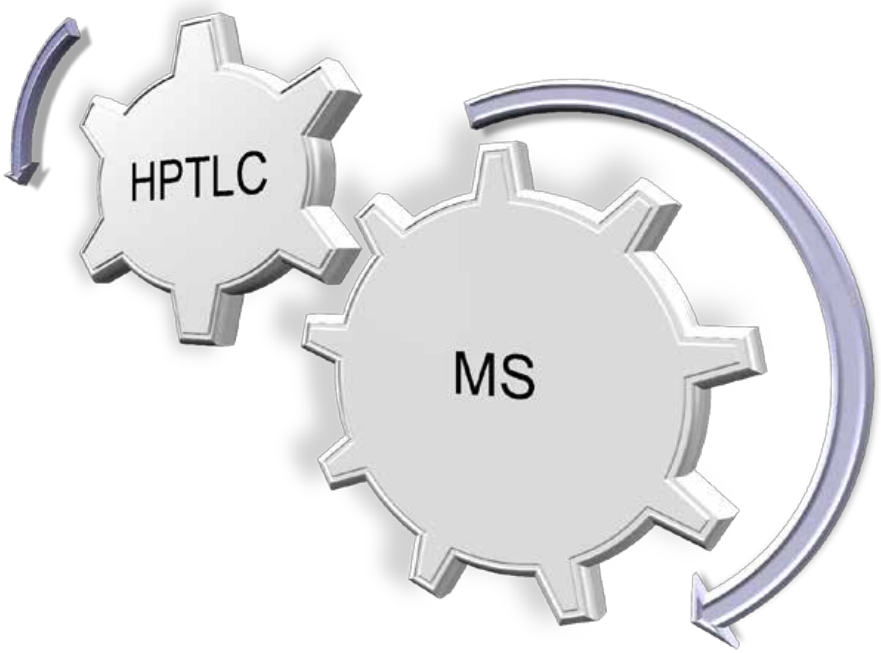
HPTLC-MS

Overview, quantitation and comparison



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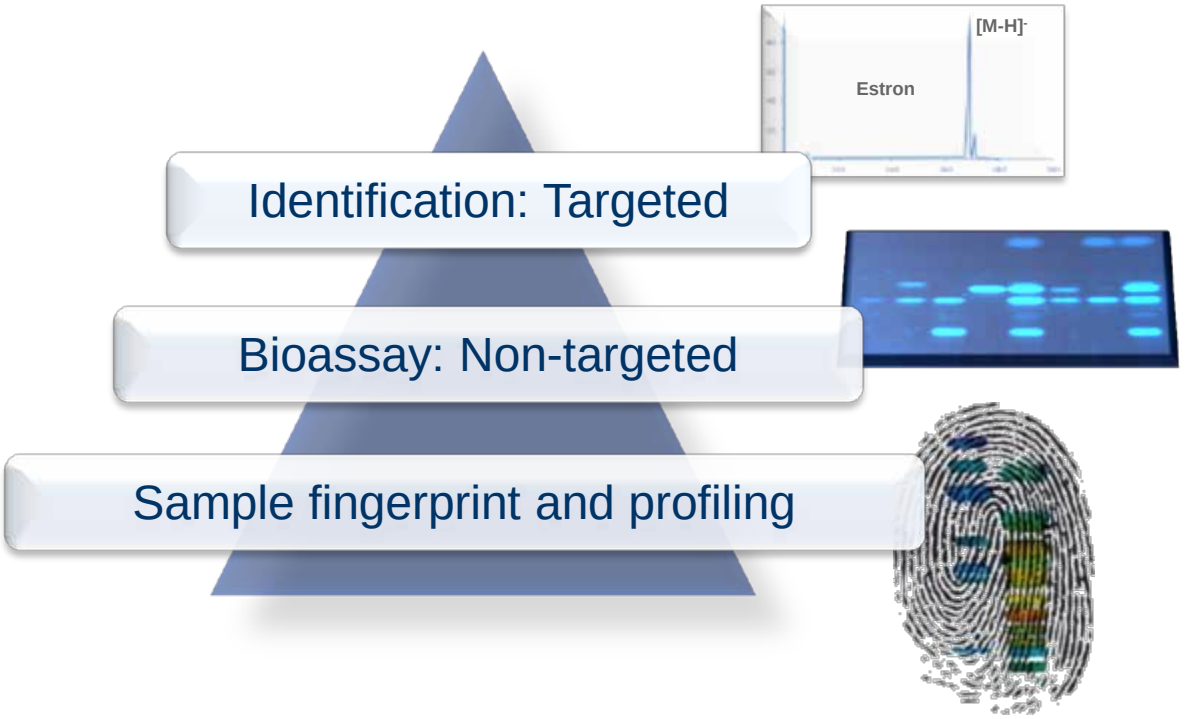
Justus Liebig University Giessen

