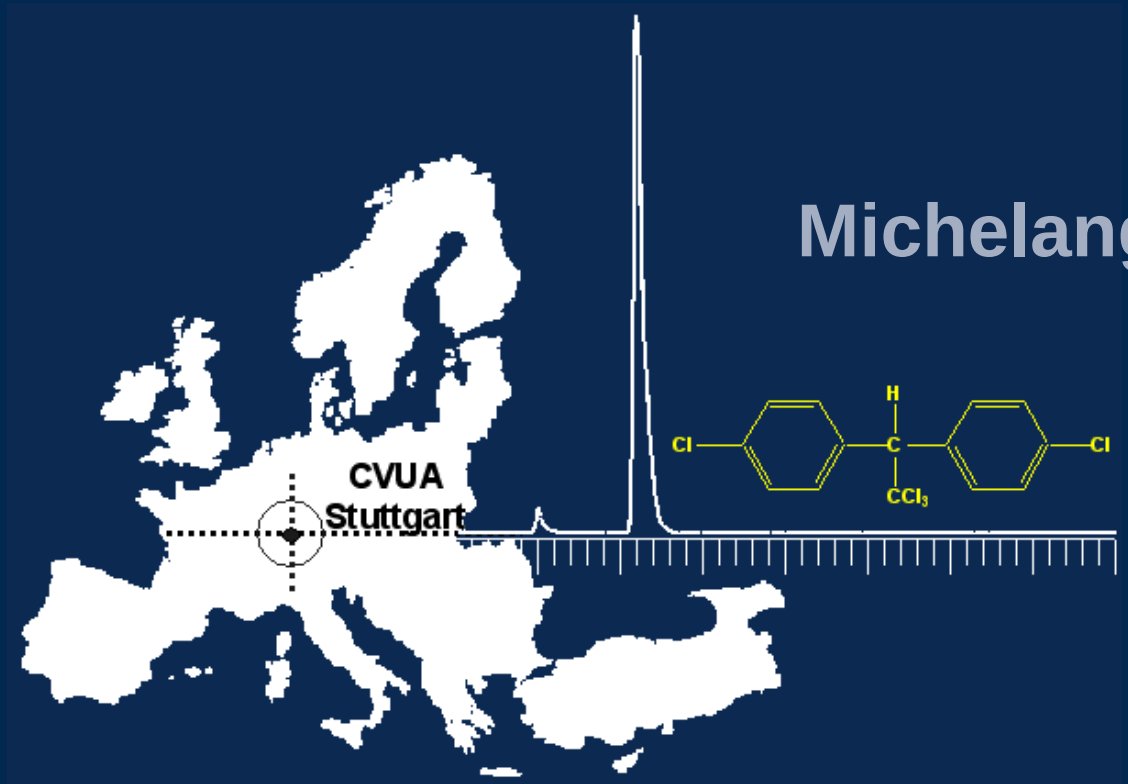
The QuEChERS Method – Background Information and Recent Developments



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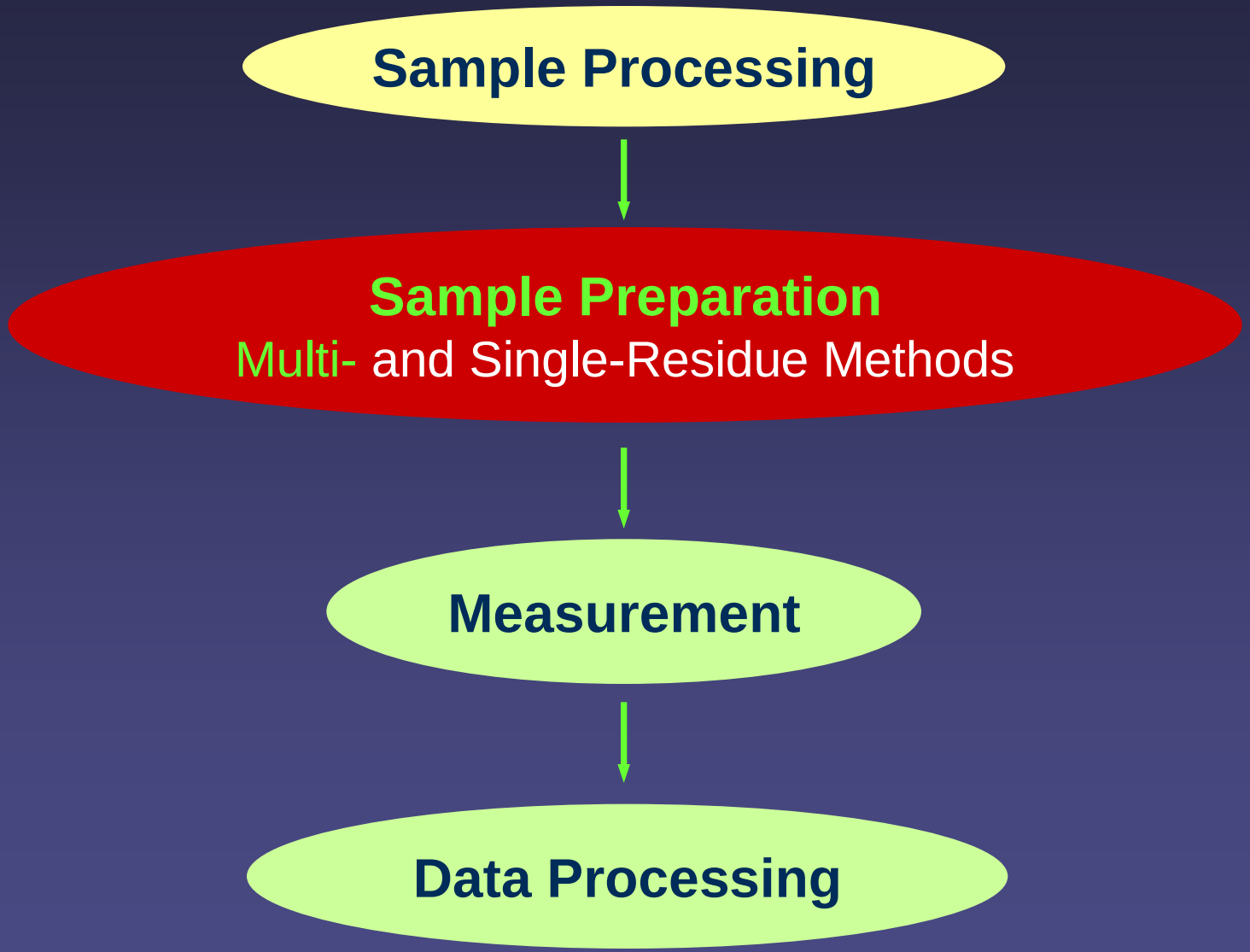
Outlook

- Classical Multiresidue Methods (MRMs)
  - Evolution
  - Limitations and Expectations
- Original QuEChERS-Method
  - Strategy of Method Development (Background Info)
- Recent Developments in QuEChERS Methodology
  - pH-Adjustment (during extraction, in final extracts)
  - Improved Selectivity (extraction, cleanup)
  - Expanded Matrix Spectrum (dry food, fatty food)
- Experiences of its Implementation in the Lab
- Method Validation
  - EU-Proficiency Tests (incurred and fortified residues)
  - Inter-laboratory Ring Tests

Pesticide Residue Analysis:



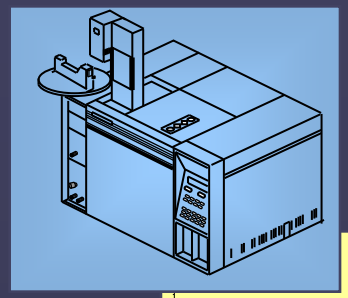
Multiresidue Methods (MRMs):

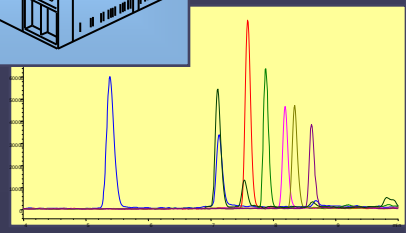
Aim of MRMs:

Cover **as many pesticides as possible** from a single sample portion employing a single sample preparation procedure

But, still

more than one determinative analysis run is required to cover all analytes of interest with sufficient selectivity and sensitivity…





The broader the spectrum of analytes covered by the MRM,

- ✓ **The less additional methods** are required to cover all analytes
- ✓ T**he more efficient and economical** the analysis
  - ➢ Less time, personnel, materials...

