racemosus, both of the liliaceae family. HPTLC of saponins and sapogenins on silica gel with chloroform - methanol - water 32:25:5 for saponins and with petroleum ether - ethyl actate 4:1 for sapogenins. Detection of saponins by spraying with anisaldehyde reagent and with Ehrlich‘s reagent, followed by densitometric analysis at 620 and 510 nm. Detection of sapogenins by spraying with Liebermann Burchard reagent, followed by densitometric analysis at 580 nm.

herbal, quality control, densitometry, qualitative identification, HPTLC, quantitative analysis 32e

103 108 S. KHATOON*, N. SINGH, N. SRIVASTAVA, A. K. S. RAWAT, S. MEHROTRA (*Pharmacognosy and Ethnopharmacology Division, National Botanical Research Insitute, Rana Pratp Marg, Lucknow 226001, India, sayyadak@yahoo.com, sayyadak@nbri.res.in): Chemical evaluation of seven Terminalia species and quantification of important polyphenols by TLC. J. Planar Chromatogr. 21, 167-171 (2008). HPTLC of gallic acid and ellagic acid on silica gel with toluene - ethyl acetate - formic acid 5:5:1 and 40:25:4 in a twin trough chamber saturated for 30 min. Detection under UV light at 254 and 366 nm and under visible light after derivatization with anisaldehyde reagent. Quantitative determination by absorbance measurement at 272 nm.

103 109 Katarzyna KULIG*, B. MALAWSKA (*Department of Physicochemical Drug Analysis, Faculty of Pharmacy, Jagiellonian University Medical College, Medyczna 9, 30-688 Kraków, Poland; mfkkulig@cyf-kr.edu.pl): RP-TLC determination of the lipohilicity of 1-substituted pyrrolidin-2-one derivatives. Correlation of lipophilicity with affinity for alpha-adrenoceptors. J. Planar Chromatogr. 22, 141-144 (2009). Determination of the relative lipophilicity of 14 1-substituted pyrrolidin-2-one by TLC on RP-18 with mixtures of acetonitrile and pH 7.0 Tris buffer with volume fractions of acetonitrile between 20 and 80 %, with chamber saturation for 2 h. Detection under UV light. Retention data obtained by this method were exponentially dependent on aceto-

nitrile concentration and enabled estimation of the relative lipophilicity corresponding to pH 7.0 Tris buffer as mobile phase.

pharmaceutical research, qualitative identification, physicochemical organic chemistry 32a

103 110 V. KUMAR, K. MUKHERJEE, S. KUMAR, M. MAL, P.K. MUKHERJEE* (*School of Natural Product Studies, Department of Pharmaceutical Technology, Jadavpur University, Kolkata 700032, India; Pknatprod@yahoo.co.in): Validation of HPTLC method for the analysis of taraxerol in Clitora ternatea. Phytochem. Anal. 19, 244-250 (2008). HPTLC of taraxerol on silica gel in a saturated twin trough chamber with hexane - ethyl acetate 4:1. Detection by spraying with anisaldehyde reagent. Quantitative determination by absorbance measurement at 420 nm. Limits of detection and quantification were 31 and 105 ng/spot, respectively.

herbal, HPTLC 32e

103 111 N.S. KUMAR*, M. VENKATESH, S. PONNUSANKAR, P. VENKATESH, A. GANTAIT, N. NEMA, S. BHADRA, D. MUKHERJEE, S. PANDIT, P. MUKHERJEE (*School of Natural Product Studies, Dept. of Pharmaceutical Tech., Jadavpur University, Kolkata, India): Mahanimbine in Murraya koenigi - Marker analysis and acetyl cholinesterase enzyme inhibition. Abstract No. 0983, IHCB (2009). HPTLC of mahanimbine (a carbazole alkaloid isolated from Murraya koenigi) on silica gel with petroleum ether - chloroform 13:7. The hRf value of mahanimbine was 60. Quantitative determination by absorbance measurement at 254 nm. The method was linear in the concentration range of 50-250 ng/spot. Enzyme inhibition activity of methanol, petroleum ether, and chloroform exctracts of the plant and of mahanimbine was evaluated using galantamine as standard inhibitor of the enzyme.

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

103 112 S. KUMAR*, R. MADAAN, A. SHARMA (*S. D. College of Pharmacy, Barnala 148101, India).: Estimation of apigenin, an anxiolytic constituent, in Turnera aphrodisiaca. Ind. J. of Pharma Sci. 70(6), 847-851 (2008). HPTLC of apigenin in segregated parts (leaves, stems, flowers and fruits) of Turnera aphrodisiaca on silica gel with toluene - ethyl acetate 1:4. Quantitative determination by absorbance measurement at 336 nm. Flowers were found to contain maximum amount of apigenin whereas leaves contained the least. Apigenin contents in methanolic extracts of aerial plant parts were fourteen times lower than in acid hydrolyzed methanolic extracts, indicating the presence of most of apigenin in glycosidic form. The plant material collected in September showed maximum contents of apigenin.

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

103 113 K.J. KUMAR*, S. JAYARAMAN, S. NARASIMHAN (*Department of Pharmaceutical Sciences, Birla Institute of Technology, Mesra, Ranchi 835215, India; jayaram_res@yahoo.com): A simple and rapid method of estimation of nimbolide, an anticancer constituent in neem leaves. J. Planar Chromatogr. 21, 263-265 (2008). HPTLC of nimbolide on silica gel in a twin trough chamber with ethyl acetate - hexane 1:1. Quantitative determination by absorbance measurement at 254 nm. The limits of detection and quantification were 3.3 and 1.0 µg/spot, respectively.

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

103 114 P.J. LOKHANDE, J. K. VERMA* (*Inorganic Chemistry Laboratory, K. J. Somaiya College of Science and Commerce, Vidyavihar, Mumbai 400 077, Maharashtra, India; jkverma7@rediffmail.com): Quantification of negundoside in Vitex negundo Linn. leaf by high-performance thin-layer chromatography. J. Planar Chromatogr. 22, 225-228 (2009). HPTLC of negundoside on silica gel (prewashed with methanol) with ethyl acetate - methanol - water - glacial acetic acid 78:12:7:3 in a twin trough chamber saturated for 20 min. Quantitative determination by absorbance measurement at 267 nm.

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

103 115 V. MADHAVAN*, Hema BASNETT, A. CENDIL KUMAR, S. YOGANARASIMHAN (*M.S. Ramaiah College of Pharmacy, Dept. of Pharmacognosy and Pharmaceutical Chemistry, Bangalore 60054, India): Fingerprinting of plumbagin in Drosera burmannii Vahl using high performance thin layer chromatography. Ind. J. Pharma Sci. 70(6), 798-800 (2008). HPTLC of plumbagin in Drosera burmannii Vahl on silica gel with toluene - glacial acetic acid 55:1 (for alcoholic extracts) and toluene - chloroform - glacial acetic acid 10:10:1 (for aqueous extracts) with chamber saturation for 60 min. Evaluation in visible light at 425 nm. The alcoholic extract showed seven components, the main zone with hRf value of 56 corresponded to plumbagin. The aqueous extract showed two zones but no plumbagin.

pharmaceutical research, quality control, herbal, HPTLC 32c

103 116 Manasi MANTRI*, Nancy PANDITA (*SVKM's, NMIMS University, Mumbai, India): Authentication of Hoodia and hang off capsules by TLC photo documentation. Abstracts No. 9156, IHCB (2009). TLC of Hoodia gordonii on silica gel with ethyl acetate - methanol - water 36:7:5. Photodocumentation and estimation was carried out by the DISTA method. The authentication of hang-off capsules, which contained several plant ingredients used to prevent hang-over, was performed by TLC on silica gel with toluene - ethyl acetate 9:1. Detection by spraying with vanillin-sulfuric acid reagent.

pharmaceutical research, quality control, herbal 32e

103 117 G. MATYSIK, Agnieszka SKALSKA-KAMINSKA*, B. STEFANCZYK, M. WÓJCIAK-KOSIOR, D. RAPA (*Department of Chemistry, Laboratory of Planar Chromatography, Medical University, Staszica 6, 20-081 Lublin, Poland; agnieszkask@wp.pl): Application of a new technique in two-dimensional TLC separation of multicomponent mixtures. J. Planar Chromatogr. 21, 233-236 (2008). Connection of two-dimensional chromatography with multiple gradient development. 2D HPTLC of anthraquinone derivatives (1,8-dihydroxyanthraquinone, franguloemodin A, aloeemodin, rhein, frangulin A, aloin, sennoside B) on silica gel. Bandwise application at the right and left corner of the plate, then development by use of multiple gradient development [G. Matysik, Chromatographia 43, 39-41 (2004)], e.g. for step 1 hexane - dichloromethane 1:1, for step 2 (hexane - dichloromethane - ethyl acetate - 80 % formic acid 50:40:10:1) and for step 3 hexane - dichloromethane - ethyl acetate - methanol - formic acid 40:40:20:5:1. After drying the plate was developed from the left and right edge with hexane - dichloromethane - formic acid 60:40:1 for step 1 and with hexane - dichloromethane - ethyl acetate - formic acid 60:35:5:1 for step 2 . Quantitative determination by absorbance measurement at 440 nm. Deteciton of anthranoids in daylight and after derivatization with 10 % potassium hydroxide in methanol.

herbal, qualitative identification, HPTLC, densitometry 32e

103 118 S. MENNICKENT*, R. FIERRO, M. VEGA, M, DIEGO, G. GODOY (*Department of Pharmacy, Faculty of Pharmacy, University of Concepcion, Concepcion, Chile, smennick@udec.cl): Instrumental planar chromatographic method for determination of carbamazepine in human serum. J. Sep. Sci. 32, 1454-1458 (2009). HPTLC of carbamazepine in human serum on silica with ethyl acetate - toluene - methanol - glacial acetic acid 10:8:1:1. Detection by dipping in a solution of perchloric acid 60 % - ethanol - water 1:1:1, followed by heating at 120-150 °C for 5-10 min. Quantitative determination by fluorescence measurement at 366/>400 nm. The accuracy and precision did not exceed 3.2 % RSD at any level. The hRf value of carbamazepine was 55 and selectivity regarding matrix was given. Linearity was between 3 and 20 ng/µL. The intra-assay and inter-assay precision, expressed as the RSD, were in the range of 0.4 - 1.2 % (n = 3) and 2.2 - 3.2 % (n = 9), respectively. The limit of detection and quantification was 0.19 and 0.57 ng, respectively. Recovery (by standard addition) was between 99.0 and 102.0 %.

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

103 119 D.B. MESHRAM*, S.B. BAGADE, M.R. TAJNE (*University Department of Pharmaceutical Sciences, RTM Nagpur University, Nagpur 440033, Maharashtra, India; dbmeshram@yahoo.com): TLC-densitometric analysis of clotrimazole and metronidazole in combined dosage forms. J. Planar Chromatogr. 21, 277-282 (2008). TLC of clotrimazole and metronidazole on silica gel in a twin trough chamber saturated for 10 min with toluene - ethyl acetate - methanol - acetic acid 55:2:6:1. Quantitative determination by absorbance measurement at 220 nm.

quality control, densitometry, quantitative analysis 32a

103 121 Jadwiga MIELCAREK*, P. GROBELNY, T. OSMALEK (*Department of Inorganic and Analytical Chemistry, Poznan Univerity of Medical Sciences, Grunwaldzka 6, 60-780 Poznan, Poland; jmielcar@amp.poznan.pl): Identification of photoproducts of fluvastatin in solutions. J. Planar Chromatogr. 22, 137-140 (2009). HPTLC of fluvastatin and photochemical decomposition products on RP-18 with phosphate buffer (pH 4.0) - methanol 3:17 at 4 +/- 0.5 °C with chamber saturation and under light protection. Before development the plates were thermostated for 10 min in the dry chamber. Detection of fluvastatin and its photoproducts under UV 254 nm. Isolation of photoproducts by preparative TLC on RP-18.

quality control, HPTLC, qualitative identification, preparative TLC 32a

103 122 M.A.A. MOHAMMAD*, N.H. ZAWILLA (*Department of Pharmaceutical and Medicinal Chemistry, Faculty of Pharmacy, Cairo University, Kasr El-Aini 11562, Cairo, Egypt; mohammadazim97@yahoo.com): Thin-layer and column-chromatographic methods for simultaneous analysis of ambroxol hydrochloride and doxycycline hyclate in a binary mixture. J. Planar Chromatogr. 22, 201-206 (2009). TLC of ambroxol hydrochloride and doxycycline hyclate on silica gel with ethyl acetate - ethanol - glacial acetic acid - water 90:40:5:10 with chamber saturation for 1 h. Detection under UV 254 nm. Quantitative determination by absorbance measurement at 254 nm for ambroxol hydrochloride and 270 nm for doxycycline hyclate.