News in HPTLC-EDA



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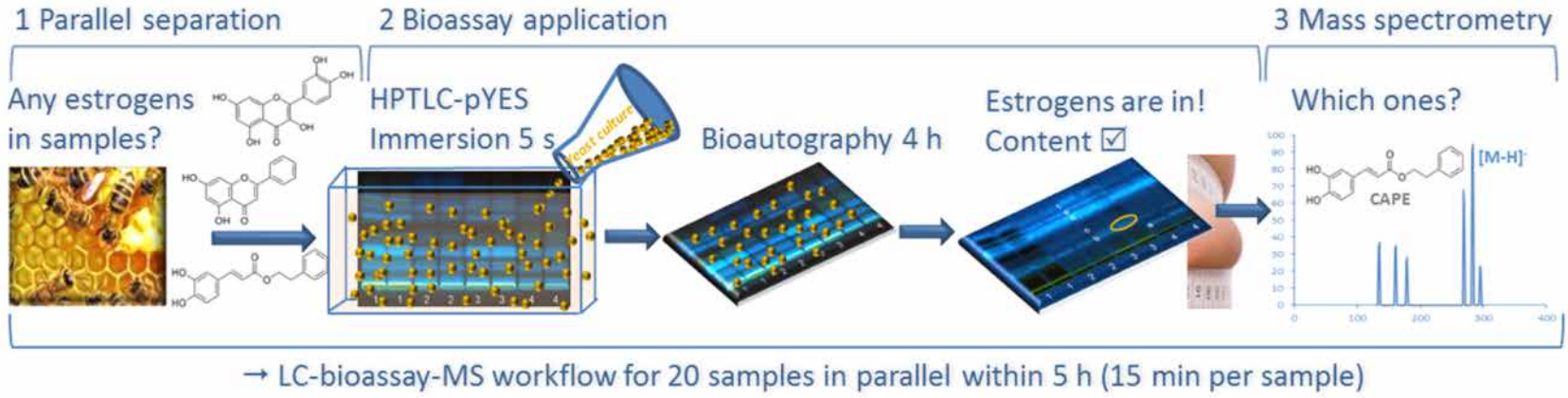
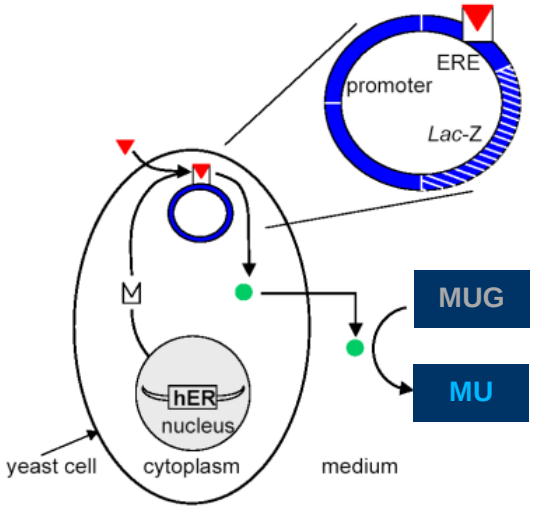
Detection of endocrine disrupting compounds (EDCs)





Modified from draft of pYES expert group

Planar yeast estrogen screen (pYES)

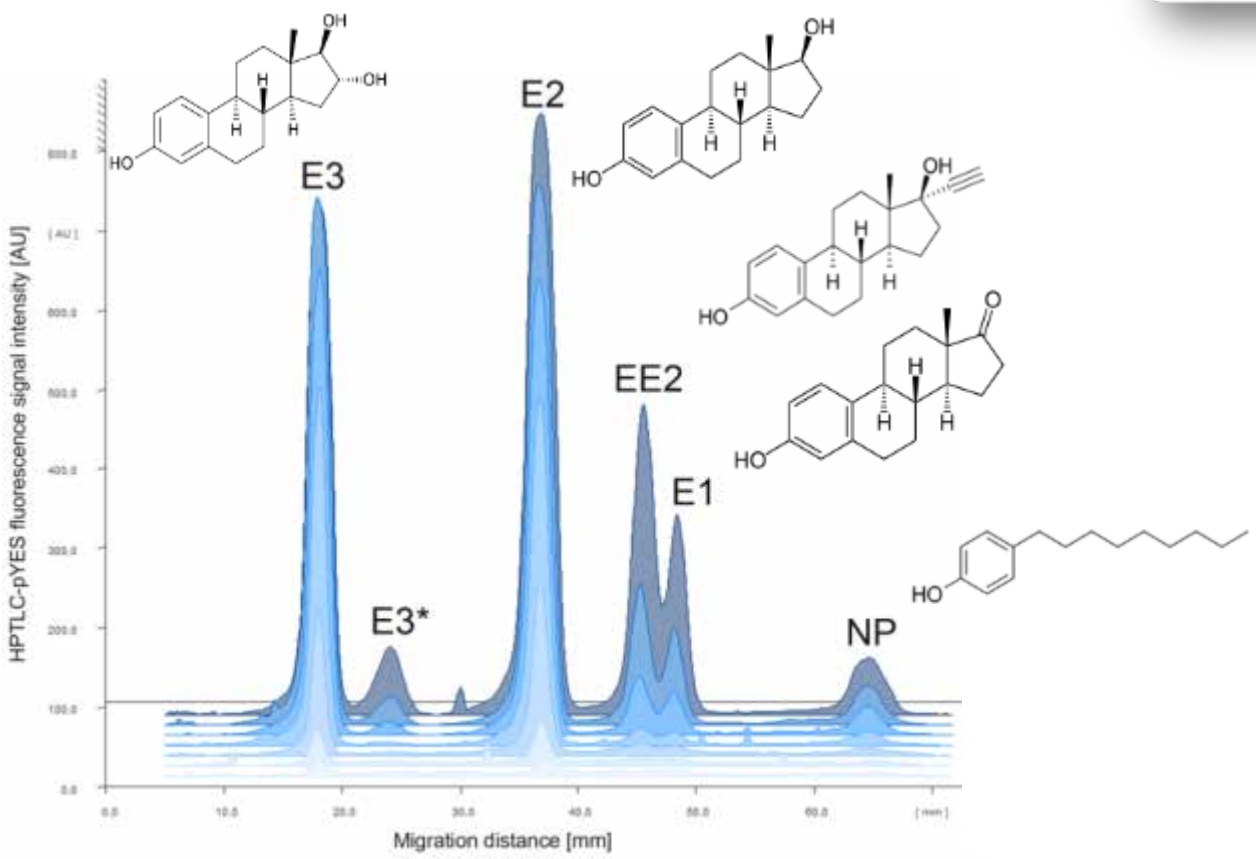
→ using human estrogen receptor hERα

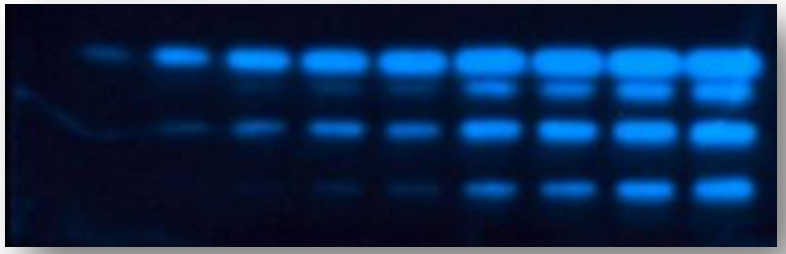
→ in *Saccharomyces cerevisiae*

1. Routledge & Sumpter, Environ. Toxicol. Chem. 15 (1996) 241
2. McDonnell *et al.*, J. Steroid Biochem. Mol. Biol. 39 (1991) 291