MRM Evolution...

Early

Simple but of narrow scope (OCs)

Intermediate

Expanded scope (to cover polar OPs)

Very complex since determ. analysis instr. of poor selectivity and specificity

Novel

Simplicity, streamlining, cost reduction miniaturization, automation

1960 1970 1980 1990 2000 2010

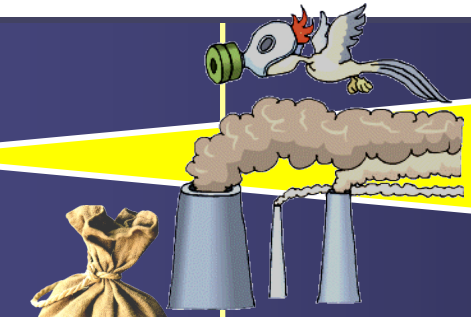
MRM Evolution:

"Technical Development always follows the way from the Primitive via the Complicated to the Simple …"

Antoine de Saint-Exupéry (1900-1944)

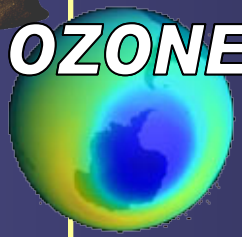
Factors that pushed the Developm. of New Approaches

Environmental, Health-Related and Economic Factors



Need to Assess Risks for Humans and Environment

Need to Improve Productivity and Sample Throughput & to Reduce TAT and Costs



Need to Reduce Solvent Consumpt.

Advancements in Instrumentation (Electronics, Robotics, IT...)

GC/ITD, MSD, MS-MS, TOF, PTV

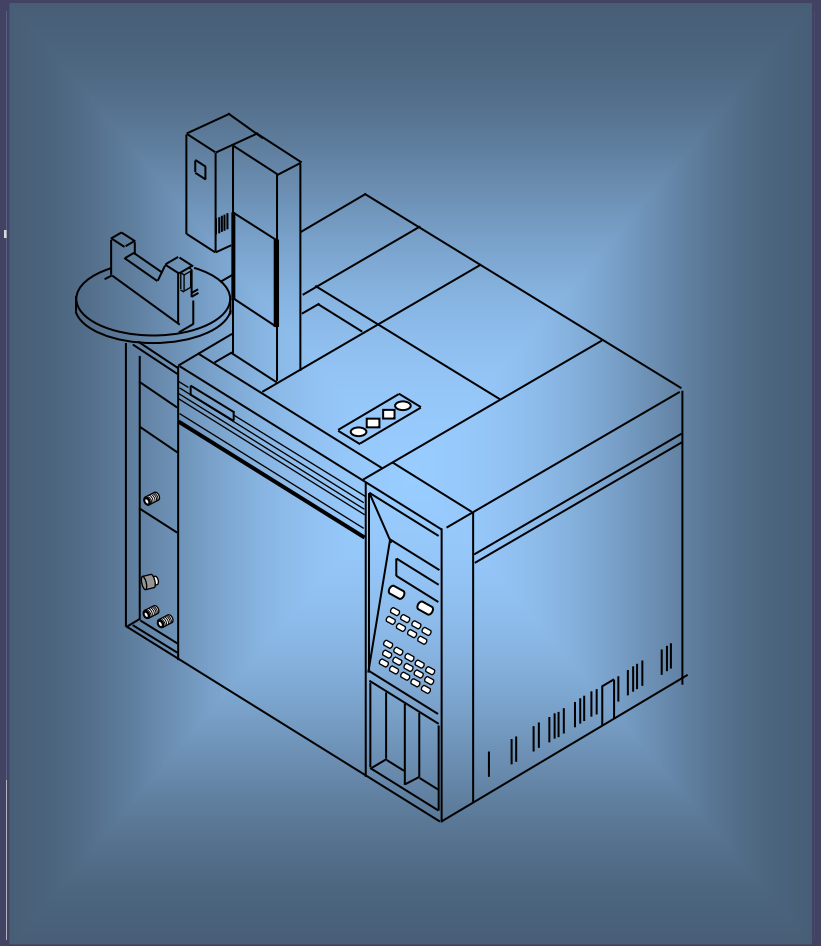
LC/MS, MS-MS, TOF

„Selective" detectors ECD, NPD, FPD

1960 1970 1980 1990 2000 2010

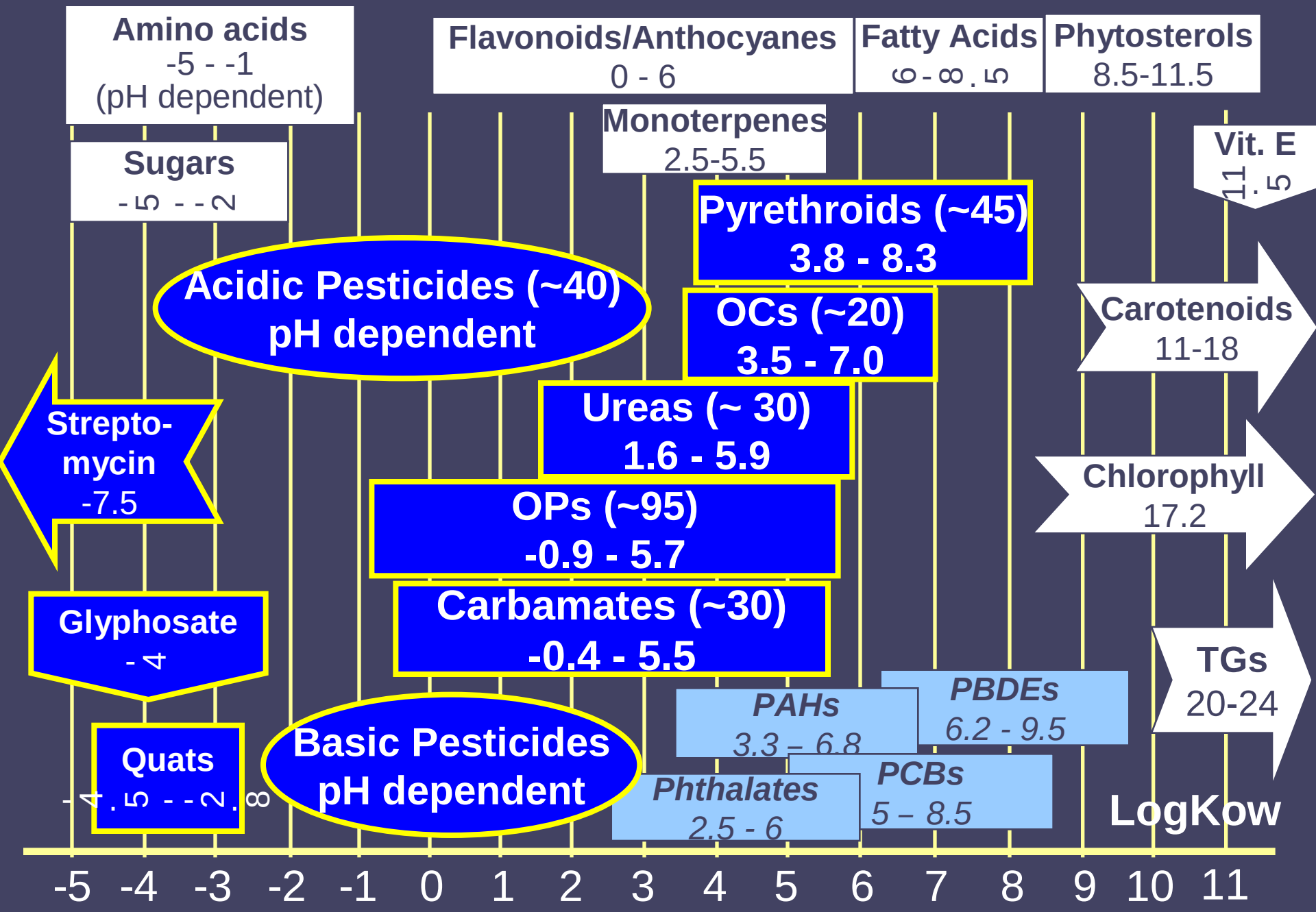
Typical Classical MRM

- Weigh sample (e.g. 50 g)
- Add acetone and blend
- Filter by suction
- Add non-polar solvent (and salts), perform (multiple) partitioning
- Dry and filter organic phase
- Evaporate
- Reconstitute, perform GPC cleanup
- Evaporate
- Perform fractionated cleanup on silica
- Evaporate
- Transfer fractions in GC-Vials