quality control, densitometry, quantitative analysis 32a

103 123 M. MORENO*, A. VILORIA, D. HIDALGO (*Simon Rodriguez University, Valencia, Venezuela, morenoalvarez@cantv.net): A new method for the isolation of betalaines by HPTLC. Rev. Fac. Agron. 21, 155-160 (2004). HPTLC of betaxantin and betacyanin in the roots of Beta vulgaris on cellulose in two one-dimensional developments with 1) isopropanol - ethanol - water - acetic acid 6:7:6:1 and 2) isopropanol - ethanol - water - acetic acid 11:4:4:1. Qualitative identification under UV light. Betaxantin and betacyanin showed maximum absorbances at 537 and 465 nm, respectively. The hRf values of betaxantin and betacyanin were 22 and 34, respectively. herbal,

HPTLC, qualitative identification 32e

103 124 O. MORINAGA, T. UTO, S. SAKAMOTO, W. PUTALUN, S. LHIEOCHAIPHANT, H. TANAKA, Y, SHOYAMA* (*Faculty of Pharmaceutical Science, Nagasaki International University, 2825-7 Huis Ten Bosch, Sasebo, Nagasaki 859-3298, Japan; shoyama@niu.ac.jp): Development of eastern blotting technique for sennoside A and sennoside B using anti-sennoside A and anti-sennoside B monoclonal antibodies. Phytochem. Anal. 20, 154-158 (2009). TLC of the purgative constituents of rhubarb (sennoside A and sennoside B) on silica gel with 1-propanol - ethyl acetate - water - acetic acid 40:40:30:1. After drying detection by spraying with 10 % sulfuric acid and heating. For the eastern blotting assay the developed TLC plate was dried and sprayed with isopropanol - methanol - water 1:4:16. Transfer by using a PVDF membrane sheet for further treatment.

herbal, traditional medicine, quality control, qualitative identification 32e, 3e

103 125 S.K. MOTWANI *, R.K. KHAR, F.J. AHMAD, S. CHOPRA, K. KOHLI, S. TALEGAONKAR, Z. IQBAL (*Department of Pharmaceutics, Faculty of Pharmacy, Jamia Hamdard, Hamdard Nagar, New Delhi 110062, India): Stability indicating high-performance thin-layer chromatographic determination of gatifloxacin as bulk drug and from polymeric nanoparticles. Anal. Chim. Acta, 576 (2), 253-260 (2006). Description of a simple, sensitive, selective, precise and stability

indicating method for determination of gatifloxacin both as a bulk drug and from polymeric nanoparticles by TLC on silica gel with n-propanol - methanol - 25 % ammonia 50:10:9. The hRf value of gatifloxacin was 60. Separation from degradation products (produced under acidic and basic conditions and upon wet and dry heat treatment) was good. Quantitative determination by absorbance measurement at 292 nm. Linearity was 400 - 1200 ng/spot, with a correlation coefficient of 0.9953. The limit of detection and quantitation was 3 and 8 ng/spot, respectively.

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

103 126 S. MURALIDHARAN*, S.N. MEYYANATHAN, N. MURUGANANTHAM, S. RAJAN, K. KRISHNARAJ, B. SURESH (*Department of Pharmaceutical Analysis, JSS College of Pharmacy, Rocklands, Ooty, Ootacamund 643 001, India; murali23pharm@rediffmail.com): Validated HPTLC method of analysis of dexibuprofen in its formulation. J. Planar Chromatogr. 22, 207-210 (2009). HPTLC of dexibuprofen on silica gel (prewashed with methanol) in a twin trough chamber with hexane - ethyl acetate - glacial acetic acid 15:5:1. Quantitative determination by absorbance measurement at 217 nm.

quality control, HPTLC, densitometry, quantitative analysis 32a

103 127 P.S. NAYAK*, S.D. UPADHYAYA, A. UPADHYAYA (*Department of Crop and Herbal Physiology, College of Agriculture, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur M. P. 482004 India; preetisagarnayak@rediffmail.com): HPTLC method for analysis of whithaferin-A in Ashwagandha (Withania somnifera). J. Planar Chromatogr. 22, 197-200 (2009). HPTLC of whithaferin A on silica gel with toluene - ethyl acetate - formic acid 5:5:1 in a twin trough chamber saturated for 20 min at 25 +/- 2 °C and 50 +/- 5 % relative humidity. Detection under UV light and by dipping into freshly prepared p-anisaldehyde reagent, followed by heating at 110 °C for 10 min. Quantitative determination by absorbance measurement at 200 nm. The limit of detection and quantification was 100 and 800 ng/zone, respectively. The high sample throughput is useful for routine analysis of the preparation in industrial quality control and regulatory laboratories

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

103 128 H.J. PANCHAL*, B.N. SUHAGIA, N.J. PATEL, B.H. PATEL (*S.S.K. Patel College of Pharmaceutical Education and Research, Ganpat Vidyanagar, Kherva, Mehsana-382711, Gujarat, India; hir_143_2003@yahoo. com): A simple and sensitive HPTLC method for quantitative analysis of pitavastatin calcium in tablets. J. Planar Chromatogr. 21, 267-270 (2008). HPTLC of pitavastatin calcium on silica gel (prewashed with methanol) in a twin trough chamber saturated for 30 min at room temperature with toluene - methanol - glacial acetic acid 190:59:1. Quantitative determination by absorbance measurement at 238 nm. The limits of detection and quantification were 7 and 20 ng/band, respectively.

quality control, HPTLC, quantitative analysis, densitometry 32a

103 129 S. PANDIT*, S. PONNUSANKAR, M. VENKATESH, A. GANTAIT, A. BANDYOPADHYAY, P. MUKHERJEE (*School of Natural Product Studies, Dept. of Pharmaceutical Tech., Jadavpur University, Kolkata, India): Standardization, validation and safety evaluation of Emblica officinalis Linn. Abstract No. 9148, IHCB (2009). HPTLC of gallic acid in Emblica officinalis (an important constituent of Triphala, a popular ayurvedic formulation) on silica gel with toluene - ethyl acetate - formic acid 10:10:3. The hRf value of gallic acid was 45. Quantitative determination by absorbance measurement at 254 nm. The method was linear in the range of 150-530 ng/spot. The extract was found to contain 7.5 mg/g of gallic acid.

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

103 130 M. PARAMASIVAM*, R. POI, H. BANERJEE, A. BANDYOPADHYAY (*Department of Agricultural Chemicals, Faculty of Agriculture, Bidhan Chandra Krishi Viswavidyalaya, Mohanpur,

Nadia 741252, India, sivam25@gmail.com): High-performance thin layer chromatographic method for quantitative determination of curcuminoids in Curcuma longa germplasm. Food Chem. 113, 640-644 (2009). HPTLC of curcumin (1), demethoxycurcumin (2), and bisdemethoxycurcumin (3) from the rhizomes of Curcuma longa on silica gel with chloroform - methanol 24:1. Quantitative determination by absorbance measurement at 425 nm. The hRf values of (1), (2), and (3) were 66, 48, and 30, respectively. Selectivity regarding matrix was given. Linearity was between 1 and 20 µg/spot for all curcuminoids. The intermediate precision of the method was satisfactory. Recovery was 98.7 % for (1), 96.3 % for (2), and 97.2 % for (3). The limit of detection for the substances was 100 ng/spot.

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

103 131 S.K. PATEL*, N. PATEL, P.U. PATEL, D.B. PATEL, A.M. PRAJAPATI, S.A. PATEL (*College of Pharmaceutical Education and Research, Ganpat University, Kherva, Dist-Mehsana, Gujarat, India 382 711; skpatel_2@rediffmail.com): Validation of a stability-indicating HPTLC method for analysis of duloxetine hydrochloride in capsule dosage form. Separation and analysis of duloxetine hydrochloride and olanzapine in a synthetic mixture. J. Planar Chromatogr. 22, 121-126 (2009). HPTLC of duloxetine hydrochloride (and degradation products after acidic and basic hydrolysis, oxidation and photodegradation) on silica gel with toluene - methanol - 10 % ammonia 60:26:1 in a twin trough chamber saturated at 25 °C. Quantitative determination by absorbance measurement at 231 nm. The hRf value was 39. Linearity was between 60-480 ng/band. The limit of detection and quantification was 20 and 60 ng/spot, respectively. For separation of duloxetine hydrochloride and olanzapine in a synthetic mixture HPTLC with acetone - methanol - triethylamine 10:6:1. Quantitative determination by absorbance measurement at 240 nm. The hRf values were 63 and 77, respectively. Linearity was between 100-800 ng/band.

quality control, HPTLC, densitometry, quantitative analysis 32a

103 132 M.B. PATEL*, V.M. KADAKIA, S.H. MISHRA (*Pharmacy Dept., Kalabhavan Faculty of Technology and Engineering, The M. S. University of Baroda, Vadodara 390001, India, shmishra48@rediffmail.com): Simultaneous estimation of andrographolide and wedelolactone in herbal formulations. Ind. J. Pharm. Sci. 70(5), 689-693 (2008). HPTLC of andrographolide and wedelolactone (active components of Andrographis paniculata respectively Eclipta alba) on silica gel with toluene - acetone - formic acid 9:6:1. Quantitative determination by absorbance measurement at 254 nm. The hRf of andrographolide was 52 and of wedelolactone 58. The method was linear in the range of 200-400 ng/spot (andrographolide) and 100-200 ng/spot (wedelolactone). The herbal formulation contained 1.4 mg andrographolide and 1.7 mg wedelolactone per tablet. The proposed method could be applied for routine analysis of both compounds.

herbal, HPTLC, densitometry, quantitative analysis 32e

103 133 Anna PELANDER, DANIEL BACKSTRÖM, I. OJANPERÄ* (*Department of Forensic Medicine, P.O. Box 40, University of Helsinki, 00014 Helsinki, Finland): Qualitative screening for basic drugs in autopsy liver samples by dual-plate overpressured layer chromatography. J. Chromatogr. B 857 (2), 337-340 (2007). OPLC of basic drugs in autopsy liver samples prepared by enzymatic digestion with trypsin and liquid - liquid extraction with butyl chloride. Separation by dual-plate analysis with trichloroethylene - methyl ethyl ketone - n-butanol - acetic acid - water 17:8:25:6:4 for OPLC 1 and butyl acetate - ethanol (96.1 %) - tripropylamine - water 340:37:20:3 for OPLC 2. Identification by automated comparison of corrected hRf values and in situ UV spectra with library values by dedicated software. Determination of the identification limit for 25 basic drugs in liver ranging from 0.5 to 10 mg/kg. The method is suitable for routine screening of basic drugs in post-mortem samples of varying quality, combining the benefit from moderately high separation power with the ease of disposable plates.