→ blue fluorescent 4-methylumbelliferone

Biological detection of estrogens

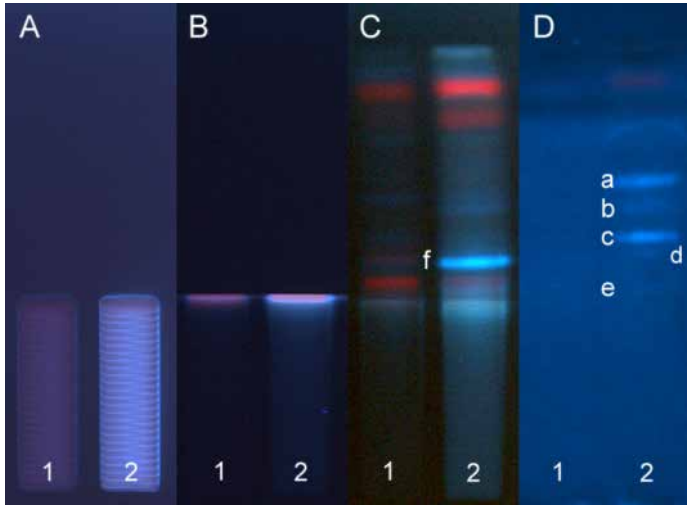


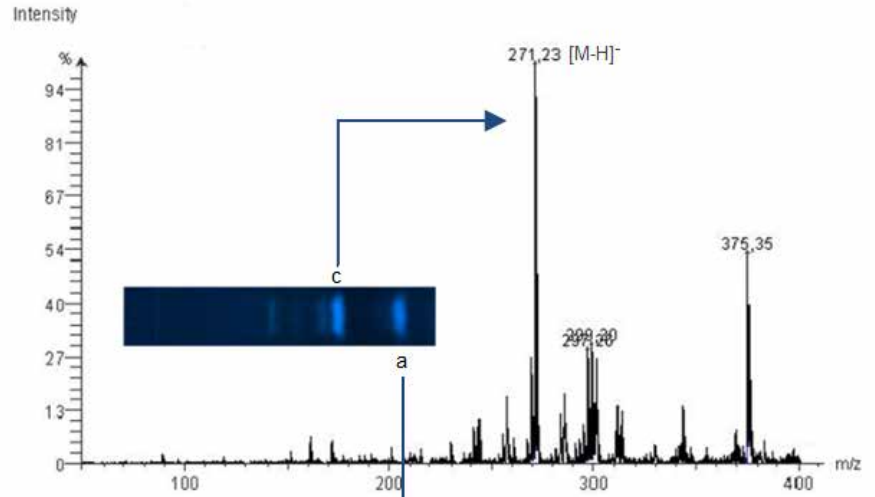


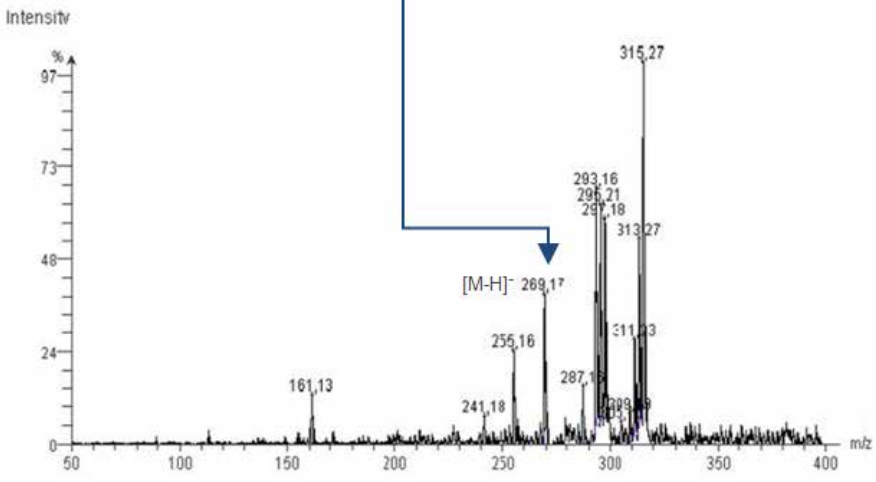
	pg/band
E1	25 – 1000
EE2	0.5 – 20
E2	0.5 – 20
E3	25 – 1000

I. Klingelhöfer, G. Morlock, J Chromatogr A 1360 (2014) 288-295

Discovery in surface and waste water samples







Substance	LOD [ng/L]	LOQ [ng/L]
E2	1.0	2.5
EE2	2.5	5.0
E1	4.3	15.0
E3	75.0	250.0
BPA	1.6 x 10^3	5.0 x 10^3
NP	15.0 x 10^3	65.0 x 10^3

I. Klingelhöfer, G. Morlock, Anal Chem 87 (2015) 11098−11104

Biouantitation by microorganisms´ response

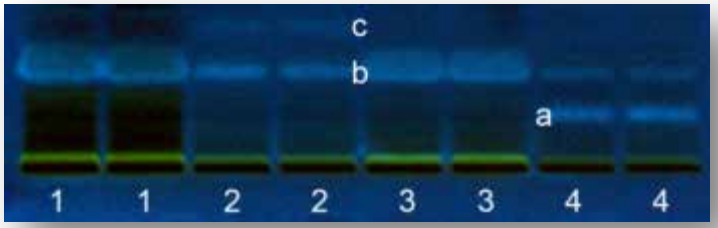
Content [ng/L]	E2	E1	E3
STP influent	10.1	40.5	98
	6.7	17.6	nd
	3.1	12.4	nd
	4.9	36.4	150
	12.6	49.7	210
STP effluent (x5)	nd	nd	nd
Lückebach	1.6	20.5	nd
	8.3	12.7	nd
Flachsbach	6.6	16.6	nd
	nd	nd	nd
Wetter (x2)	nd	nd	nd
Weidgraben	2.1	11.6	nd
	**	4.9	nd
Wieseck (x2)	nd	nd	nd