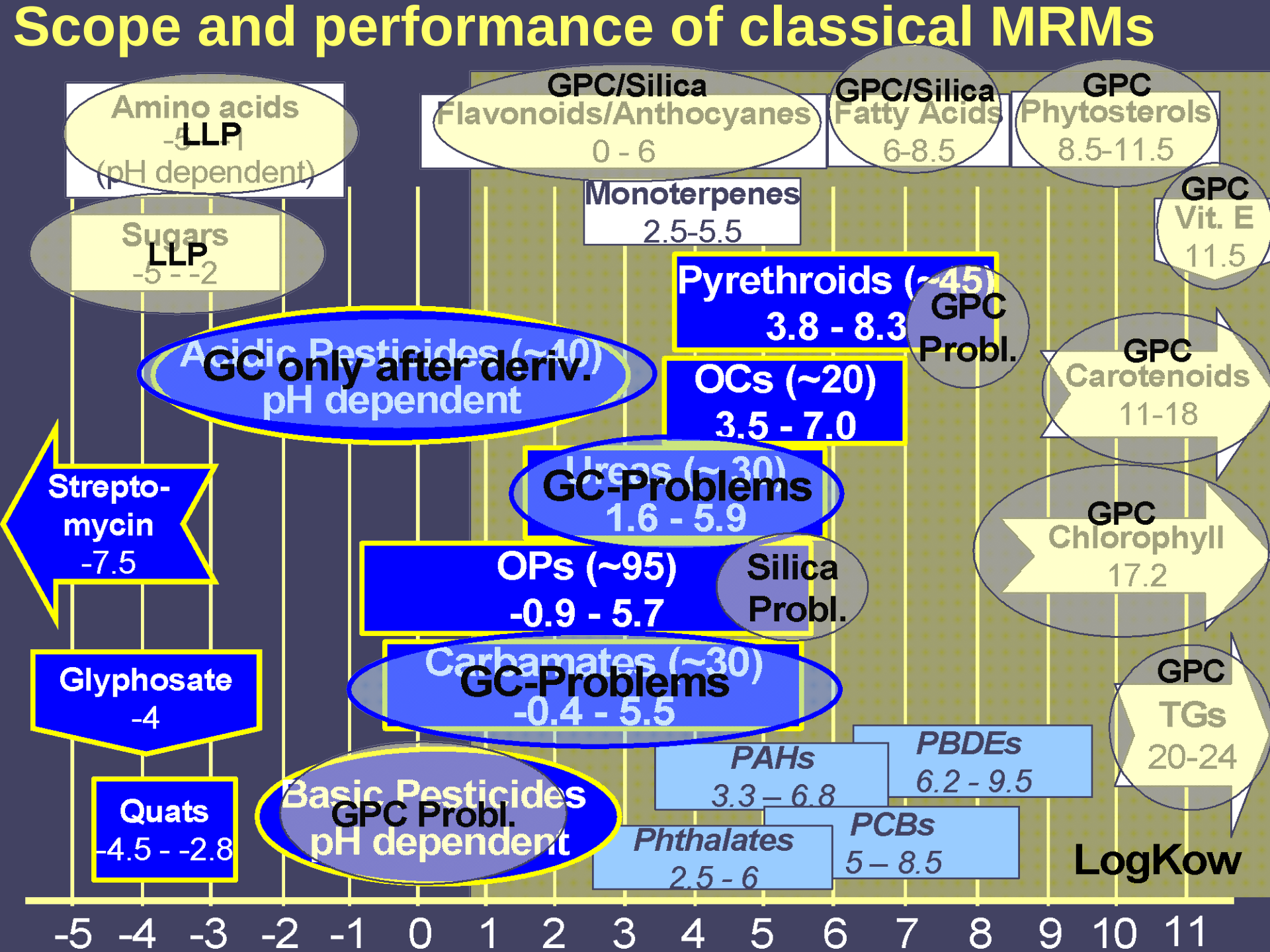


Analysis
by GC-ECD, FPD, NPD

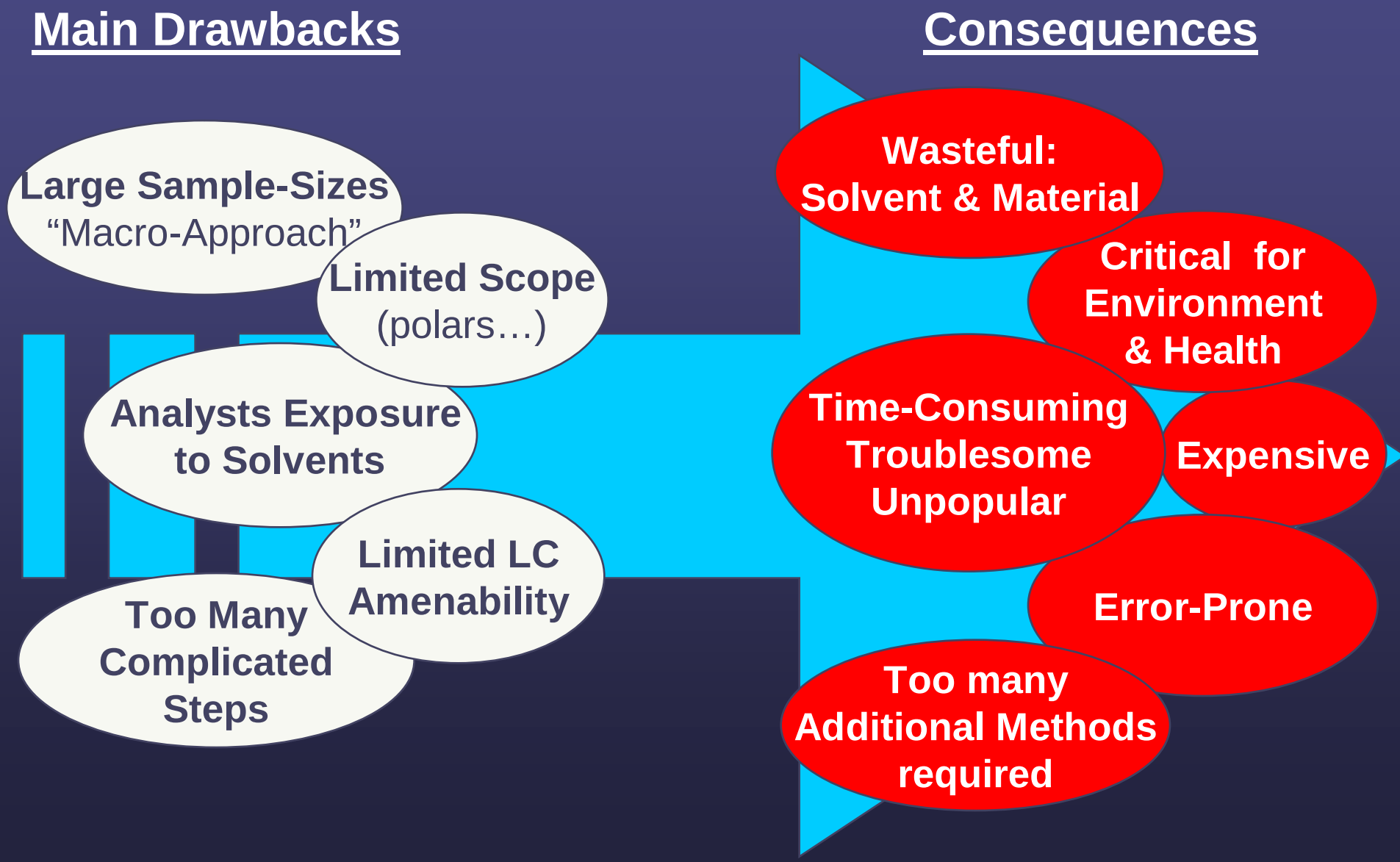
Pesticides and Co-extractives...