OPLC 32f

103 134 S. PONNUSANKAR*, S. PANDIT, M. VENKATESH, A. BANDOPADHYAY, P. MUKHERJEE (*School of Natural Studies, Jadavpur University, Kolkata 700032, India, and Molecular Endocri-

nology Lab, Indian Institute of Chemical Biology, Kolkata, India): Standardization, validation and safety evaluation of Terminalia chebula Retz. Abstract No. 9145, IHCB (2009). Terminalia chebula (an important constituent of Triphala, a popular ayurvedic formulation) was standardized for gallic acid content by HPTLC on silica gel with toluene - ethyl acetate - formic acid 10:10:3. The hR_f value of gallic acid was 45. Quantitative determination by absorbance measurement at 254 nm. The method was linear in the range of 150-550 ng/spot. Hydroalcoholic extracts of the fruit pulp contained 10.5 mg/g gallic acid.

pharmaceutical research, quality control, herbal, HPTLC, densitometry 32e

103 135 O. POTTERAT*, R. FELTEN, P. DALSGAARD, M. HAMBURGER (*Institute of Pharmaceutical Biology, University of Basel, Klingelbergstrasse 50, Basel, Switzerland, olivier.potterat@unibas.ch): Identification of TLC markers and quantification by HPLC-MS of various constituents in noni fruit powder and commercial noni-derived products. J. Agric. Food Chem. 55, 7489-7494 (2007). HPTLC of fruit powder and commercial products of Noni (Morinda citrifolia) on silica gel with chloroform - methanol 9:1 or chloroform - methanol - water 13:6:1. Detection under UV 254 and 366 nm after spraying with vanillin reagent (1 % vanillin in ethanol containing 2 % sulphuric acid), followed by heating at 105 °C. The following compounds were identified as markers (hR_f): ursolic acid (60), linoleic acid (55), scopoletin (53), 3-methyl-1,3-butanediol (27).

food analysis, herbal, HPTLC, qualitative identification 32e

103 136 K. RAVIKANTH*, M. THAKUR, B. SINGH, M. SAXENA (*Research and Development Centre, Ayurvet Ltd., Katha, P.O. Baddi, Solan, HP, India): TLC based method for standardization of Pongamia pinnata (Karanj) using karanjin as marker. Chromatographia 69 (5-6), 597-599 (2009). TLC of karanjin in the seeds of Pongamia pinnata on silica gel with toluene - ethyl acetate 7:3. Quantitative determination by absorbance measurement at 260 nm. The limit of detection was 100 ng, linearity was 50 - 300 ng. Four samples of P. pinnata from different geographical locations were screened for their karanjin content.

pharmaceutical research, quality control, traditional medicine, herbal, quantitative analysis, qualitative identification, densitometry 32e

103 137 E. REICH*, Valeria WIDMER (*CAMAG Laboratory, Sonnenmattstr. 11, 4132 Muttenz, Switzerland, Lab@camag.com): HPTLC for rapid identification of Black Cohosh. LC-GC Europe, The Applications 19 (2006). HPTLC of actein, chlorogenic acid, caffeic acid, and isoferulic acid on silica gel with toluene - ethyl formate - formic acid 5:3:2 with chamber saturation for 20 min and at a relative humidity of 5 % (automatic development chamber ADC2 with molecular sieve). Detection by dipping in sulfuric acid reagent (20 mL of sulfuric acid in 80 mL of methanol) followed by heating at 100 °C for 5 min. Evaluation under daylight and under UV 254 and 366 nm.

herbal, HPTLC, qualitative identification, postchromatographic derivatization 32e

103 138 K.K. ROUT*, S.K. MISHRA (*Pharmacognosy and Phytochemistry Division, University Department of Pharmaceutical Sciences, Utkal University, Vani Vihar, Bhubaneswar 751004, Orissa, India; kd_rout@yahoo.co.in): Efficient and sensitive method for quantitative analysis of 6-gingerol in marketed Ayurvedic formulation. J. Planar Chromatogr. 22, 127-131 (2009). HPTLC of 6-gingerol, extracts of Suprabha and market samples of ginger on silica gel (prewashed with methanol) with n-hexane - acetone 18:7 in a twin trough chamber saturated for 4 min at 20 +/- 4 °C and 36 +/- 3 % relative humidity. Quantitative determination by absorbance measurement at 286 nm. The limit of detection and quantification was 40 and 150 ng/band, respectively.

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

103 139 A. SAHU*, A. GANTAIT, P. VENKATESH, P. MUKHERJEE (*School of Natural Product Studies, Dept. of Pharmaceutical Technology, Jadavpur University, Kolkata 700032, India): A vali-

dated method for standardization of Coccinia grandis extract. Abstract No. 9154, IHCB (2009). Standardization of Coccinia grandis extract by HPTLC of taraxerol on silica gel with n-hexane - ethyl acetate 9:1 in a twin trough chamber. Detection by spraying with anisaldehyde reagent, followed by heating at 105 °C. Quantitative determination by absorbance measurement at 420 nm. The dichloromethane extract of the plant contained 3.7 % (w/w) of taraxerol.

pharmaceutical research, herbal, HPTLC,densitometry, quantitative analysis 32e

103 140 M.O. SALAZAR, R.L.E. FURLAN* (*Cátedra de Farmacognosia, Facultad de Ciencias Bioquímicas y Farmacéuticas, Universidad Nacional de Rosario, Suipacha 531, S2002LRK Rosario, Argentina; rfurlan@fbioyf.unr.edu.ar): A rapid TLC autographic method for the detection of glucosidase inhibitors. Phytochem. Anal. 18, 209-212 (2007). Separation of extracts of Solanum diflorum and Setaria parviflora by TLC on silica gel, cellulose, and RP-18 and by HPTLC on cyano and diol phase with hexane - ethyl acetate 1:1 and n-butanol - formic acid - water 5:1:4 (upper phase). Detection after distribution of ß-glucuronidase staining solution (52.5 mg agar and 0.9 mL 0.5 % iron(III) chloride solution). After solidification of the staining solution, the TLC plate was incubated for 120 min at 37 °C and immersed in a 0.2 % solution of esculin. Autography showed enzyme inhibition zones with hRf of 14 (Solanum diflorum) and 46 (Setaria parviflora).

food analysis, qualitative identification, HPTLC, TLC autography 32e

103 141 P.U. SANGANALMATH, K.M. SUJATHA, S.M. BHARGAVI, V.G. NAYAK, B.M. MOHAN* (*Toxicology Division, Forensic Science Laboratory, Madivala, Bangalore 560068, Karnataka State, India; mohandfsl@yahoo.co.in): Simple, accurate and rapid HPTLC method for analysis of theophylline in post-mortem blood and validation of the method. J. Planar Chromatogr. 22, 29-33 (2009). HPTLC of theophylline (extracted at pH 8.5 with chloroform - isopropanol 4:1 from post-mortem blood after acid hydrolysis) on silica gel in a twin trough chamber with chloroform - methanol 9:1. Quantitative determination by absorbance measurement at 277 nm. Polynomial regression in the range of 0.5-20 µg/mL. The limit of detection was 0.5µ g/mL (S/N = 3). Intra-day and inter-day repeatability was between 0.5-0.8 % and 0.5-1.3 %, respectively, for three different concentrations in the range of 0.5-10 µg/mL. Recovery was 89.1-93.4 % at a concentration of 10 µg/mL and pH 8.3-8.6. An average analytical recovery of 89.9 % was achieved at pH 8.5 with a relative standard deviation of 2.2 %. Theophylline was stable in methanolic solution and during chromatography.

toxicology, densitometry, HPTLC, quantitative analysis 32d

103 142 R.S. SANGWAN*, N.S. SANGWAN, P.K. SHARMA, N.D. CHAURASIYA, S.K. MISHRA, B.R. TYAGI, A.K. SRIVASTAVA (*Central Institute of Medicinal and Aromatic Plants (CIMAP), PO CIMAP, Kukrail Picnis Spot Road, Lucknow 226015, India; sangwan.lab@gmail.com): Carbonate extraction process for the metabolic, isozymic and proteomic profiling of rose-scented geranium (Pelargonium sp.), a hyper-acidic plant. Phytochem. Anal.19, 104-115 (2008). TLC of geraniol on silica gel with chloroform - methanol - water 97:24:2 and of geranylacetate with toluene - ethyl acetate 93:7. Detection by spraying with vanillin sulfuric acid reagent.

herbal, qualitative identification 32e

103 143 A. SARASWATHY*, D.S. NANDINI, D. RAMASAMY (*Captain Srinivasa Murti Drug Research Institute for Ayurveda (CCRAS), Anna Hospital Campus, Arumbakkam, Chennai 600106, India, saraswathy20042000@yahoo.co.in): Chemical analysis of Terminalia alata and Terminalia arjuna stem bark. Indian Drugs 46(2), 109-112 (2009). HPTLC of chloroform extracts of stem bark of Terminalia alata and Terminalia arjuna on silica gel with chloroform - methanol 9:1. Arjunolic acid and maslinic acid were used as marker compounds. Detection by treatment with vanillin-sulphuric acid reagent. Quantitative determination by absorbance measurement at 254 nm. The fingerprint profile along with other physico-chemical data helped in the correct authentification and standardization of both plant species.

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

103 144 S.B. SEGAN, D.M. OPSENICA, B.A. SOLAJA, Dusanka M. MILOJKOVIC-OPSENICA* (*Institute of Chemistry, Technology and Metallurgy, Njegoseva 12, 11000 Belgrade, Serbia; dusankam@chem.bg.ac.yu): Planar chromatography of cholic acid-derived cis-trans isomeric bis-steroidal tetraoxanes. J. Planar Chromatogr. 22, 175-181 (2009). TLC of 14 tetraoxanes on silica gel with ethyl acetate - toluene and ethyl acetate - petroleum ether, on cyano phase with methanol - water and acetone - water, and on RP-18 with water - organic modifier (methanol, acetone, or dioxane) in various combinations, with chamber saturation for 15 min at ambient temperature (22 +/- 2 °C). Detection by spraying with 50 % sulfuric acid, followed by heating until spots became visible.

pharmaceutical research, qualitative identification 32a

103 145 S.A. SHAH*, I.S. RATHOD, B.N. SUHAGIA, D.A. PATEL, V.K. PARMAR, B.K. SHAH, V.M. VAISHNAVI (*Department of Quality Assurance, L. M. College of Pharmacy, Ahmedabad 380009, India): Estimation of boswellic acids from market formulations of Boswellia serrata extract and 11-keto-beta-boswellic acid in human plasma by high-performance thin-layer chromatography. J. Chromatogr. B 848 (2), 232-238 (2007). HPTLC of boswellic acids in Boswellia serrata extract and 11-keto-beta-boswellic acid in human plasma on silica gel with hexane - chloroform - methanol 10:10:1. Quantitative determination by absorbance measurement at 260 nm. The linearity range for 11-keto-beta-boswellic acid spiked in 1 mL of plasma was 29 - 146 ng/spot, with a recovery of 91.7 %. The limit of detection and quantification for 11-keto-beta-boswellic acid in human plasma was 9 and 29 ng/mL, respectively. The method was applied for the determination of 11-keto-beta-boswellic acid plasma levels in a clinical pilot study.

quality control, pharmaceutical research, traditional medicine, herbal, HPTLC, quantitative analysis, qualitative identification,densitometry 32e

103 146 C. SHARMA*, B. SUHAGIA, N. SHAH, R. SHAH (*Shri B. M. Shah College of Pharmaceutical Education and Research, Modasa 383315, Gujarat India): Development and validation of a HPTLC method for the estimation of sumatriptan in tablet dosage forms. Indian J. Pharma. Sci. 70(6), 831-834 (2008). HPTLC of sumatriptan on silica gel (pre-washed) with methanol - water - glacial acetic acid 40:80:1 with chamber saturation for 10 min. Quantitative determination by absorbance measurement at 230 nm. The hRf value was 64. The method was linear in the range of 200-800 ng/spot.