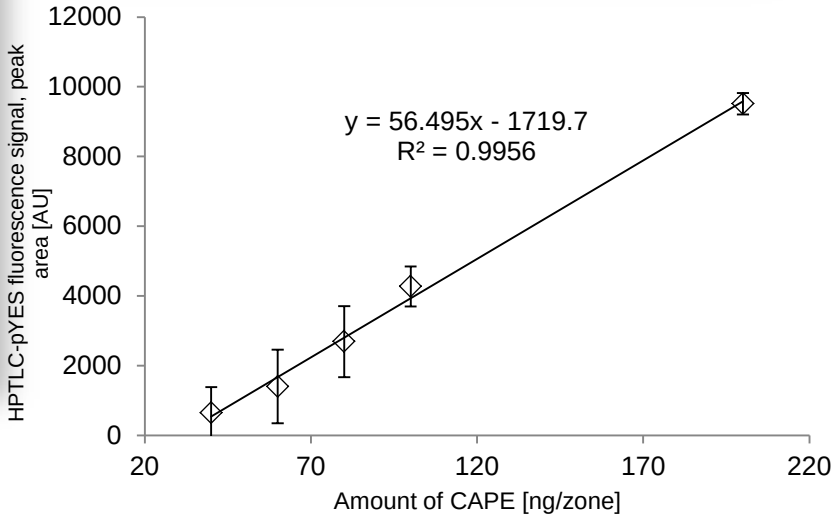
I. Klingelhöfer, G. Morlock, Anal Chem 87 (2015) 11098−11104

Bioquantitation of CAPE in propolis

Propolis sample	CAPE content in sample [µg/mL]	CAPE content [µg/g] referred to propolis weight (n=2)
P1 (30 %)	481	2028
P2 (30 %)	476	2009
P3 (25 %)	471	2387
P4 (62 %)	348	710
P5 (not specified)	380	380[3]
P6 (250 mg/capsule)	359[1]	1435
P7 (30 mg/lozenge)	22[2]	1089

[1]µg/capsule, [2]µg/pastille, [3]µg/mL

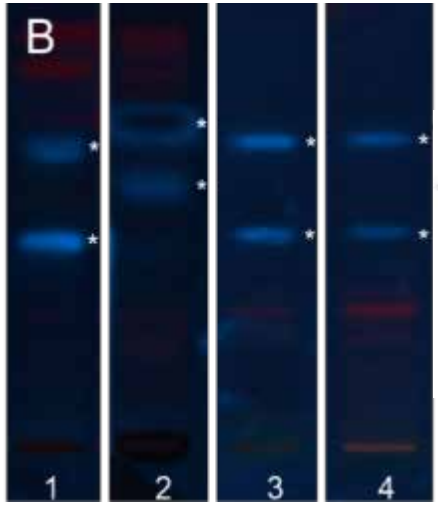




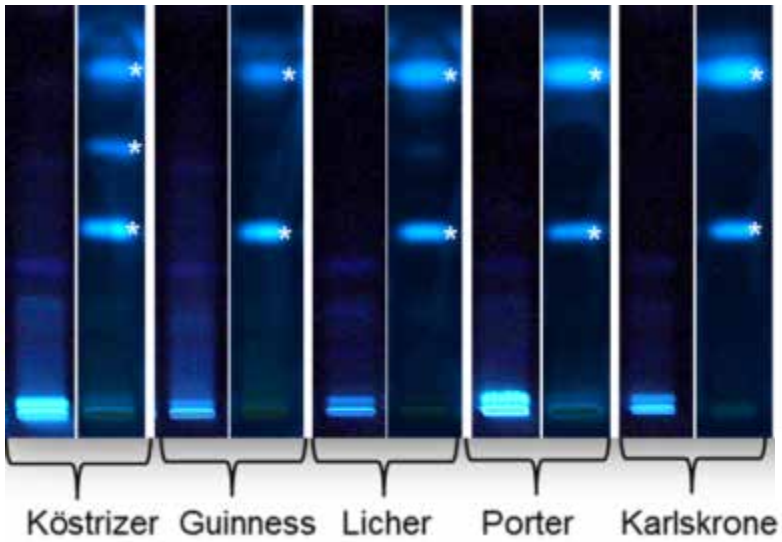
G. Morlock, I. Klingelhöfer, Anal Chem 86 (2014) 8289–8295

Biological detection of estrogen-effective comp.

→ Spices



→ Beer samples

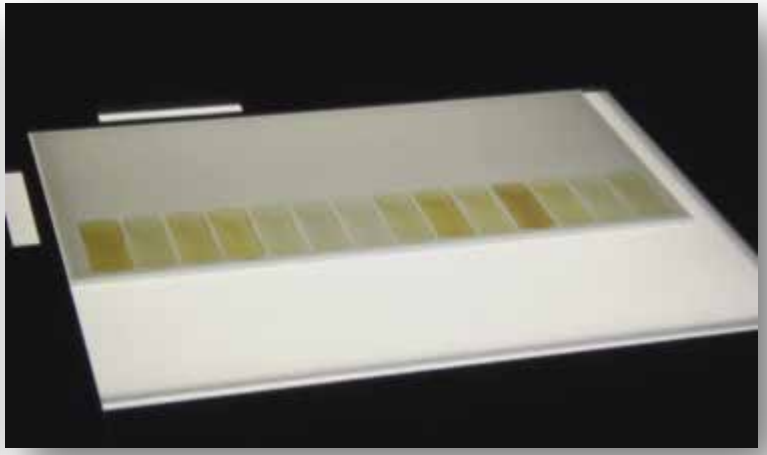


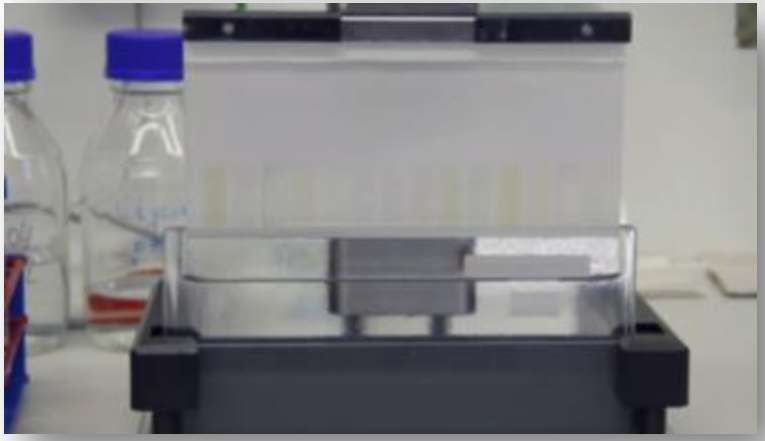
I. Klingelhöfer, G. Morlock, J Chromatogr A 1360 (2014) 288-295