Scope and performance of classical MRMs



Typical inefficiencies of classical MRMs



Sample Processing

Sample Preparation
has traditionally been
the Bottleneck
of Pesticide Residue Analysis

Measurement

Data Processing

Desirable Characteristics of MRMs

- Fast (as Few Steps as Possible)
- Easy to Perform
- Inexpensive
- Low Solvent Consumption
- Safe for Personnel and Environment
- Selective
- Rugged and precise
- Achieve Good Recoveries for a Broad Analyte Spectrum
  - Thus Reducing the need to run Single (-Group) Residue Methods

Some Novel Sample Preparation Techniques

- Focusing on Automation
  - SFE
  - PLE
- Focusing on Automation and/or Miniaturization
  - SPME/SBSE
  - MSPD
- Focusing at Simplification of Classical Methods
  - SPE of water-diluted extracts
  - Partitioning on Macroporous Sorbents
  - QuEChERS

QuEChERS - Original-Method

Weigh 10 g of Sample (50 mL Teflon-Tube)

Add 10 mL Acetonitrile

Shake Vigorously 1 min

Add 4 g $MgSO_4$ and 1 g NaCl

Shake Vigorously 1 min

Add ISTD-Solution

Shake 30 s and Centrifuge

Take Aliquot and Add $MgSO_4$ and Sorbent(s)

Shake 30 s and Centrifuge

(Add "Analyte Protectants", adjust pH)

GC-MS (and LC-MS)

Anastassiades et al.

JAOAC Int. 86 (2003) 412-431

Procedure in Pictures – 1. Initial Extraction Step

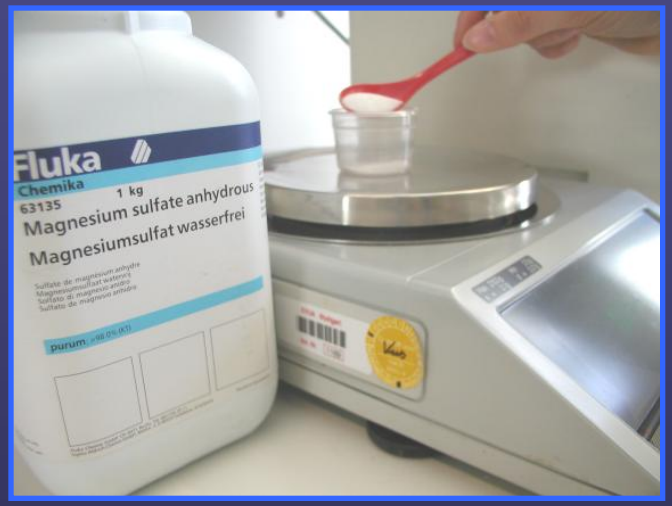
Weigh 10 g Sample

Add 10 mL MeCN

Shake Intensively

for 1 min

Procedure in Pictures – 2. Extraction/Partitioning Step



(Pre-)Weigh

4 g $MgSO_4$ + 1 g NaCl

Add to the Tube

Shake Intensively

for 1 min

Procedure in Pictures – 3. Addition of ISTD and Centrifugation

Add ISTD

Shake for 30 s

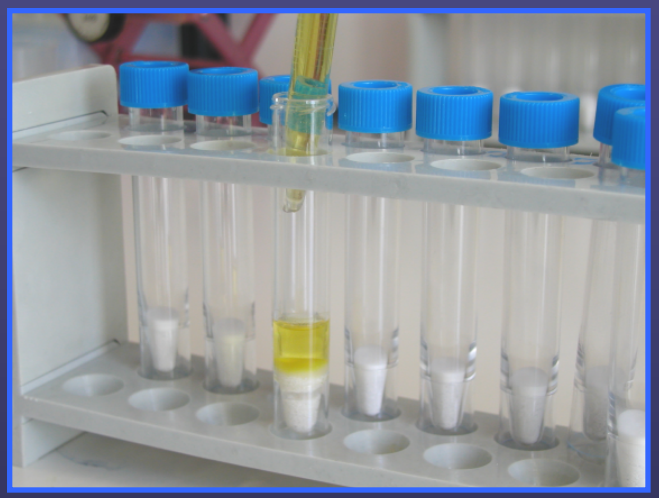
Centrifuge (ca. 5 min)

Separated Raw Extract

Procedure in Pictures – 4. Dispersive SPE Step

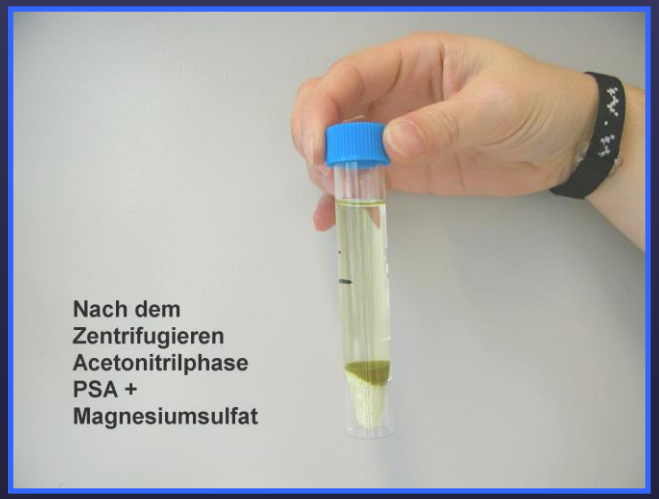
(Pre-) Weigh $MgSO_4$ and PSA

Add Extract to Tube and Shake ca. 30 s



Centrifuge (ca. 2 min)

Cleaned up Extract

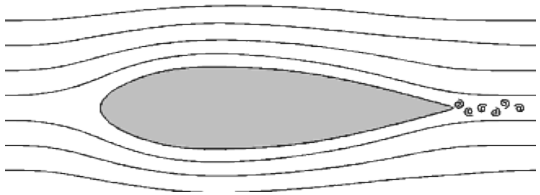


Simplifications Introduced in the Method

Time Consuming, Complicated or Error Prone Steps of traditional MRMs	Simplified Alternatives
Use of Ultra-Turrax during Initial Extraction	Shaking
Filtration	Centrifugation
Multiple LL-Partitioning Steps and Isolation of Entire Extract	Single Partitioning ("On-Line-Approach") Take Aliquots (Use ISTD)
Use of a Lot of Glassware	Extraction/Partitioning in Single Vessel
Evaporation/Reconstitution	Large Volume Injection; Sensitive Instr.
Trad. Cleanup w. Columns (SPE, GPC)	Dispersive SPE
Sample Processing/Homogenization	No Way Around this!!

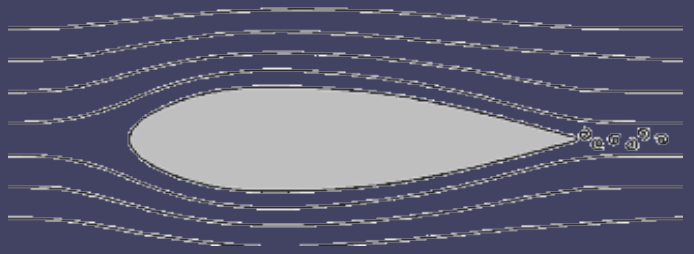
⇨**Goal achieved: Simple and Streamlined MRM**

- **Few working steps,**
- **Convenient to perform**
- **Low Material- and Solvent consumption**



Strategies in the Development of the QuEChERS-Method

STREAMLINED AND SIMPLE...