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

103 147 A. SHIKOV*, D. DEMCHENKO, Svetlana IVANOVA, Marina KARLINA, Vera KOSMAN, Olga POZHARITSKAYA, Irina URAKOVA (*Saint Petersburg Institute of Pharmacy, 47/5, Piskarevsky prospect, 195067, St-Petersburg, Russia, spb.pharmacy@gmail.com): HPTLC determination of ginkgolides A, B, and C and bilobalide in Ginkgo biloba. CBS 102, 10-12 (2009). HPTLC of diterpene lactones (ginkgolides A, B, C, and bilobalide) in aqueous Ginkgo biloba leaf extracts on silica gel (impregnated by dipping into a 4 % solution of sodium acetate in methanol - water 3:2 for 5 s followed by drying at room temperature for 1 h) with toluene - acetone 7:3 with chamber saturation for 20 min. After twofold development over 60 mm the plate is heated at 150 °C for 1 h for detection of active compounds. Quantitative determination by absorbance measurement at 254 and 400 nm. The correlation coefficients of the calibration curves were ≤ 0.9971 and relative standard deviations were ≤ 2.0 %. Intraday precision was 1.1-1.2 %, interday precision 1.1-1.3 % and recovery 98.5-104.6 %. The average content in leaves was 0.078 (in % of dry weight) for ginkgolide A , 0.072 for ginkgolide B, 0.076 for ginkgolide C, 0.062 for bilobalide and 0.29 % for total diterpene lactones.

herbal, HPTLC, qualitative identification 32e

103 148 A.A. SHIRKHEDKAR*, S.J. SURANA (*Department of Pharmaceutical Chemistry, R. C. Patel College of Pharmacy, Shirpur, Dist:Dhule (M.S.) 425 405, India; atulshirkhedkar@rediffmail.com): Application of a stability-indicating densitometric RP-TLC method for analysis of pioglitazone hydrochloride in the bulk material and in pharmaceutical formulations. J. Planar

Chromatogr. 22, 191-196 (2009). TLC of pioglitazone hydrochloride ((+/-)-5-{p-[2-(5-ethyl-2-pyridyl)ethoxy]benzyl}-2,4-thiazolidinedione hydrochloride and degradation products (after acid and alkaline hydrolysis, oxidation, photochemical and thermal treatment) on RP-18 with acetone - acetic acid - water 40:10:1 in a twin trough chamber saturated for 30 min at room temperature (25 +/- 2 °C). Quantitative determination by absorbance measurement at 225 nm. The limit of detection and quantification was 47 and 141 ng/zone, respectively.

quality control, densitometry, quantitative analysis 32a

103 149 R. SKIBINSKI*, L. KOMSTA, I. KOSZTYLA (*Department of Medicinal Chemistry, Medical University of Lublin, Jaczewskiego 4, 20-090 Lublin, Poland; robert.skibinski@am.lublin.pl): Comparative validation of quetiapine determination in tablets by NP-HPTLC and RP-HPTLC with densitometric and videodensitometric detection. J. Planar Chromatogr. 21, 289-294 (2008). HPTLC of quetiapine on silica gel with hexane - dioxane - propylamine 5:45:2 and on RP-8 with tetrahydrofuran - phosphate buffer (pH 9.0) 1:1 in horizontal chambers. Quantitative determination by absorbance measurement at 243 nm and by videodensitometry at 254 nm. The precision and accuracy of the four methods were fully comparable and no significant differences were observed.

quality control, HPTLC, quantitative analysis, densitometry 32a

103 150 T.G. SONI*, N.P. CHOTAI, P.H. PATEL, L. HINGORANI, R. SHAH, N. PATEL, T. R. GANDHI (*Anand Pharmaceutical College, Near Town Hall, Grid Chokdi, Anand, Gujarat, India; tejalsoni_2973i@yahoo.com): Evaluation of an optimum regression model for high-performance thin-layer chromatographic analysis of aceclofenac in plasma. J. Planar Chromatogr. 22, 101-107 (2009). HPTLC of aceclofenac on silica gel (prewashed with methanol) with toluene - ethyl acetate - methanol - glacial acetic acid 70:20:20:1 in a saturated twin trough chamber. Quantitative determination by absorbance measurement at 270 nm. Linearity was between 0.8 and 14 µg/mL. Average relative residual at each calibration point combined with accuracy and precision were compared using the parameter rank approach. The 1/ x 2 weighted regression model provided best estimates with small relative residuals and the best accuracy and precision.

HPTLC, quantitative analysis, densitometry 32a

103 151 Malgorzata STAREK*, J. KRZEK, M. STOCH (*Jagiellonian University, Collegium Medicum, Department of Inorganic and Analytical Chemistry, Medyczna 9, Cracow, Poland; mstarek@interia.pl): Densitometric analysis of 2-arylpropionate derivatives in pharmaceutical preparations. J. Planar Chromatogr. 21, 251-258 (2008). TLC of tiaprofenic acid, ketoprofen, naproxen, dexibuprofen, flurbiprofen, alminoprofen, and ibuprofen on silica gel with chloroform - acetone - toluene 12:5:2 and chloroform - ethyl acetate 1:1. Quantitative determination by absorbance measurement at 225 nm (for naproxen, dexibuprofen, and ibuprofen) and at 270 nm (for tiaprofenic acid, ketoprofen, flurbiprofen, and alminoprofen).

quality control, densitometry, quantitative analysis, qualitative identification 32a

103 152 G.S. SUBRAMANIAN*, A. KARTHIK, S. B. KAMATH, K. PRABAHAR, A. RANJITHKUMAR, S. PATHAK, N. UDUPA (*Department of Pharmaceutical Quality Assurance, Manipal College of Pharmaceutical Sciences, Manipal, Karnataka 576104, India; ganrajesh@gmail.com): Stability-indicating HPTLC determination of capsaicin in the bulk drug. J. Planar Chromatogr. 21, 271-275 (2008). HPTLC of capsaicin and its degradation products (after acidic and alkaline hydrolysis, oxidation and thermal degradation) on silica gel with toluene - ethyl acetate 3:2. Quantitative determination by absorbance measurement at 280 nm. The limit of detection and quantification was 10 and 60 ng/band, respectively.

quality control, HPTLC, densitometry, quantitative analysis 32a

103 153 P. TENG (Teng Peng)*, X. ZHANG (Zhang Xu), Y. DENG (Deng Yun) (*Pharm. Coll., Chengdu Chinese Trad. & Pharm. Univ., Chengdu, Sichuan 610075, China): (Study of the quality standard

for Qufu Zhuanggu capsules) (Chinese). Chinese J. Modern Appl. Pharm. 25 (1), 66-69 (2008). TLC of Qufu Zhuanggu capsule extracts on silica gel with 1) n-hexane - ethyl acetate 4:1; 2) benzene - ethyl acetate 9:1; 3) chloroform - methanol 19:1; 4) cyclohexane - ethyl acetate 5:2; 5) n-hexane - acetone 3:1. Detection 1) under UV 365 nm; 2) by spraying with 10 % sulfuric acid in ethanol and heating at 105 ºC until the spot were visualized; 3) by treatment with ammonia vapors for 10 min; 4) by spraying with 20 % HClO4 in ethanol. Identification by comparison of the chromatograms with those of the standard psoralen.

quality control, traditional medicine, pharmaceutical research, HPTLC, quantitative analysis, qualitative identification 32e

103 154 A.K. THAKUR*, P.D. HAMRAPURKAR (*Department of Pharmaceutical Analysis, Prin. K. M. Kundanani College of Pharmacy, Jote Joy Building, Rambhau Salgaonkar Road, Cuff Parade, Mumbai 400005, Maharashtra, India; achalthakur@gmail.com): Quantitative densitometric HPTLC analysis of purpurin in the parts of Rubia cordifolia and in pharmaceutical dosage forms. J. Planar Chromatogr. 22, 109-113 (2009). HPTLC of purpurin on silica gel with toluene - ethyl acetate - formic acid 98:2:1 in a twin trough chamber saturated for 20 min at 25 +/- 2 °C. Quantitative determination by absorbance measurement at 255 nm. The limit of detection and quantification was 50 and 100 ng/band, respectively.

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

103 155 R. TIAN (Tian RunTao)*, P. XIE (Xie PeiShan), H. LIU (Liu HePing) (*Chromap Institute of Herbal Medicine Research, Zhuhai, Guangdong, China): Evaluation of traditional Chinese herbal medicine: Chaihu (Bupleuri Radix) by both high-performance liquid chromatographic and high-performance thin-layer chromatographic fingerprint and chemometric analysis. J. Chromatogr. A 1216 (11), 2150-2155 (2009). Chaihu roots (Bupleuri radix), roots of Bupleurum chinense and Bupleurum scorzonerifolium are monographed in the Chinese Pharmacopoeia. Evaluation of the quality of 33 lots of authenticated Chaihu samples versus 31 lots of commercial samples by HPLC-ELSD and HPTLC analysis of the principal bioactive components (saikosaponins). Data acquired from HPLC fingerprints and HPTLC fluorescent images was analyzed by chemometrics for similarity and pattern recognition, including artificial neural networks, k-nearest neighbor (k-NN) and an expert‘s panel. The k-NN classifier showed good performance with sufficient flexibility for processing HPTLC fingerprint images. These images were otherwise not easily dealt with by other algorithms due to the shift of hRf values and varying hue/saturation of the band colors between different TLC plates. The two chromatographic fingerprint methods are complementary measures for quality control. Chaihu roots from different species of the genus Bupleurum could readily be distinguished from each other. Commercial samples of Chaihu can easily be classified by investigating the content of major saikosaponins.

herbal, traditional medicine, HPTLC, qualitative identification 32e

103 156 A. TILAY, M. BULE, J. KISHENKUMAR, U. ANNAPURE* (*Food Engineering and Technology Department, Institute of Chemical Technology, University of Mumbai, Matunga, Mumbai, India, usa@udct.org): Preparation of ferulic acid from agricultural wastes: its improved extraction and purification. J. Agric. Food Chem. 56, 7644-7648 (2008). HPTLC of ferulic acid from agricultural waste (maize bran, rice bran, wheat bran, wheat straw, sugar cane baggasse, pineapple peels, orange peels and pomegranate peels) on silica gel with benzene - dioxane - acetic acid 85:15:1. Detection under UV 254 nm. Purification by adsorption chromatography followed by preparative layer chromatography.

agricultural, HPTLC, preparative TLC 32e

103 157 P. TRIVEDI*, K. PUNDARIKAKSHUDU (*Department of Pharmaceutical Chemistry, K. B. Institute of Pharmaceutical Education and Research, Sector-23, GH-6, Gandhinagar, 382023, Gujarat, India): Novel TLC densitometric method for quantification of solasodine in various So-

lanum species, market samples and formulations. Chromatographia 65 (3-4), 239-243 (2007). TLC of solasodine in various Solanum species on silica gel with a developing solvent containing an organic acid to form ion pair complexes of solasodine with the acid dye. The resulting colored complex was quantified by absorbance measurement at 461 nm. Linearity was between 79 and 495 ng/spot with a correlation coefficient of 0.995. The method eliminates post derivatization steps and the problem of background interference. Application of the method to determine solasodine content in various herb samples, herb extract and their formulations showed an accuracy of 98.5 ± 2.8 % without matrix interference.

quality control, pharmaceutical research, traditional medicine, herbal, densitometry, qualitative identification, quantitative analysis 32e

103 158 E. TYIHÁK*, Á. MÓRICZ, P. G. OTT, M. L. HAJNOS, K. GLOWNIAK (*Plant Protection Institute, Hungarian Academy of Sciences, Herman O. u. 15, POB 102, 1525 Budapest, Hungary; etyih@nki.hu): New approach to mechanism of action of paclitaxel by means of BioArena studies. J. Planar Chromatogr. 21, 331-336 (2008). TLC and OPLC of paclitaxel on silica gel with a variety of mobile phases, e. g. chloroform - methanol 9:1 with chamber saturation. After drying bioautography by immersion in the bacterial suspension of Pseudomonas savastanoi for 20 s. Visualization of the chromatograms with MTT was performed either after a short draining period or after an overnight incubation.

pharmaceutical research, qualitative identification, bioautography 32e

103 159 K. UNIKRISHNAN*, S. RAJA, Indira BALACHANDRAN (*Central for Medicinal Plants Research, Arya Vaidya Sala, Kottakkal, Malappuram 676503, India).: A reverse phase HPLC-UV and HPTLC methods for determination of plumbagin in Plumbago indica and Plumbago zeylanica. Ind. J. of Pharma Sci. 70(6), 844-847 (2008). HPTLC of plumbagin in alcoholic root extracts of Plumbago indica and Plumbago zeylanica on silica gel with n-hexane - ethyl acetate 4:1. The hRf value of plumbagin was 67. Quantitative determination by absorbance measurement at 265 nm. Plumbago indica showed significantly higher plumbagin contents than P. zeylanica. The HPTLC method was the method of choice for simultaneous analysis of several samples, whereas the RP-HPLC method was very sensitive.

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

103 160 A. VENKATACHALAM*, V.S. CHATTERJEE (*Bhavan's College, Department of Chemistry, Andheri West, Mumbai 400058, India): Stability-indicating high performance thin layer chromatography determination of paroxetine hydrochloride in bulk drug and pharmaceutical formulations. Anal. Chim. Acta 598 (2), 312-317 (2007). TLC of paroxetine hydrochloride on silica gel with butanol - acetic acid - water 16:4:1. The hRf value of paroxetine HCl was 48 and separation from the degradation products (produced by acid and alkali hydrolysis, oxidation and photodegradation) was good. Quantitative determination by absorbance measurement at 295 nm. The linearity was in the range of 300 - 1500 ng/spot and the correlation coefficient was 0.9903. The limit of detection and quantitation was 50 and 150 ng/zone, respectively.