Video HPTLC-EDA

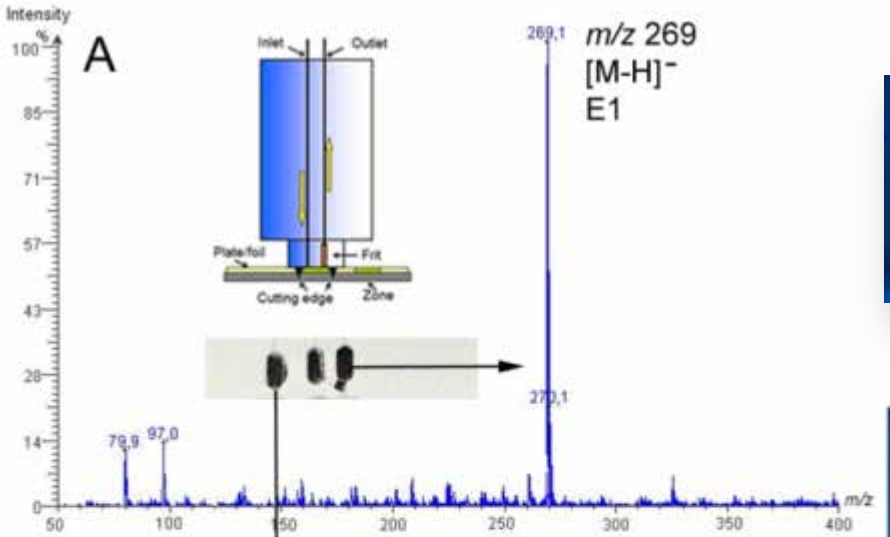
How to discover estrogen-effective compounds in beer?

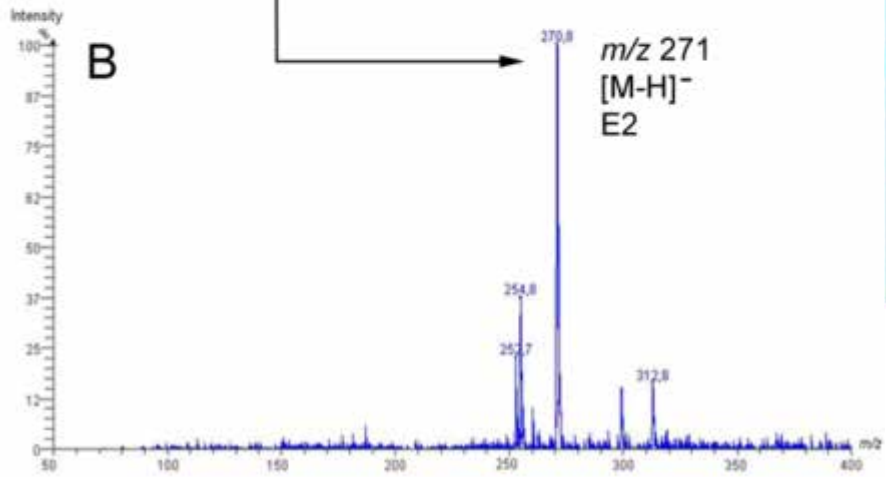
https://youtu.be/Q7AGuljcFvQ

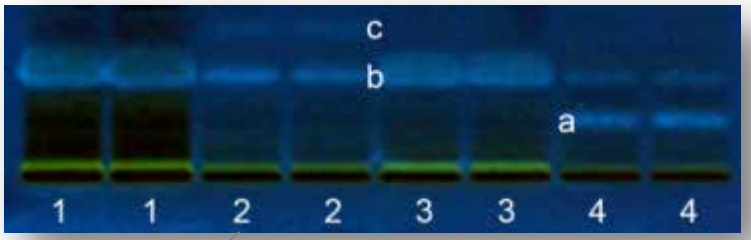


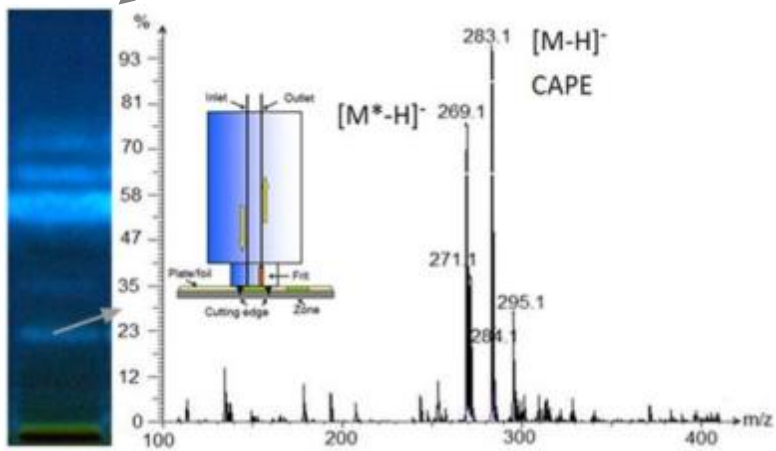


Confirmation by MS





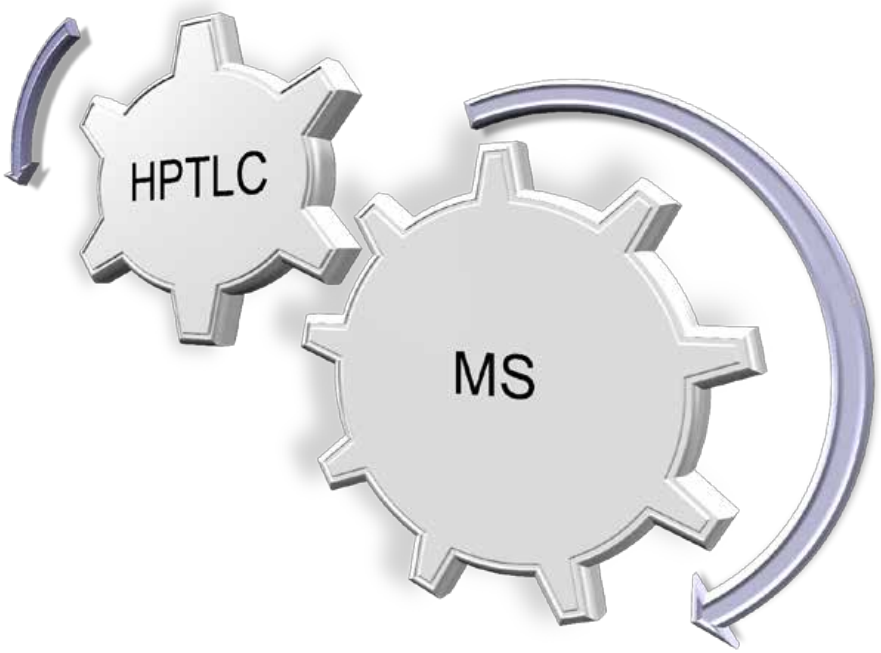




I. Klingelhöfer, G. Morlock, J Chromatogr A 1360 (2014) 288-295
G. Morlock, I. Klingelhöfer, Anal Chem 86 (2014) 8289–8295