ECONOMICAL...

FIT FOR PURPOSE...

Method Development - Aspects Considered

- **Initial Extraction & Extraction/Partitioning Step**
  - Choice of Extraction Solvent and Sample/Solvent Ratio
  - Sample Amount
  - Blending Vs. Shaking (Incurred Residues)
  - Influence of Sample pH on Recov. (Ionization, Degradation)
  - Type and Amount of Salts Used to Induce Phase Separation
  - Selectivity (Gravimetric Anal. of Extracts, GC-Interferences)
  - Use of ISTD (Check that Recovery-Correction is minimal)

- **Cleanup (Dispersive SPE)**
  - Type and Amount of Sorbent and $MgSO_4$
  - Selectivity (Gravim. Anal. of Extracts, GC-Interferences)

- **Instrumental Analysis**
  - Matrix Effects (Influence of Cleanup)
  - Use of “Analyte Protectants”

Method Development –
Choice of Acetonitrile as Solvent

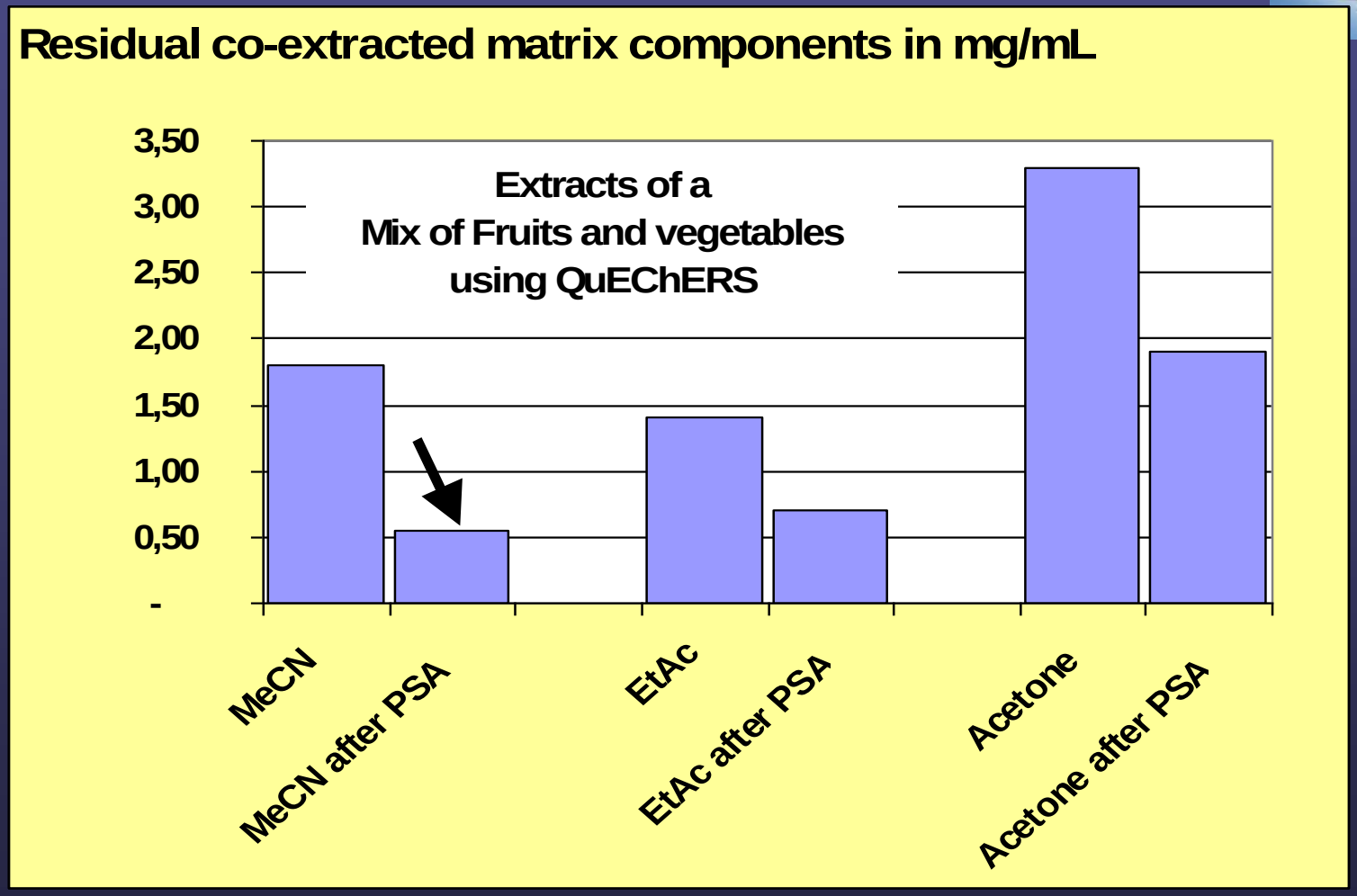
- ☑ Selective (Few Co-Extractives but still broad pesticide Spectrum covered)
- ☑ Compatible with LC- and SPE-Applications
- ☑ Not Chlorinated
- ☑ Miscible with Water (Good for Initial Extraction)
- ☑ Separ. from Water-Phase by Salt-Add. (No Non-Polar Solv. Needed)
- ☑ Easier to Remove Water (with $MgSO_4$) than from Acetone

- ☒ Difficult to Evaporate
- ☒ High Expansion Volume
- ☒ Not Compatible With NPD

But PTV with Solvent Venting could be used

- ☒ Not Compatible with GPC ***(But, Lipid-Co-Extraction is Low)***
- ☒ Low Lipid Solubility
  - ➢ *Losses of non-polar pesticides* ***(Recov. consistent at same Lipid/solvent ratio)***
  - ➢ *Accessibility problems of pesticides enclosed in Lipid particles* ***(Ultra Turrax)***
- ☒ Rel. Toxic ***(But, Method Performed in a Closed Vessel, thus minimal exposure)***

Method Development –
Acetonitrile vs. other Solvents



For More Details and Comparison with EtAc , Acetone *(see AOAC publ.)*

Method Development –
Sample Amount and Sample/Solvent Ratio

Sample Amount: 10 g

Miniaturization improves efficiency

- Less material consumption
- Reduced costs

Important:

- Good homogenization is needed (e.g. use of dry ice)

Studies: Acceptable variations for 10 g subsamples using cryogenic milling

Sample:Solvent Ratio: 1:1 (w/vol)

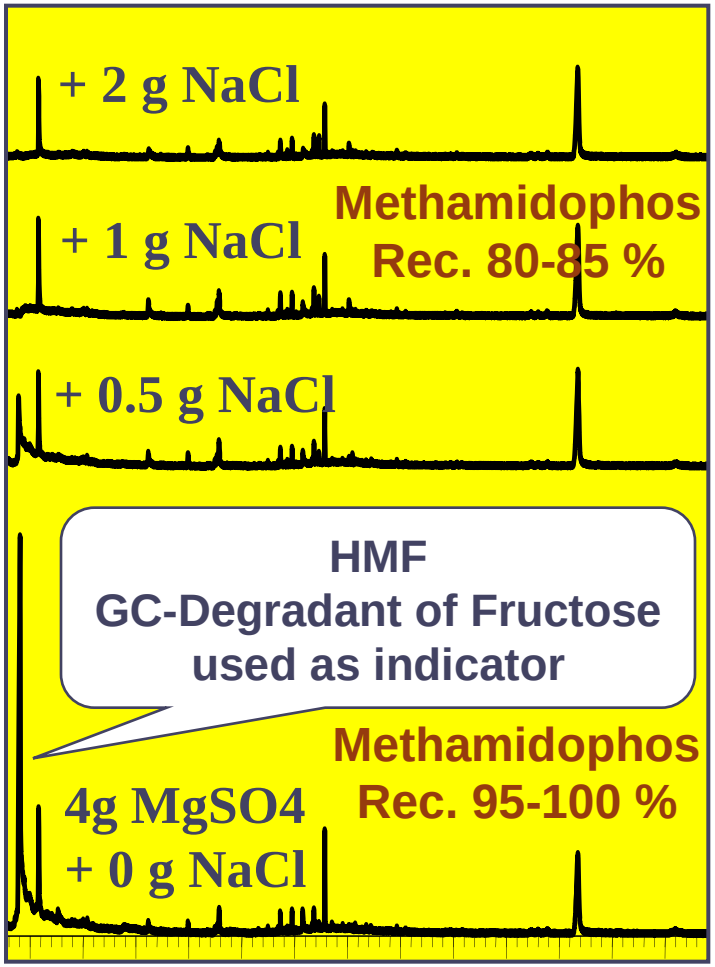
⏩ **gives 1g/mL**

- Still good recoveries of incurred residues (polar and non-polar)
- No evaporation of final extract necessary…
  - 1g/mL is enough when using modern instruments (PTV in GC is better)