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

103 161 G. XU (Xu Guangying)*, X. WANG (Wang Xun), X. DAI (Dai Xixi), M. CHEN (Chen Min) (*Nanjing Univ. TCM, Nanjing 210046, China): (Study of the quality standard for Shujin Huoxue capsules) (Chinese). J. Chinese Trad. Patent Med., 30 (11), 1638-1642 (2008). TLC of Shujin Huoxue capsule extracts on silica gel with 1) petroleum ether (60-90 ºC) - ethyl formate - formic acid 15:5:1; 2) cyclohexane - ethyl acetate 9:1; 3) toluene - acetone - ethanol - ammonia 16:12:1:4. Detection 1) by exposure to iodine vapor; 2) under UV 365 nm; 3) under UV 254 nm. Identification by comparison with the standards of the component drugs. Quantification of paeo-

niflorin by HPLC.

quality control, traditional medicine, pharmaceutical research, HPTLC, quantitative analysis, qualitative identification 32e, 4d

103 162 H. YAN (Yan Hai)*, J. ZHOU (Zhou Jieming), L. WANG (Wang Lisheng), B. ZHOU (Zhou Benhong) (*Pharm. Coll., Wuhan Univ., Wuhan 430072, China): (Study on the quality standard for Sanjin Ganmao pills) (Chinese). J. Chinese Trad. Patent Med. 30 (11), 1635-1638 (2008). TLC of Sanjin Ganmao pill extracts on silica gel with 1) chloroform - methanol 17:3; 2) chloroform - methanol - formic acid 35:5:2; 3) chloroform - methanol - formic acid 90:10:1. Detection 1) by spraying with 10 % sulfuric acid in ethanol and heating; 2) by exposure to ammonia vapor. Identification by comparison with the standard hyperin and other standards of the component drugs.

quality control, pharmaceutical research, traditional medicine, HPTLC, quantitative analysis, qualitative identification 32e

103 163 X. YAN (Yan Xingdong)*, H.NIU (Niu Huifen), J. XU (Xu Jiaxi), G. BAI (Bai Guiying), L. PEI (Pei Lin) (*Hebei Provin. Coll. Chinese Trad. Parm. & Med., Shijiazhuang, Hebei 050031, China): (Study of the quality standard for Tianbing Tiaodu capsules) (Chinese). J. Chinese Trad. Patent Med. 29 (4), 540-542 (2008). TLC of Tianbing Tiaodu capsule extracts on silica gel with cyclohexane - ethyl acetate 4:1. Detection 1) by spraying with vanillin reagent (5 % vanillin in sulfuric acid - ethanol 1:6) and heating until the zones were visualized; 2) under UV 365 nm. Identification by comparison of the chromatograms with that of the standard ferulic acid.

quality control, pharmaceutical research, traditional medicine, herbal, qualitative identification, HPTLC, quantitative analysis 32e

103 164 G. ZHANG (Zhang Guoxia)*, P. LI (Li Ping), B. HAN (Han Biao) (*No. 2 People‘s Hosp., Lanzhou Univ., Lanzhou 330030, China): (Study on the quality standard for Yangxue Fuzheng capsules) (Chinese). J. Chinese Trad. Patent Med. 29 (4), 536-540 (2007). TLC of Yangxue Fuzheng (a traditional Chinese patent medicine) capsule extracts on silica gel with 1) chloroform - methanol - water 13:7:2, 2) chloroform - ethyl acetate - methanol - water 15:40:22:10, 3) ethyl acetate - butanone - methanol - water 10:1:1:1, 4) chloroform - methanol 40:3, 5) petroleum ether (60-90 ºC) - ethyl acetate 1:1. Detection 1) under UV 365 nm, 2) in daylight after spraying with 10 % sulfuric acid in ethanol and heating at 105 ºC.

pharmaceutical research, traditional medicine, quality control, herbal, HPTLC, qualitative identification 32e

103 165 S. ZHANG (Zhang Songan)*, Y. CAI (Cai Yaling), J. YUAN (Yuan Jinlan), X. WU (Wu Xiaohui), CH. FENG (Feng Chao), L. ZHOU (Zhou Lumin), Y. YAO (Yao Youqi), D. ZHOU (Zhou Daonian) (*Pharm. Coll., Tongji Med. Inst., MidChina Univ. Technol., Wuhan, Hubei 430030, China): (Study of the quality standard for Fangji Guanjie pills) (Chinese). J. Chinese Trad. Patent Med. 30 (10), 1478-1482 (2008). TLC of Fangji Guanjie pill extracts on silica gel with 1) petroleum ether (60-90 ºC) - ethyl acetate 10:1; 2) ethyl acetate - formic acid - glacial acetic acid - water 15:1:1:2; 3) petroleum ether (60-90 ºC) - ethyl acetate 17:3; 4) chloroform - methanol - water 40:10:1; 5) diethyl ether - chloroform - methanol 4:2:1. Detection 1) under UV 254 nm; 2) by spraying with 10 % sulfuric acid in ethanol; 3) by spraying with dinitrophenylhydrazine in ethanol and heating at 105 ºC; 4) by spraying with 5 % potassium iodobismuthate solution. Identification by comparison with the standards fangchinoline and tetrandrine.

quality control, pharmaceutical research, traditional medicine, qualitative identification, HPTLC, quantitative analysis 32e

103 166 B. ZHAO (Zhao Bin)*, N. LI (Li Na), J. WU (Wu Jianming), J. LI (Li Jianzhi) (*Sichuan Acad. TCM, Chengdu 610031, China): (Study on the quality standard for Baijian Cichuang suppository) (Chinese). J. Chinese Trad. Patent Med. 30 (10), 1475-1478 (2008). TLC of Baijian Cichuang

suppository extracts on silica gel with 1) n-butanol - glacial acetic acid - water 7:1:2; 2) chloroform - ethyl acetate - formic acid 6:4:1; 3) petroleum ether (60-90 ºC) - diethyl ether - toluene 14:4:3. Detection 1) under UV 365 nm; 2) by spraying with 5 % vanillin in sulfuric acid, followed by heating. Identification by comparison with the standard berberine chloride and other standards of the component drugs.

quality control, pharmaceutical research, traditional medicine, HPTLC, quantitative analysis, qualitative identification 32e

33. Inorganic substances

103 167 Katharina HIEGEL, B. SPANGENBERG* (*University of Offenburg, Institute of Process Engineering, Badstrasse 24, 77652 Offenburg, Germany): New method for the quantification of dequalinium cations in pharmaceutical samples by absorption and fluorescence diode array thin-layer chromatography. J. Chromatogr. A 1216(25), 5052-5056 (2009). Diode array HPTLC of dequalinium chloride on silica gel with methanol - 7.8 % aqueous ammonium acetate 17:3. The hRf value was 58 for dequalinium chloride. Measurement of pure dequalinium chloride in the wavelength range from 200 to 500 nm showed several by-products. By densitometric scanning of a single track in absorption and fluorescence mode 600 spectra in the range from 200 to 1100 nm were measured within 30 s. Sample preparation for an ointment was done by dissolution in methanol. Quantitative determination by absorbance measurement from 315 to 340 nm, by fluorescence measurement from 355 to 370 nm, and by fluorescence measurement from 445 to 485 nm after derivatization with a solution of sodium tetraphenyl borate HCl in water. The linearity range of absorption and fluorescence measurements was between 10 and 2000 ng/zone. Sugar-containing preparations like liquids or lozenges can be reliably quantified only in fluorescence mode, otherwise further sample preparation is necessary.

HPTLC, diode array TLC 33a

103 168 J.P. LAFLEUR, E.D. SALIN* (*Department of Chemistry, McGill University, 801 Sherbrooke Street W. Montreal, Canada H3A 2K6; eric.salin@mcgill.ca): Speciation of chromium by high-performance thin-layer chromatography with direct determination by laser ablation inductively coupled plasma mass spectrometry. Anal. Chem. 80, 6821-6823 (2008). HPTLC of Cr3+ and Cr 6+ on silica gel in a saturated chamber with distilled deionized water and Triton-X-100 in concentrations between 0.001 and 0.1%, which is around the critical micelle concentration. Separation was achieved in seconds over 1 cm. Laser ablation was used to volatilize the chromium species directly from the chromatographic material prior to ICP-MS detection. The reliability of calibration was satisfying with precisions between 3 - 30 % and detection limits in the low ng-range. Silicium, which is present in the silica gel plate, was discussed as suitable internal standard.

toxicology, HPTLC, quantitative analysis 33a, 4e

35.Other technical products and complex mixtures

103 169 Vera BAUMGARTNER*, C. HOHL, U. HAURI (*Kantonales Laboratorium Basel-Stadt, Department Non-Food, Kannenfeldstrasse 2, 4012 Basel, Switzerland; vera.baumgartner@bs.ch): Bioactivity-based analysis of sunscreens using the luminescent bacteria Vibrio fischeri. J. Planar Chromatogr. 22, 19-23 (2009). HPTLC of 26 UV filter substances and their photodegradation products on LiChrospher silica gel (prewashed with methanol) by automated multiple development with mixtures of tert-butyl ether - n-hexane. Detection under UV light at 254 and 366 nm. Bioassay by immersion of plates for 1 s in a suspension of Vibrio fischeri bacteria followed by evaluation in dark.

cosmetics, quality control, HPTLC, AMD 35c

103 170 Iva REZIC*, D. KRSTIC, L. BOKIC (*Laboratory of Analytical Chemistry, Department of Applied Chemistry, Faculty of Textile Technology, University of Zagrab, Prilaz baruna Filipovica 28a, 10000 Zagreb, Croatia; iva_rezic@net.hr): Analysis of waxes on historical samples by thin-layer chromatography. J. Planar Chromatogr. 22, 171-173 (2009). TLC of waxes (carnauba, can-

dellia, lanolin, japanese, spermaceti, bees, and paraffin) on silica gel with petroleum ether - diethyl ether - acetic acid 90:10:1 with chamber saturation. Detection under UV 254 nm.

qualitative identification 35d

38. Chiral separation

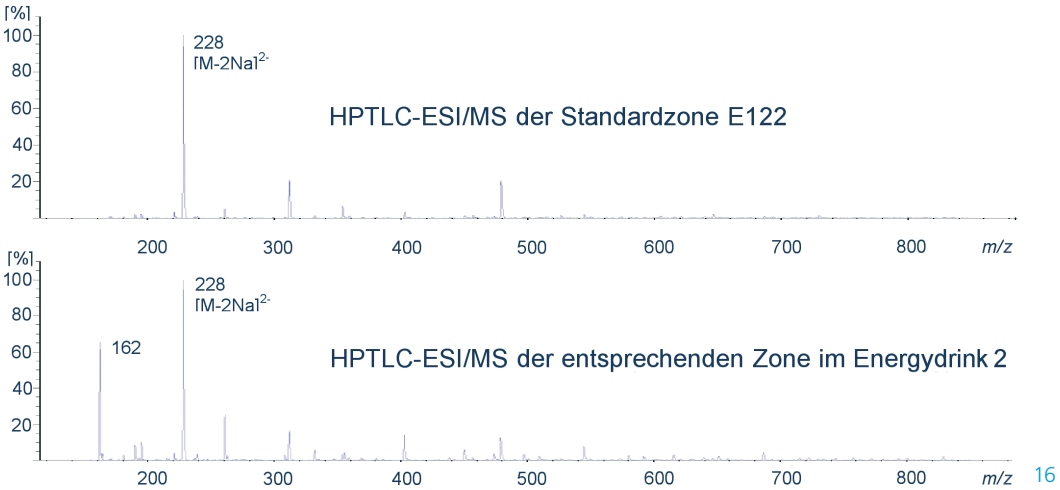
103 171 R. BHUSHAN*, CH. AGARWAL (*Department of Chemistry, Indian Institute of Technology Roorkee, Roorkee 247667, India): Direct enantiomeric TLC resolution of dl-penicillamine using (R)-mandelic acid and L-tartaric acid as chiral impregnating reagents and as chiral mobile phase additive. Biomed. Chromatogr. 22 (11), 1237-1242 (2008). TLC of dl-penicillamine on silica gel 1) impregnated with L-tartaric acid; 2) impregnated with (R)-mandelic acid; and 3) with no impregnation but with chiral modifier added to the mobile phase. The mobile phases used were 1) acetonitrile - methanol - water 5:1:1; 3) ethyl acetate - methanol - water 3:1:1; and 2) acetonitrile - methanol - 0.5 % aqueous L-tartaric acid (pH 5) - glacial acetic acid 70:10:11:7. Detection by exposure to iodine vapors. The limit of detection of both was 120 ng/zone by use of L-tartaric acid (either by impregnation or added to mobile phase), and 110 ng/zone by use of (R)-mandelic acid. (R)-mandelic acid was found to be successful as a chiral impregnating reagent.

pharmaceutical research, HPTLC, quantitative analysis, qualitative identification 38

103 172 M. SAJEWICZ, M. GONTARSKA, D. KRONENBACH, Teresa KOWALSKA* (*Insitute of Chemistry, Silesian University, 9 Szkolna Street, 40-006 Katowice, Poland, kowalska@us.edu.pl): Thin-layer chromatographic and polarimetric investigation of the oscillatory in-vitro chiral inversion of S-(+)-ketoprofen. J. Planar Chromatogr. 21, 349-353 (2008) TLC of S-(+)-ketoprofen on silica gel (prewashed by development with methanol - water 9:1 and impregnated by dipping for 2 s in a solution of L-arginine in methanol) at 22 +/- 2 °C with acetonitrile - water 5:1 acidified with several drops of glacial acetic acid (to maintain the pH <4.8) in one-dimensional and two-dimensional modes. Quantitative determination by absorbance measurement at 252 nm.

qualitative identification, densitometry 38

Fortsetzung von Seite 7



16

HPTLC-ESI/MS-Spektren der Standardzone E122 (m/z 228 $[M-2Na]^{2-}$) und der entsprechenden Zone in Energydrink 2 (m/z 162 aus der Matrix, Chromatographie ohne Essigsäure)

Je nach vorliegender Analyse kann der Grad der Auswertung von visueller Begutachtung über die Aufnahme von Vis-Spektren bis hin zur Aufnahme von Massenspektren erfolgen. Daher ist das offline-Prinzip optimal um Kosten zu minimieren und ein sehr hohes Probenaufkommen leicht zu bewältigen.