HPTLC-MS

Overview, quantitation and comparison



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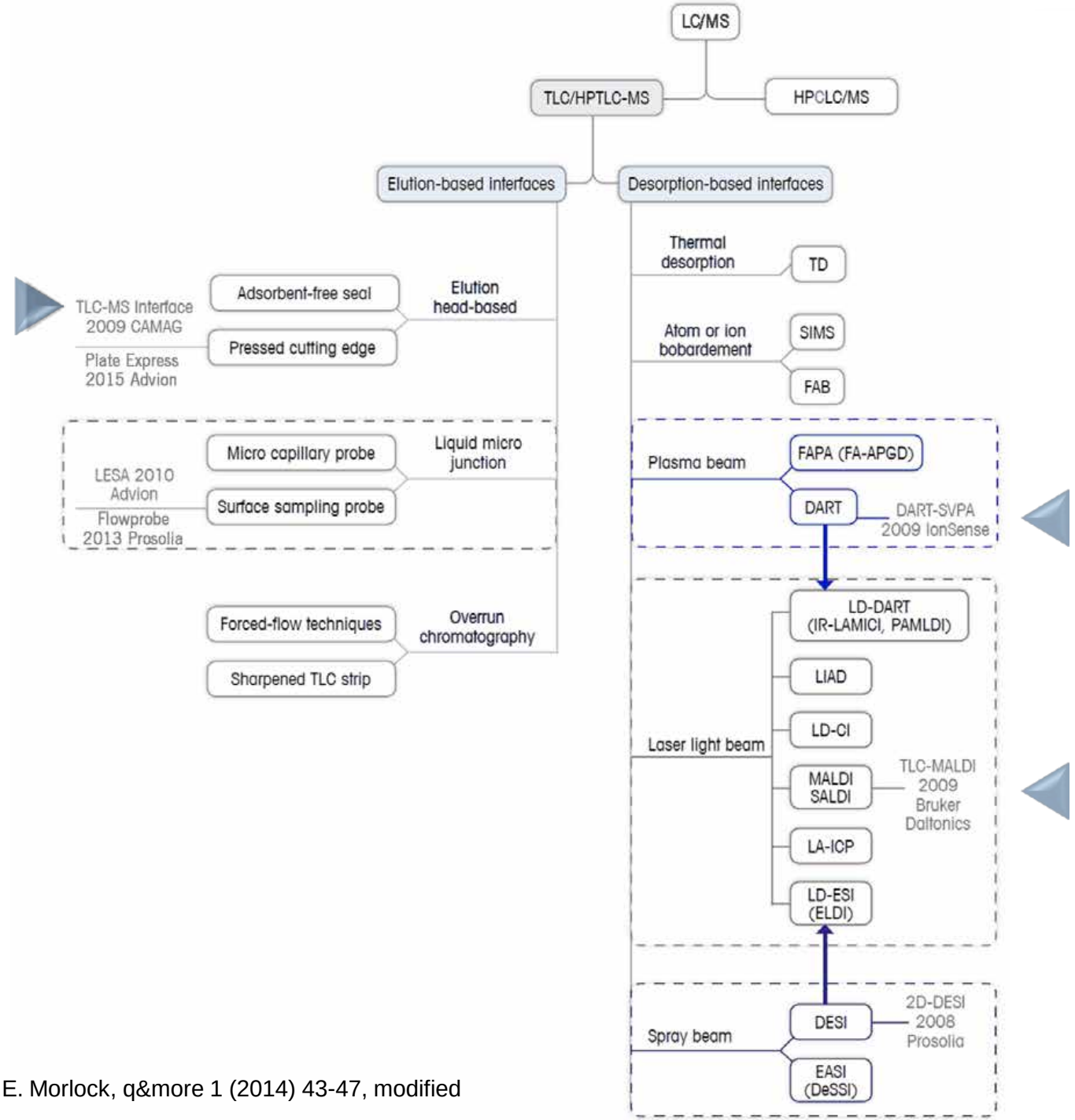


G.E. Morlock, q&more 1 (2014) 43-47, modified

HPTLC-MS

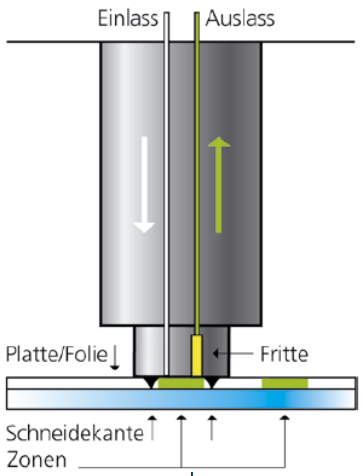
Can we just compare figures

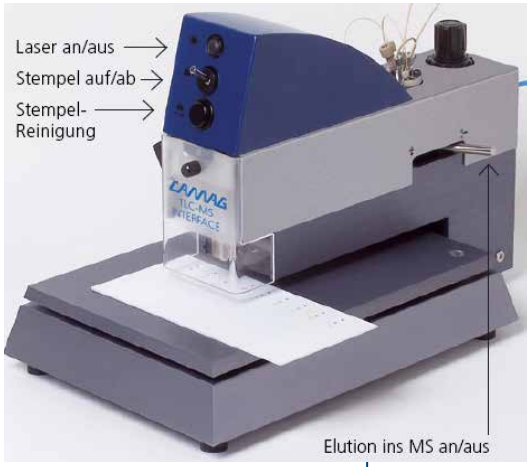
like limit of detection (LOD)?

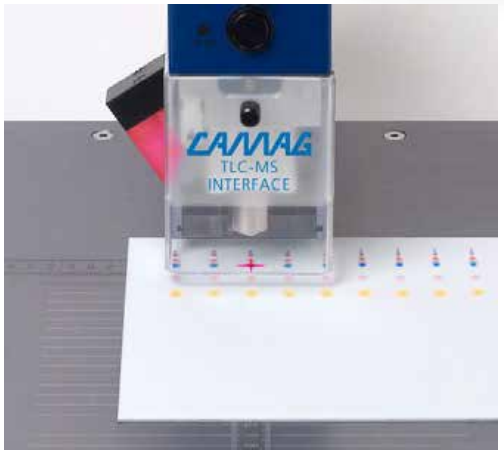
LOD 100 pg/band?