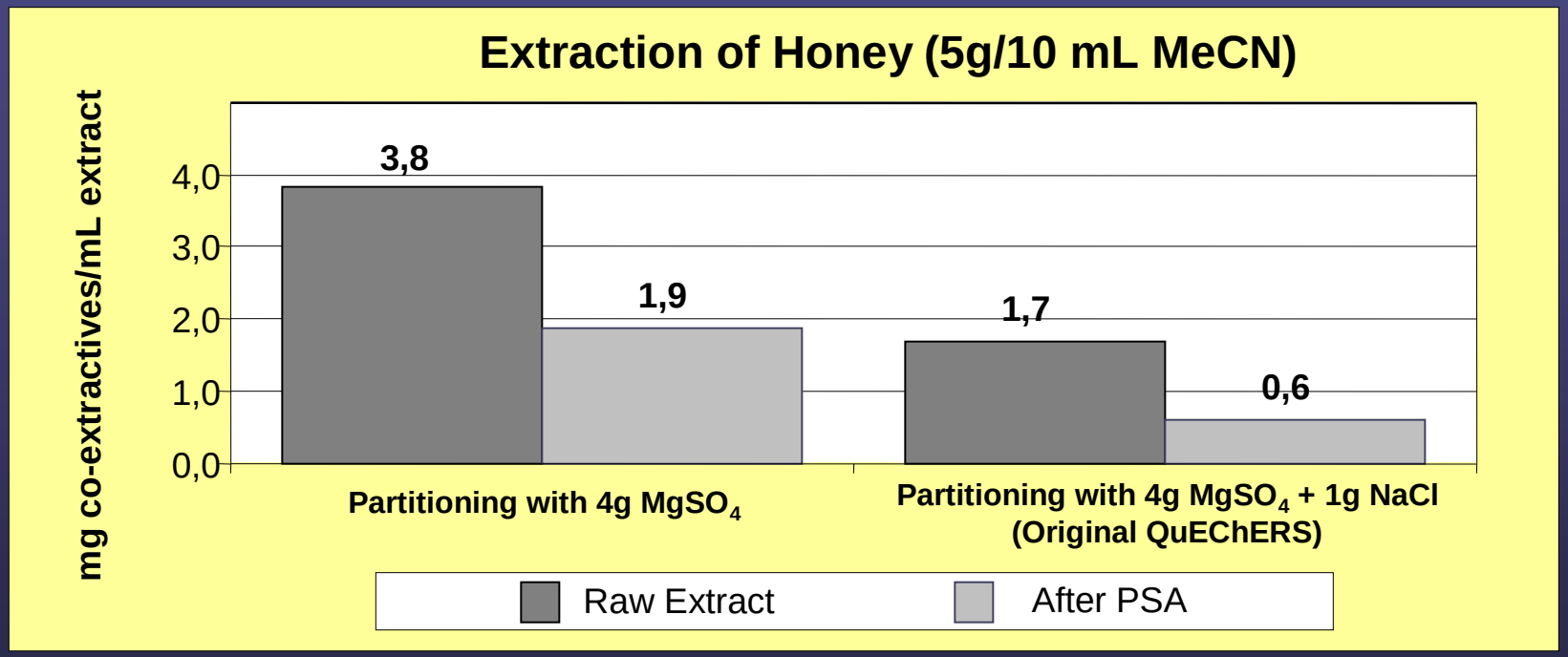
Method Development - $MgSO_4$ / NaCl 4:1 for the Partitioning Step

- Many Salts tested
- $MgSO_4$ gave best salting-out of ACN and **Best Overall Recoveries** (especially for polar pesticides)
- However: too much water in ACN-phase and **too many Polar Co-extractives** (e.g. Sugars)
- **NaCl Addition increases Selectivity**
  Less Water (and Sugar) in ACN-Phase

➔NaCl is used to Control Selectivity



Method Development - $MgSO_4$ / NaCl 4:1 for the Partitioning Step



➔NaCl reduces the amount of co-extracted matrix

Method Development – Shaking vs. Blending

Advantages

- ✓ No Exposure to Metal Surfaces
- ✓ Can Be Done by Hand and in Parallel
- ✓ No Cleaning of Jar and Blender Between Samples
- ✓ No Carry Over Between Samples
- ✓ Only One Container necessary
- ✓ Safer (Closed Vessel)
- ✓ Less Noisy than Blending
- ✓ No Frictional Heat

- May be less reproducible that blending

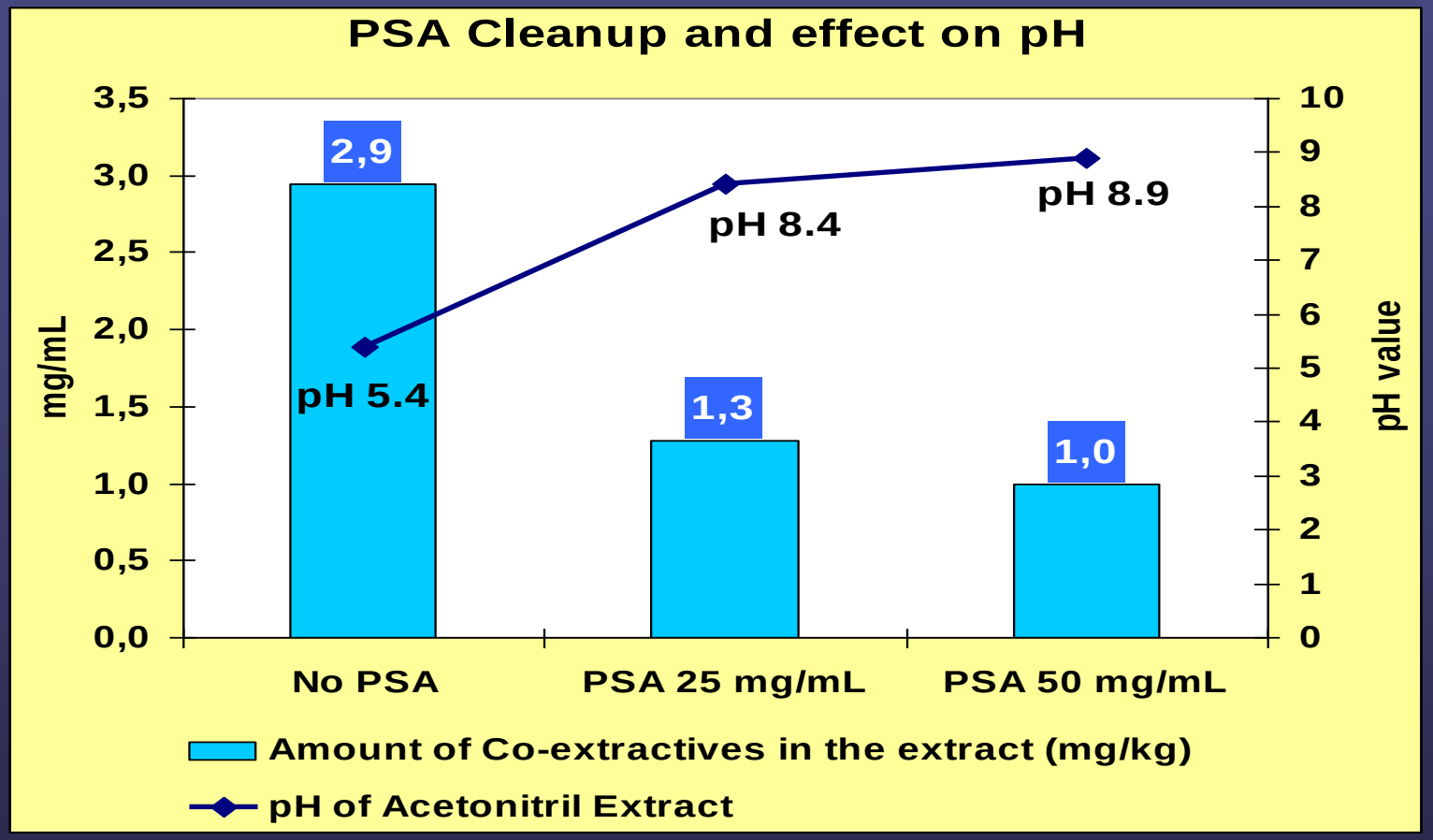
Pesticides from Fruits + Veg.: Ultra-Turrax **usually not necessary**
➔Checked with Incurred Residues (Cryogenic milling)