	HPLC [4]	HPTLC
Mobile Phase	0,58	0,003
Stationäre Phase	0,64	0,11
Entsorgung	0,04	0,0001
Kosten/Lauf (€)	**1,26**	**0,11**
		=>11 x günstiger
Auftragen/Injektion		0,50
Laufzeit	43	0,20
Detektion		0,10
Zeit/Lauf (min)	**43**	**0,80**
		=> 54 x schneller
davon Arbeitszeit/40 Läufe	keine	5 min

Weitere Informationen sind vom Autor auf Anfrage erhältlich.

[1] G. Morlock, C. Oellig, J AOAC Int 92 (2009) 745

[2] G. Morlock, W. Schwack, Die Aktuelle Wochenschau der GDCh (2009) Woche 21 und 26, www.aktuelle-wochenschau.de/index09.htm

[3] G. Morlock, W. Schwack, GIT 9 (2009) 489–492

[4] K. Minioti et al., Anal Chim Acta 583 (2007) 103

Kontakt: Dr. G. Morlock, Institut für Lebensmittelchemie, Universität Hohenheim, 70599 Stuttgart, gmorlock@uni-hohenheim.de

Screening unbekannter Pflanzenextrakte mittels Planar-Chromatographie

17

Von links nach rechts: M. Schulz, S. Minarik, C. Wirth und M. Oberle

Nicht nur bei der Herstellungskontrolle der DC/HPTLC-Schichten, auch in der Routineanalytik verwendet Merck die Planar-Chromatographie. Dieser Beitrag entstand durch eine Zusammenarbeit der Laboratorien Dünnschicht-Chromatographie und Kosmetik in der Forschungsabteilung Performance & Life Science Chemicals.

Einleitung

Die Planar-Chromatographie findet Anwendung in einer Vielzahl kosmetischer Applikationen. Typische Anwendungen sind Wirkstoff-Screening, Applikationen mit schwer aufzuschliessenden Proben und auch Quantifizierungen von Wirkstoffen wie z. B. UV-Filtersubstanzen. In dieser Arbeit wird die HPTLC als Screening-Methode zur Erstanalyse potentieller Wirkstoffe auf Pflanzenextraktbasis beschrieben. Dazu liegt einerseits der Schwerpunkt bei Substanzen, die durch ihre kosmetische Relevanz und ihren hohen Gehalt in der Pflanze auffallen und andererseits bei der Substanzklasse der Flavonoide, die in der Kosmetik als wirksame Naturstoffe anerkannt sind.

Die Vorteile der Planar-Chromatographie können hier hervorragend genutzt werden. Es werden viele Proben auf einer Platte parallel analysiert und damit geringe Analysezeiten und -kosten erreicht. Eine hohe Flexibilität in der Detektion wird durch den Einsatz verschiedener Derivatisierungsreagenzien gewährleistet. Potentielle Wirkstoffkandidaten lassen sich schnell und einfach identifizieren und im nächsten Schritt mit strukturaufklärenden Methoden weitergehend charakterisieren.

Standardlösung

Chlorogensäure, Hyperosid, Rutin, Quercetin und Kaempferol in Methanol (0.1 %ig)

Probenvorbereitung

Die Pflanze wird zerkleinert und zu einem Rohextrakt verarbeitet. Aus dem Rohextrakt werden durch flüssig/flüssig-Extraktion drei weitere Extrakte unterschiedlicher Polarität hergestellt.

Schicht

HPTLC-Platten Kieselgel 60 F_{254s} Merck, 20 × 10 cm

Probenauftragung

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

Chromatographie

In der Doppeltrogkammer 20 × 10 cm mit Ethylacetat – Ameisensäure – Eisessig – Wasser 100:11:11:27

Postchromatographische Derivatisierung

Mit dem DC-Sprühgerät werden folgende Derivatisierungen durchgeführt*:

- Naturstoffreagenz nach Neu (NSR): 1 % Diphenylborsäure-2-aminoethylester in Methanol → UV 366 nm
- Anisaldehyd-Schwefelsäure-Reagenz (AAS): 0,5 mL Anisaldehyd in 85 mL Methanol, 10 mL Eisessig und 8 mL konzentrierter Schwefelsäure (zugesetzt unter Eiskühlung) → Platte bei 90–125 °C für max. 15 min erhitzen → Weisslicht
- Diphenyl-2-picrylhydrazyl-Reagenz (DPPH): 0,1 % Diphenyl-2-picrylhydrazyl in Methanol → Platte bei 40 °C für max. 2 min erwärmen → Weisslicht
- Rhodamin B-Reagenz (RDB): 0,1 % Rhodamin B in Methanol → UV 366 nm
- Dragendorff-Reagenz (DRR): Merck Fertiglösung → Platte bei 40 °C für max. 2 min erwärmen → Weisslicht

**Anmerkung: Das Tauchen sollte dem Sprühen aus vielfältigen Gründen vorgezogen werden, wie z. B. homogenere Aufbringung des Reagenzes auf die Schicht, weitaus geringere Laborluftbelastung und sauberer Arbeitsplatz.*

Ergebnisse und Diskussion

In den zwei folgenden Beispielen wurde jeweils der Rohextrakt einer Pflanze (Bahn 1) und deren drei Extrakte von unterschiedlicher Polarität (A–C in steigender Polarität, Bahn 2–4) chromatographiert und mit unterschiedlichen Derivatisierungsreagenzien detektiert.

Reagenz	detektiert
NSR (Naturstoffreagenz nach Neu)	Flavonoide, Penicillinsäure, Kohlenhydrate, Anthocyanidine
AAS (Anisaldehyd-Schwefelsäure-Reagenz)	Ätherische Ölkomponenten, Terpene, Steroide, Sapogenine, Prostaglandine, Antioxidanzien, Antibiotika
DPPH (Diphenyl-2-picrylhydrazyl-Reagenz)	Ätherische Öle
RDB (Rhodamin B-Reagenz)	Lipophile Substanzen, Lipide, Phenole, Polyphenole, Flavonole, Tenside
DRR (Dragendorff-Reagenz)	Alkaloide

Beispiel 1

Mit dem NSR-Reagenz wurden z. B. im Extrakt A (Bahn 2) die unpolaren Chlorophyll-Verbindungen als rote Bande im Frontbereich und im Extrakt C (Bahn 4) Pflanzensäuren detektiert.

Als Hauptkomponente wurde eine Pflanzensäure angenommen, deren R_F-Wert über dem der Chlorogensäure (Bahn 5, blaue Bande bei RF 0,5-0,6) liegt. Diese Verbindung ist in den Extrakten B und C (Bahn 3 und 4) angereichert. Sie reagiert mit dem AAS- und sehr intensiv mit dem DPPH-Reagenz, was auf eine antioxidative Wirkung deutet. Weiterhin ist der Chlorophyllanteil im Rohextrakt (Bahn 1) und in den Extrakten A und B (Bahn 2 und 3) zu erkennen. Dieser zeigt auch mit DRR eine Farbreaktion. Ausser im Extrakt A können weitere Verbindungen sowohl mit höherem als auch mit niedrigerem R_F-Wert nachgewiesen werden, welche mit NSR hellblaue und mit AAS bräunliche Zonen ergeben.

Beispiel 2

Im zweiten Pflanzenbeispiel liegen als Hauptkomponenten neben Pflanzensäuren auch Flavonoide vor. Mit dem NSR-Reagenz ist dies erkennbar (1) am R_F-Bereich, (2) der gelben, grünen bzw. hellblauen Färbung der Banden und (3) der Anreicherung in den Extrakten B und C (Bahn 3 und 4). Weiterhin sieht man im Extrakt B (Bahn 3) die grüngelbe Bande, welche für ein Kaempferol-Derivat spricht, und die gelbe Bande an der Front, welche das Quercetin

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Chromatogramm-Tauchvorrichtung

Die Derivatisierung durch automatisiertes Tauchen hat gegenüber dem manuellen Sprühen in vielen Fällen eindeutige Vorteile. Durch die Einstellung einer gleichmässigen, vertikalen Tauchgeschwindigkeit (wählbar zwischen 30 und 50 mm/s) und Verweilzeit (wählbar von 1 bis 8 s sowie unendlich) lassen sich die Tauchbedingungen standardisieren. Durch die homogene Reagenzaufbringung werden Fliessmittelfront-ähnliche Tauchlinien vermieden, die bei der densitometrischen Auswertung störend wirken. Das batteriebetriebene Gerät ist auf eine Eintauchtiefe für 10 und 20 cm hohe Platten einstellbar.

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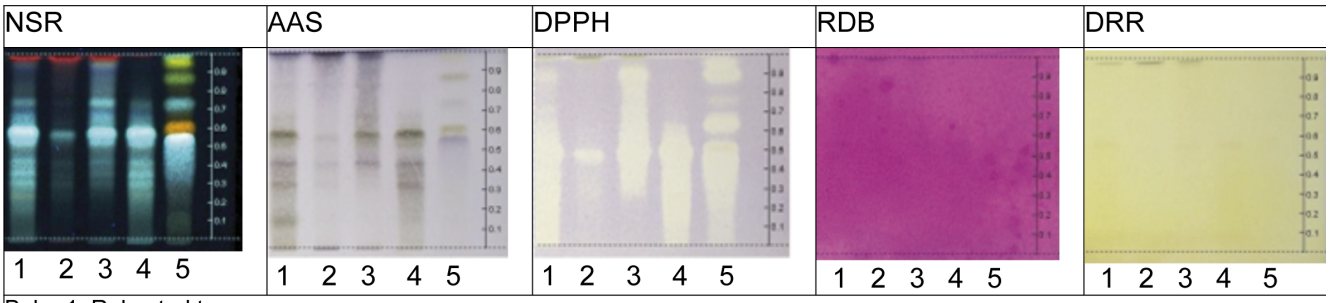
DC-Plattenheizer

Für viele Reaktionen ist eine kontrollierte Temperaturerhöhung erforderlich. Der Temperatur-Regelbereich des DC-Plattenheizers beträgt 25–200 °C und ermöglicht eine gleichmässige Erhitzung über die ganze Platte. Das CERAN®-Keramikfeld ist mit einem Raster versehen, der die richtige Plazierung der Platte erleichtert. Es ist beständig gegenüber allen üblichen Reagenzien und leicht zu reinigen.