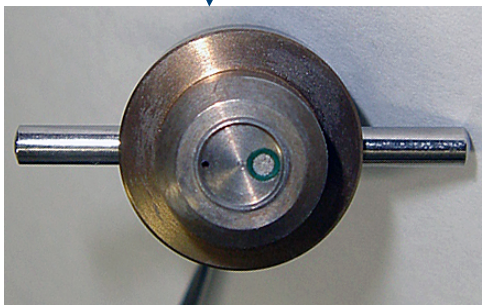
On what does LOD depend on?

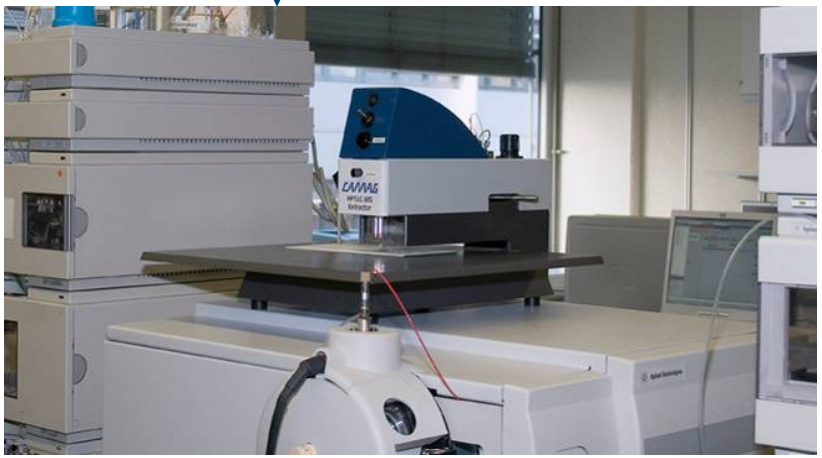
Elution head-based HPTLC-MS









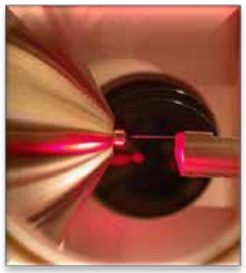


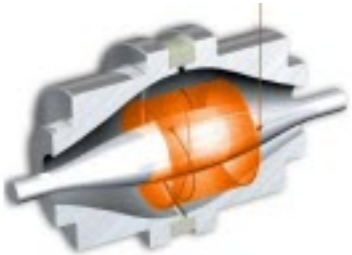
H. Luftmann, Anal Bioanal Chem 378 (2004) 964-968

A. Alpmann, G. Morlock, Anal Bioanal Chem 386 (2006) 1543-1551

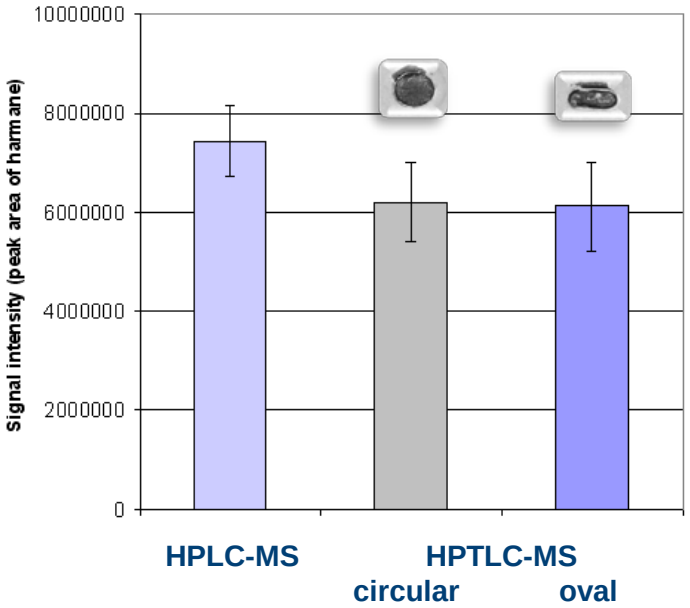
Analyzer type of the mass spectrometer

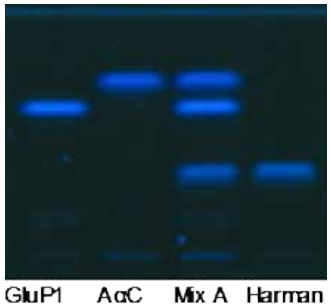
Ion source	Analyzer	Detector
• Electrospray ionization (ESI) • Atmospheric pressure chemical ionization (APCI) • Atmospheric pressure photoionisation (APPI)	• Time-of-flight MS (TOF) • Orbitrap • Ion trap • Single quadrupole • Tandem MS (2 quadrupoles)	• Micro-channel plate (MCP) • Photo multiplier (PM)