Beispiel 1



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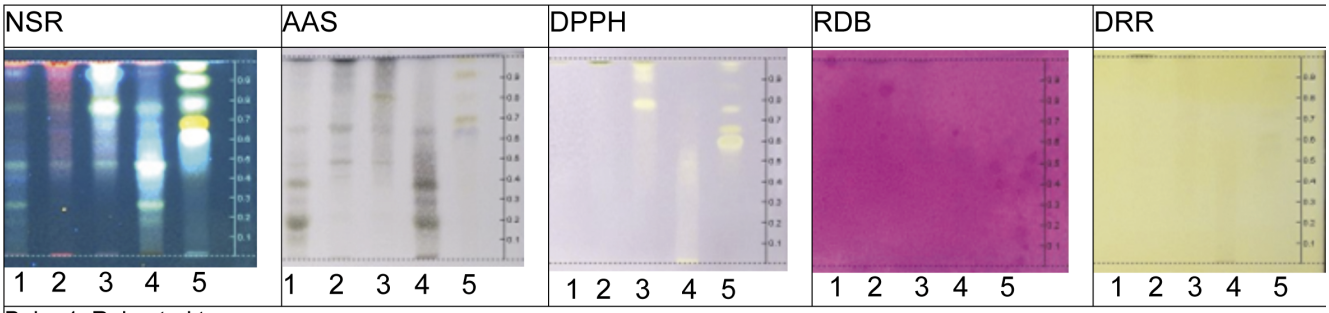
Bahn 1: Rohextrakt
Bahn 2: Extrakt A (unpolar)
Bahn 3: Extrakt B (mittelpolar)
Bahn 4: Extrakt C (polar)
Bahn 5: Standardmischung (nach aufsteigenden R_F geordnet: Chlorogensäure, Hyperosid, Rutin, Kaempferol und Quercetin)

Beispiel 2



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Bahn 1: Rohextrakt
Bahn 2: Extrakt A (unpolar)
Bahn 3: Extrakt B (mittelpolar)
Bahn 4: Extrakt C (polar)
Bahn 5: Standardmischung (nach aufsteigenden R_F geordnet: Chlorogensäure, Hyperosid, Rutin, Kaempferol und Quercetin)

zeigt. In Extrakt C (Bahn 4) sind ausserdem mit blauer Bande mehrere Pflanzensäuren detektiert. Potentielle Flavonoide zeigen mit DPPH-Reagenz eine intensive Reaktion.

Im Screening von Pflanzenextrakten, d.h. für die analytische Erstcharakterisierung der zu bewertenden Pflanzenextrakte auf angereicherte Inhaltsstoffe und Nebenkomponenten hat sich die Planar-Chromatographie bei Merck etabliert. Der hohe Probendurchsatz und die Vielzahl möglicher Derivatisierungen machen sie zu einem leistungsfähigen und sehr wichtigen Baustein im eingesetzten Methodenpool. Zur weiteren Bewertung wird der Wellenlängenbereich von 210–400 nm mit einem HPLC-DAD-System ausgewertet und ein Aktivitätsscreening durchgeführt, z. B. ein Schnelltest auf antioxidativ wirkende Substanzen. Nach Auswertung der Screening-Ergebnisse wird entschieden, welche Wirkstoffkandidaten weitergehend untersucht werden, z. B. zur Strukturaufklärung mit HPLC-MS* und NMR.

Anmerkung: Mit dem neuen TLC-MS-Interface wird von der Zone auf der Platte durch online Extraktion das Massenspektrum innerhalb einer halben Minute direkt erhalten.

Weitere Informationen sind von den Autoren auf Anfrage erhältlich.

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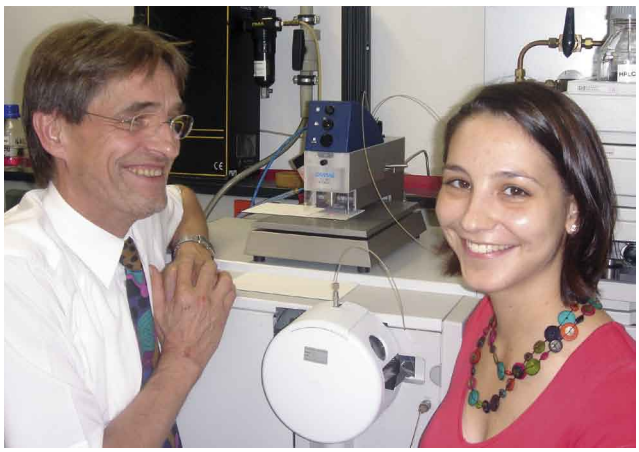
Frankfurter Str. 250, 64293 Darmstadt

HPTLC-Bestimmung von unerlaubt zugesetzten fettlöslichen Azofarbstoffen in Gewürzen



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Prof. Dr. Wolfgang Schwack und Elodie Pellissier

HPTLC ist konkurrenzlos im schnellen und matrix-robusten Screening von vielen Proben parallel. Im Falle von Azofarbstoffen mit einer starken Absorption im Weißlicht ist eine gute Detektierbarkeit zu erwarten. Neben der Bibliothekssuche der UV/Vis-Spektren bietet die Kopplung mit der Massenspektrometrie eine verbesserte Möglichkeit, positive Funde zu bestätigen. Mit MassWorks-Software kann der Analytiker sogar von niedrig auflösenden Massenspektrometern die exakte Masse und Elementarzusammensetzung erhalten. Aufgrund dieser Vorteile wurde eine schnelle und verlässliche HPTLC-Methode zur Identifizierung und Quantifizierung der relevanten Farbstoffe in verschiedenen Gewürzpulvern und Gewürzpulvermischungen (Chili, Curry, Paprika etc.) entwickelt. Die auf Pasten und Saucen erweiterte Methode ist momentan in der Validierung.

Sowohl der Beitrag Kandler et al. (S. 2 ff - Kantonales Labor Zürich) als auch der vorliegende gehen dasselbe Thema an, jedoch auf unterschiedliche Weise. Beide bieten gegenüber dem Status quo der Analytik von Azofarbstoffen deutliche Vorteile. Der Anwender mag entscheiden, welchen Weg er bevorzugt.

Die Untersuchungen wurden während der Bachelorarbeit von Elodie Pellissier (Fachhochschule Westschweiz) am Institut für Lebensmittelchemie der Universität Hohenheim in Stuttgart durchgeführt.

Einleitung

Sudan I wurde 2003 in Frankreich in einer indischen Chili-Lieferung nachgewiesen. Dies war die erste Warnung an die EU-Mitgliedstaaten durch das Schnellwarnsystem im Lebens- und Futtermittelbereich (Rapid Alert System for Food and Feed, RASFF). Danach wurden zunehmend Produkte mit unerlaubt zugesetzten, fettlöslichen Azofarbstoffen gefunden, z. B. mit Sudanrot B, Sudanorange G und Pararot sowie die von der internationalen Agentur für Krebsforschung (International Agency for Research on Cancer, IARC) als Kanzerogene der Kategorie 3 eingestuften Farbstoffe Sudan I bis IV und Sudanrot 7B. Im Jahr 2008 und im ersten Halbjahr 2009 wurden nicht weniger als 60 Fälle im RASFF dokumentiert. [1]

Standardlösungen

Sudan I (I), Sudan II (II), Sudanrot B (B), Sudanorange G (OR) und 4-Dimethylaminoazobenzen (interner Standard, IS) wurden einzeln (10 mg) in je 5 mL Aceton gelöst, Sudan III (III), Sudan IV (IV), Sudanrot 7B (7B) und Pararot (PR) in je 15 mL. Die Lösungen wurden auf 20 mL mit Methanol aufgefüllt (Stammlösungen von je 0.5 mg/mL). Für die Standardgemischlösung wurden je 200 µL der entsprechenden Stammlösungen gemischt und mit Methanol auf 10 mL aufgefüllt (Farbstoff je 10 ng/µL). Die interne Standardlösung zum Übersprühen wurde entsprechend 1:50 verdünnt.

Probenvorbereitung

Die homogenisierte Gewürzprobe (1 g) wurde in ein 20 mL Zentrifugenglas mit Schraubkappe eingewogen. Nach Zugabe von 1 mL der Stammlösung des internen Standards und 4 mL Aceton wurde das Zentrifugenglas mit dem Vortex für 1 min homogenisiert. Nachfolgend wurden 5 mL Methanol hinzugefügt. Das Zentrifugenröhrchen wurde von Hand 1 min geschüttelt und bei 4000 rpm zentrifugiert. Ohne weitere Cleanup-Schritte wurde der Überstand direkt zur HPTLC eingesetzt.

Schicht

HPTLC-Platten NANO-SIL-PAH, 20×10 cm, Macherey-Nagel (Coffein-imprägniert)

Anmerkung: Der Einsatz von bereits imprägnierten Fertigplatten ist sehr benutzerfreundlich. Notfalls kann die Imprägnierung der Schicht selbst hergestellt werden durch 20 minütiges

Tauchen einer HPTLC-Platte Kieselgel 60 in eine Coffeinlösung (1.7 g Coffein in 100 mL Acetonitril) und abschliessendes Trocknen für 20 min bei 120 °C.

Probenauftragung

Bandförmig mit ATS4, Bandlänge 8 mm, Bahnabstand 10 mm, Abstand vom unteren Plattenrand 8 mm, Abstand vom seitlichen Plattenrand min. 15 mm, Auftragevolumen 1–20 µL der Standardlösung (übersprüht mit 10 µL interner Standardlösung) und 4 µL der Probenextrakte

Chromatographie

In der ADC2 mit Isohexan – Methylethylketon 5:1 nach Kammersättigung für 10 min über eine Laufstrecke von 68 mm. Die Plattenaktivität wurde mit der Option Feuchtekontrolle über eine gesättigte Kaliumcarbonatlösung (45 % relative Luftfeuchtigkeit) für 4 min eingestellt.

Dokumentation

Im DigiStore 2 unter Weisslicht (Reflektion und Transmission)

Densitometrie

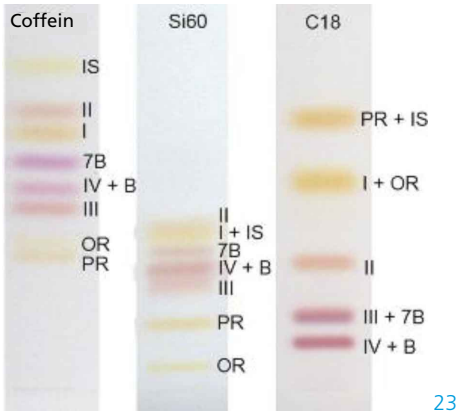
Absorptionsmessung via Mehrwellenlängen-Scan (390, 415, 500, 525 und 550 nm) und Spektrenaufnahme (320–600 nm) mit TLC-Scanner 3 und winCATS-Software.

Massenspektrometrie

Mit dem TLC-MS-Interface ausgerüstet mit dem ovalen Elutionskopf (4 x 2 mm) und verbunden mit einem Agilent 1100 LC/MSD-System, Aufnahme im positiven ESI-Modus, Zonenextraktion durch Methanol – 0.1 % Ameisensäure 95:5 bei 0.2 mL/min. Eine Chromolith RP 18-Säule (50 x 4.6 mm, Merck) wurde in die Verbindungskapillare zwischen TLC-MS-Interface und MSD eingebaut. Exakte Massen wurden mit MassWorks-Software (Cerno Bioscience, Danbury, CT, USA) berechnet.

Ergebnisse und Diskussion

Obwohl unterschiedliche chromatographische Systeme im Normal- und Umkehrphasenmodus während der Methodenentwicklung benutzt wurden, war die Chromatographie der in Gewürzen und Pflanzenölen in den letzten Jahren am häufigsten gefundenen acht Azofarbstoffe [1] auf Coffein-imprägnierten Schichten vergleichsweise am besten. Die Regioisomeren Sudan IV und Sudanrot B sind generell schwer zu trennen, auch mittels LC/MS-MS [2].



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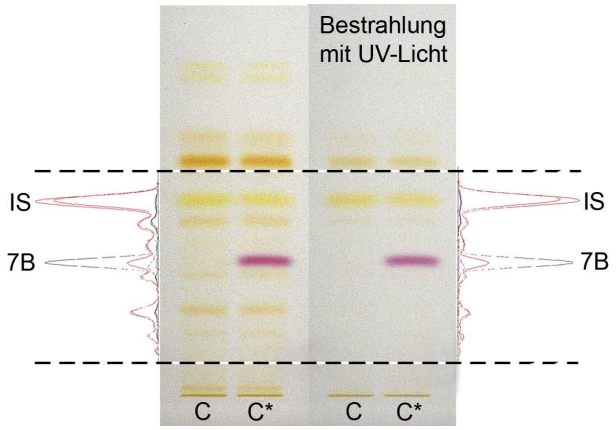
Trennung der 8 Azofarbstoffe auf Coffein-imprägnierten HPTLC-Platten NANO-SIL-PAH, HPTLC-Platten Kieselgel 60 und HPTLC-Platten RP 18 gemäß [3]

Statt zeitaufwändiger, in der Literatur beschriebener Cleanup-Schritte wurden die Probenextrakte direkt auf die HPTLC-Platten aufgetragen und nach der Chromatographie einer Bleichung unterworfen. Bleichung mit starkem UV-Licht (600 W/m^2) für max. 5 min führte zu einer von Matrixstörungen nahezu freien Basislinie.



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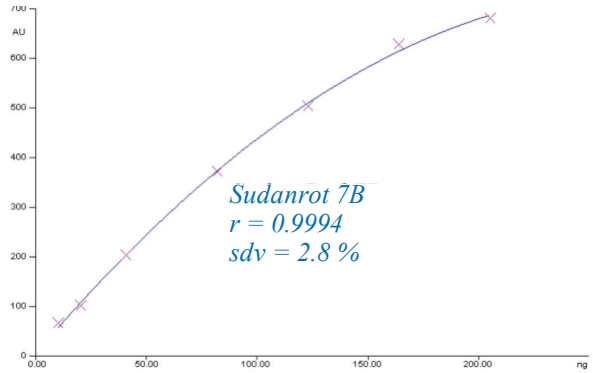
Effekt der UV-Bestrahlung auf den Matrix-Hintergrund im Chromatogramm eines originalen (C) und dotierten (C, Sudan Red 7B, 500 mg/kg) Chilipulver-Extraktes*

Nach dem Mehrwellenlängen-Scan bei 5 ausgewählten Wellenlängen wurde zur Quantifizierung die Mehrbereichskalibration mit interner Standardauswertung (zur Korrektur der Probenvorbereitung) verwendet. Die Kalibration erfolgte nach einer polynomen bzw. linearen Regression entsprechend einem weiten bzw. engen Arbeitsbereich. Gemäss der angegebenen Probenvorbereitung lagen die Nachweisgrenzen (LOD) im Bereich von 10 mg/kg, was im Hinblick auf die zu erwartenden Zusätze zur gewünschten Farbkorrektur der Produkte ausreichend war.



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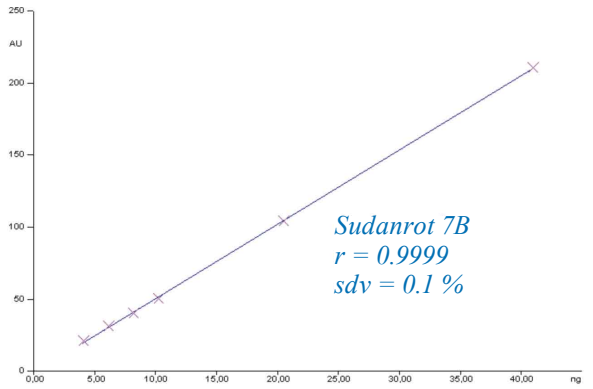

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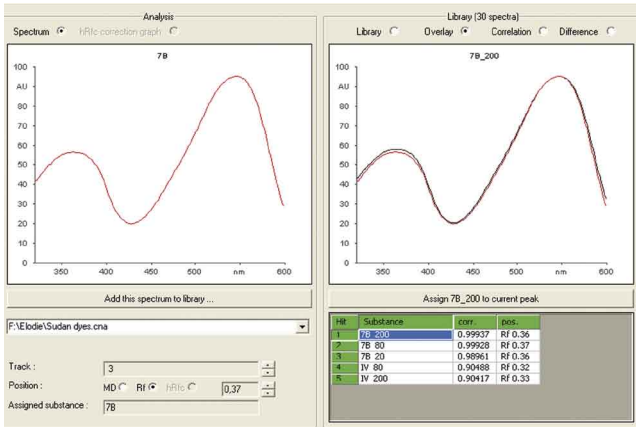
Polynome und lineare Kalibration über einen weiten (10 - 200 ng/Band) bzw. engen (4 - 40 ng/Band, ab LOD) Kalibrationsbereich am Beispiel von Sudanrot 7B

In einer Spektrenbibliothek (aufgebaut auf 3 Konzentrationen pro Azofarbstoff) kann nach passenden UV/Vis-Spektren gesucht werden, um einen verdächtigen Fund in einer Probe zu bestätigen.



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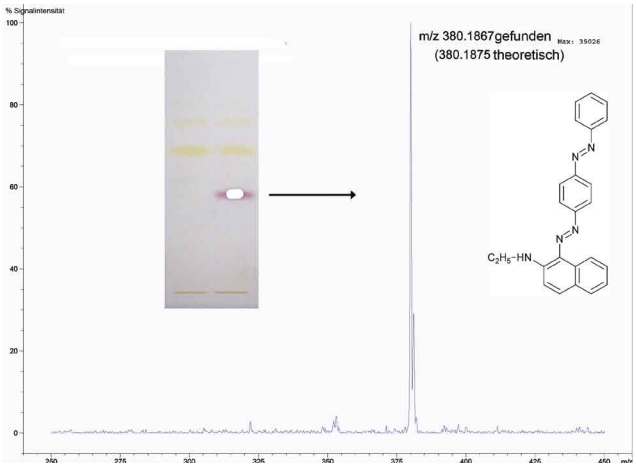
Spektrenvergleich von Sudanrot 7B in einer dotierten Probe (rot, 500 mg/kg Sudanrot 7B) und dem besten Treffer in der Spektrenbibliothek (Korrelation: 0.9994).

Zur zweifelsfreien Bestätigung positiver Funde können Massenspektren von verdächtigen Zonen mit dem TLC-MS-Interface aufgenommen werden. Die Massengenauigkeit inklusive der Elementarzusammensetzung kann durch die Verwendung der MassWorks-Software verbessert werden, sogar bei niedrig auflösenden Massenspektrometern.



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Als zweite Bestätigung des Sudanrot 7B-Fundes, das HPTLC-MS Spektrum der verdächtigen Zone mit Berechnung der »exakten« Masse durch MassWorks-Software

Die orthogonale online HPTLC-HPLC-MS-Kopplung wurde hier zum ersten Mal vorgestellt. Neben der Trennung von coextrahiertem Coffein (Schichtimprägnierung) und dem Azofarbstoff ermöglicht diese einfache Kopplung eine zweite Selektivitätsrichtung (C18-Säule versus Kieselgelplatte). Demzufolge können Azofarbstoff-Funde leicht bestätigt werden durch (1) UV/Vis-Spektren, (2) Massenspektren und (3) ein zweites chromatographisches System unterschiedlicher Selektivität. All diese Absicherungen bietet je nach Anforderung ein einziger HPTLC-Lauf - und das für viele Proben parallel.

[1] Rapid Alert System for Food and Feed, http://ec.europa.eu/food/food/rapidalert/index_en.htm

[2] H. San, F. Wang, L. Ai, J Chromatogr A 1164 (2007) 120

[3] H. Kandler, M. Bleisch, V. Widmer, E. Reich, J Liq Chromatogr Rel Technol 32 (2009) 1273

Weitere Informationen sind vom Autor auf Anfrage erhältlich.

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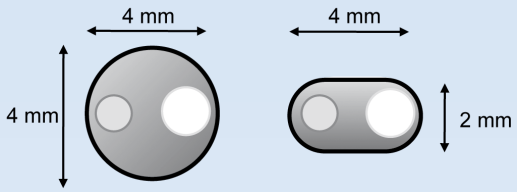
NEU

TLC-MS Interface jetzt mit rundem und ovalem Elutionskopf

Komfortabler Transfer von Trennzonen direkt von der Platte in Ihr MS



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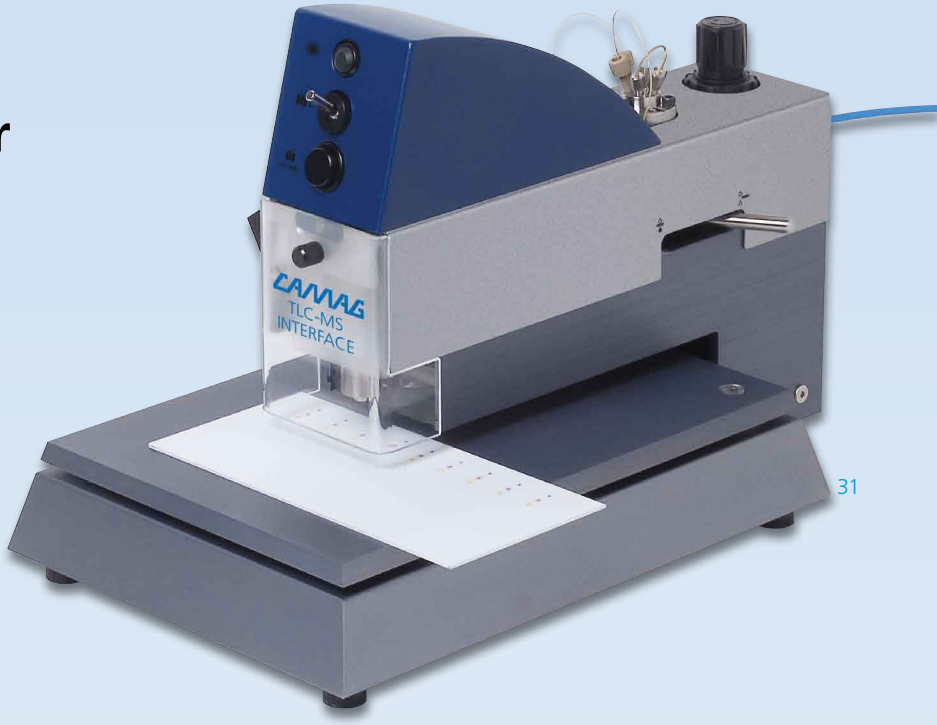
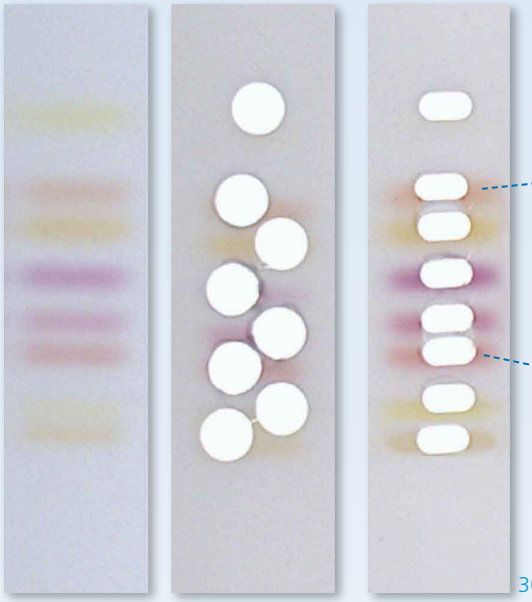
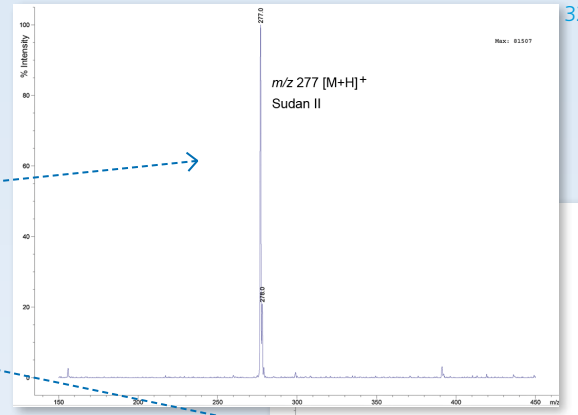
Abmessungen des runden und ovalen Elutionskopfes



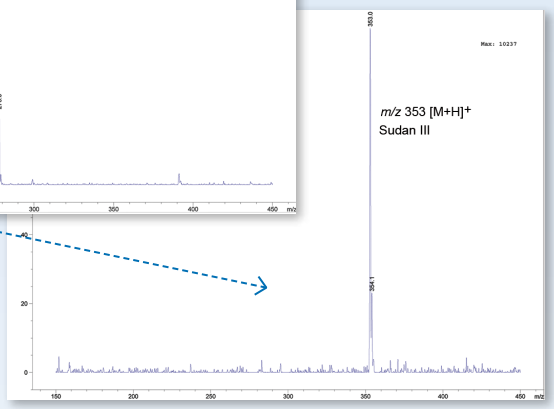
31



30



32



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Der ovale Elutionskopf ist zur Extraktion eng nebeneinander liegender Banden optimal.

CAMAG

Weltweit führend in der Planar-Chromatographie

CBS 103 D 996.1310-6