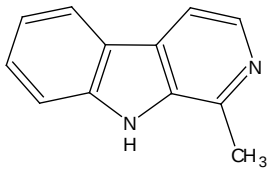


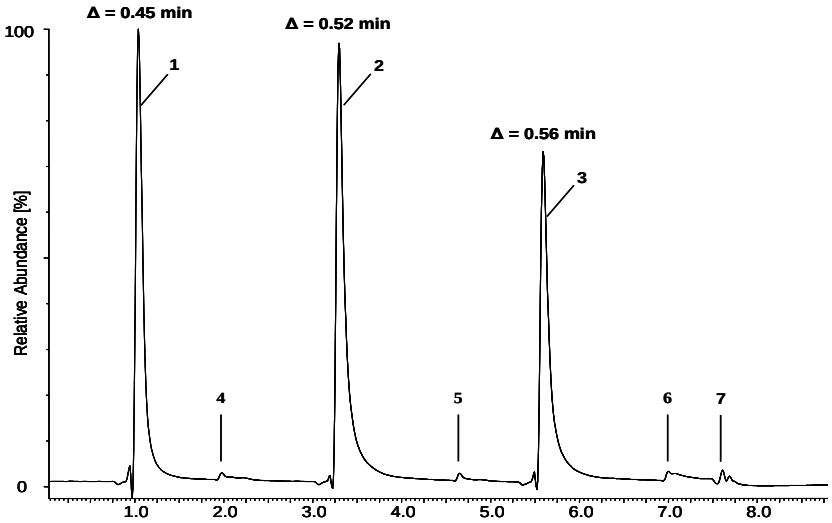
HPTLC-MS *versus* HPLC-MS





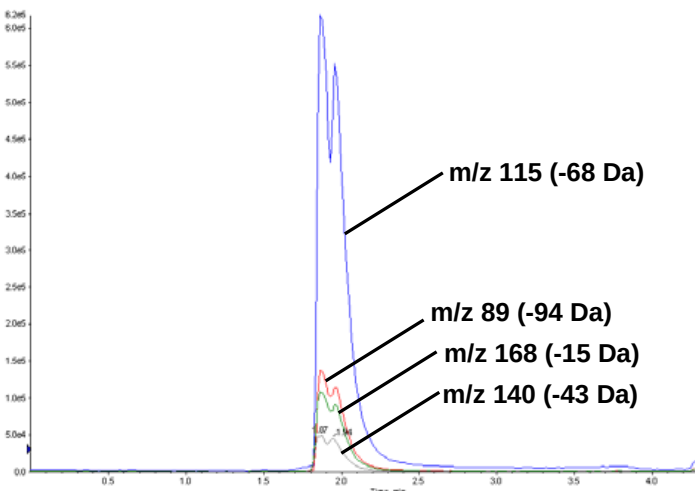
4 ng/zone harmane

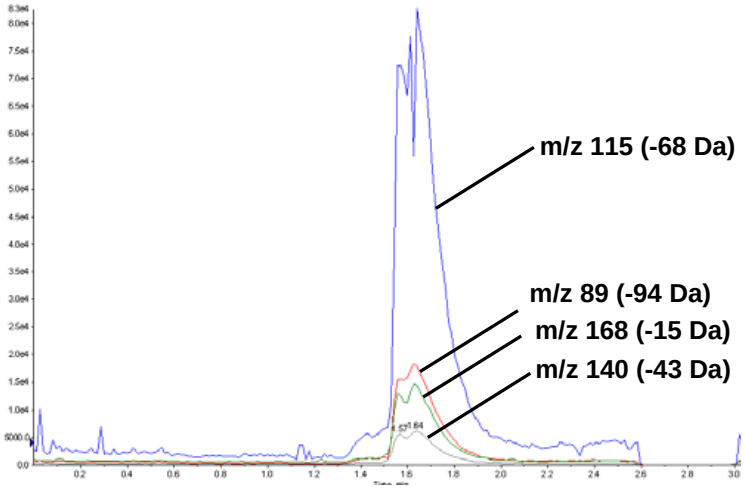




U. Jautz, G. Morlock, J Chromatogr A 58 (2006) 244-250

Detectability by HPTLC-ESI-MS/MS





200 pg/zone harman → 20 pg/zone harman

→ LOQ better than 20 pg/zone harman (S/N 20)

→ Detectability comparable to HPLC/MS

U. Jautz, G. Morlock, J Chromatogr A 58 (2006) 244-250

HPTLC-HRMS (Q Exactive plus)

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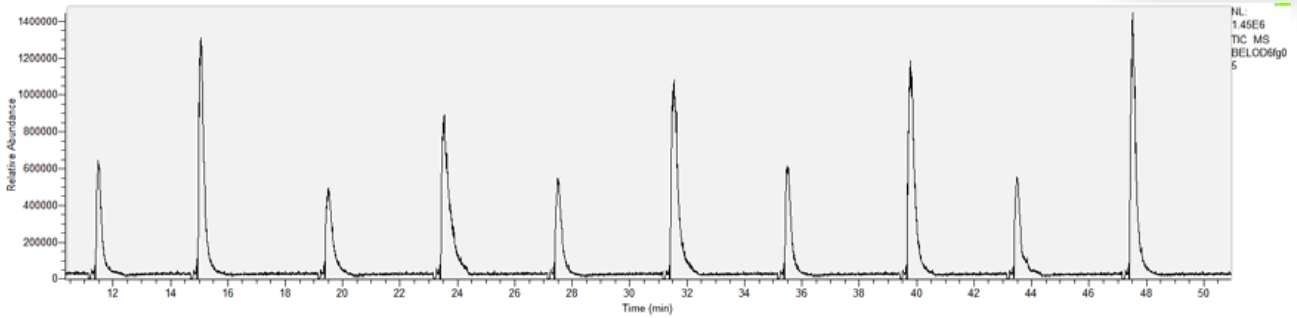
Food Science

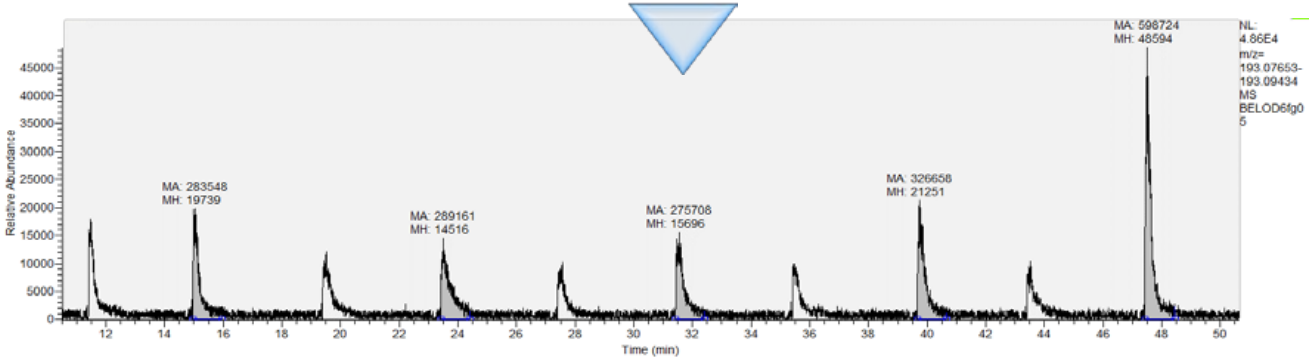
SIM

m/z 193.0861
± *m/z* 1.0



EIC

193.076
- 193.096



LOD of 24 fg/band
for butyl paraben

If 100 µL sample
volume applied:
→ LOD of 0.24 ng/L