Method Development - Dispersive SPE for Cleanup

- **Advantages over classical SPE with Cartridges**
  - ☑ No SPE Manifold, Vacuum/Pressure,
  - ☑ No Conditioning,
  - ☑ No problems w. Channeling, Flow Control, Drying-Out,
  - ☑ No Elution Step Needed,
  - ☑ No Add. Vessels for Eluent Collection,
  - ☑ No Dilution of Extract
  - ☑ No Evaporation,
  - ☑ Less Sorbent Needed,
  - ☑ Faster and Cheaper,
  - ☑ No Experience Needed.

When "**Chemical Filtration**" is needed
➔ "**Dispersive SPE**" is a serious option

Dispersive SPE –Removal of Co-extractives



Drawbacks:

- ☒ pH goes up (degradation risk)
- ☒ Matrix-Induced Analyte Protection in GC reduced

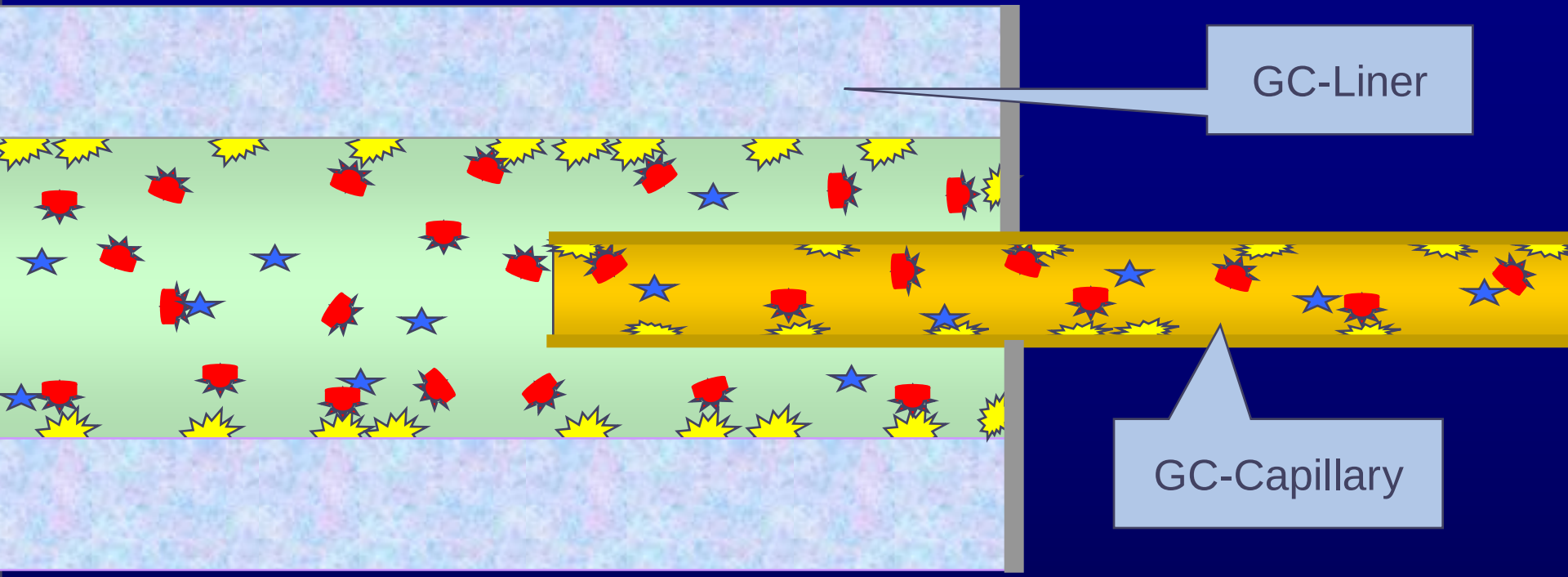
Solutions:

- ➢ Addition of Acids (see later)
- ➢ Addition of Analyte Protectants

Impact of Matrix-Effects

„Matrix-Induced Peak Enhancement“

„Matrix-Induced Peak Enhancement Effect“



Active Sites (on Surface of GC-Liner & Column)
(Siloxanes & deposited non-volatile matrix-co-extractives)

Analytes (interact with Active Sites which causes…)

- Unwanted Retention/Tailing
- Quasi-catalysed degradation (susceptible compounds)

Matrix-Components (in Excess)

- Bloc active sites and protect analytes

Analyte: Atrazine ; Matrix: Strawberry

WITH Matrix co-extractives
(Strawberry-Extract)

WITHOUT Matrix co-extractives
(in pure solvent)
e.g. Calibration standard

- Stronger Tailing
- Apex-Shift towards longer RTs!

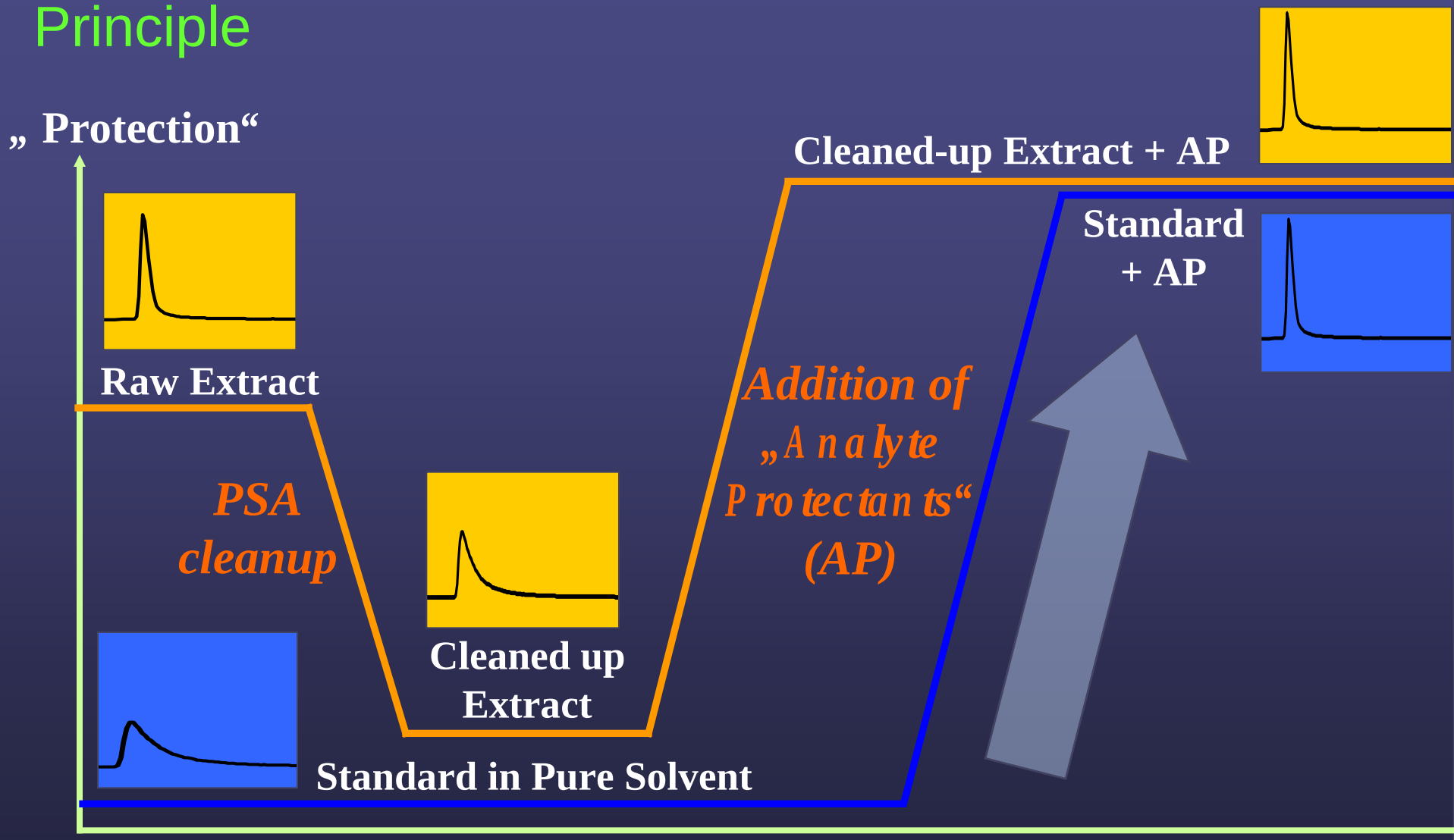
Ratios:

- Peak-Areas: ~ 1,5:1
- Peak-Heights: ~ 4:1
- Peak-Width (at half height): ~ 1:3

Matrix-Induced Peak Enhancement
⇨ OVERESTIMATION OF RESULTS!!

Analyte Protectants

Principle



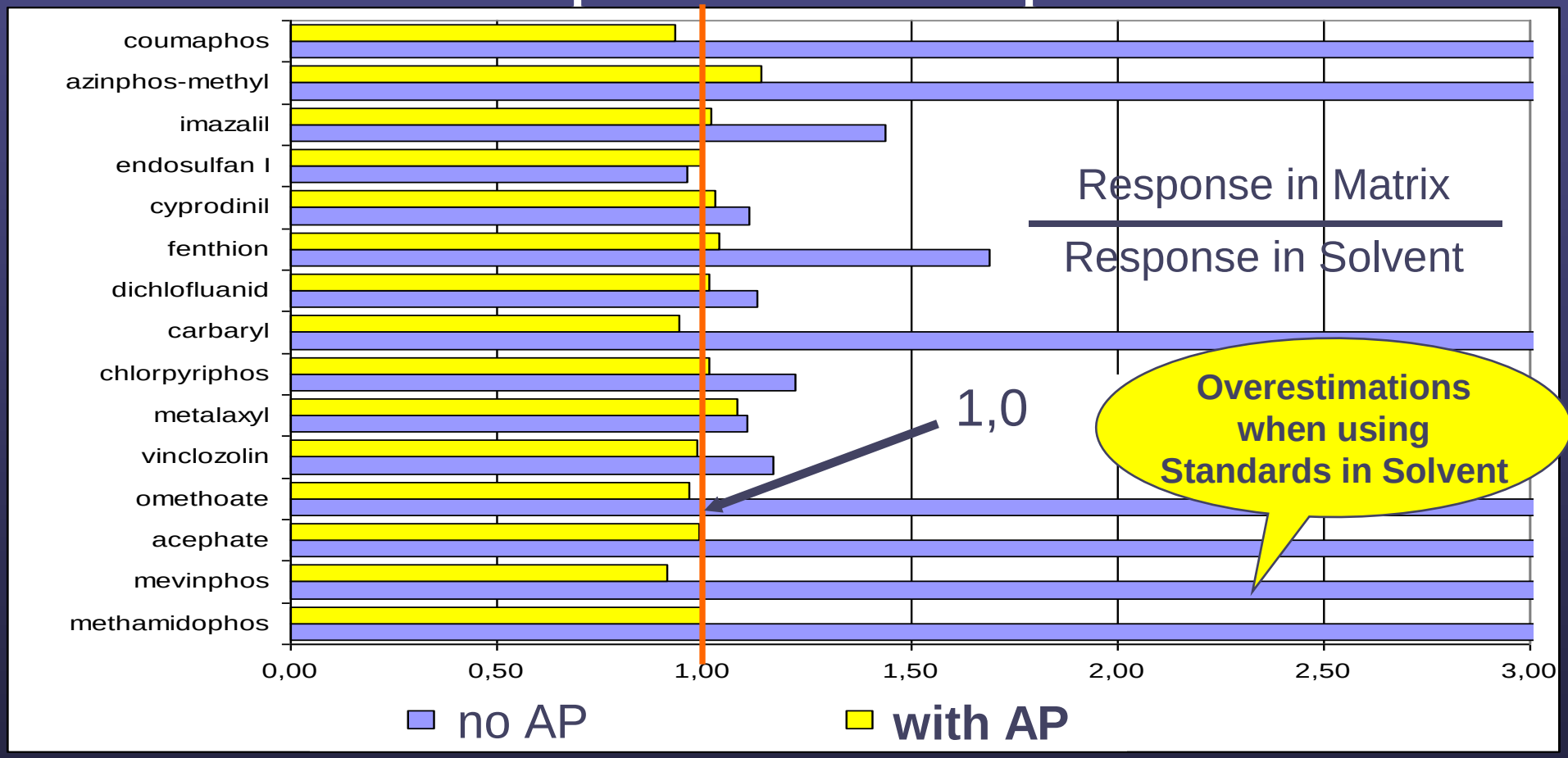
Analyte Protectants Reduce:

Analyte Interactions with Active Sites

and thus **Errors Related to Matrix-Induced Peak Enhancement in GC**

Analyte Protectants- Reduction of Matrix Induced Enhancement Errors

Errors eliminated if: Response in Matrix/Response in Solvent ~ 1

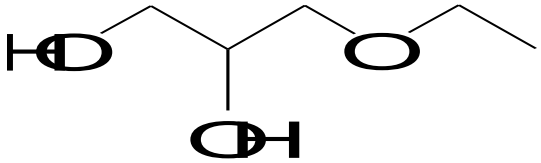


AP was added to both :
Sample Extract and Calibration Standard (in pure Solvent)

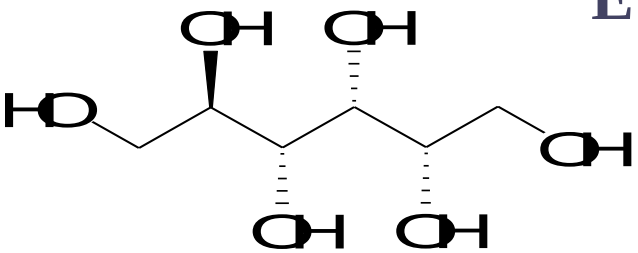
Analyte Protectants – Examples

- Various Compounds Tested for “Protective Potential”.
- **Best Protection** : **Polyhydroxy-Compounds** (sugars, ~derivatives)

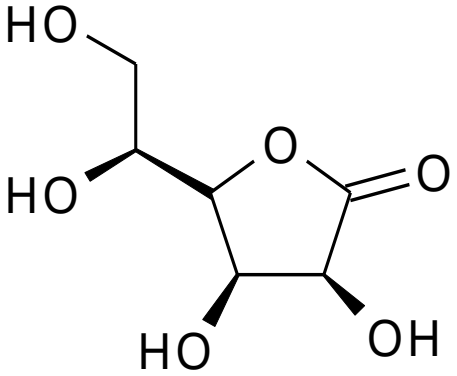
Examples:



Ethylglycerol



Sorbitol

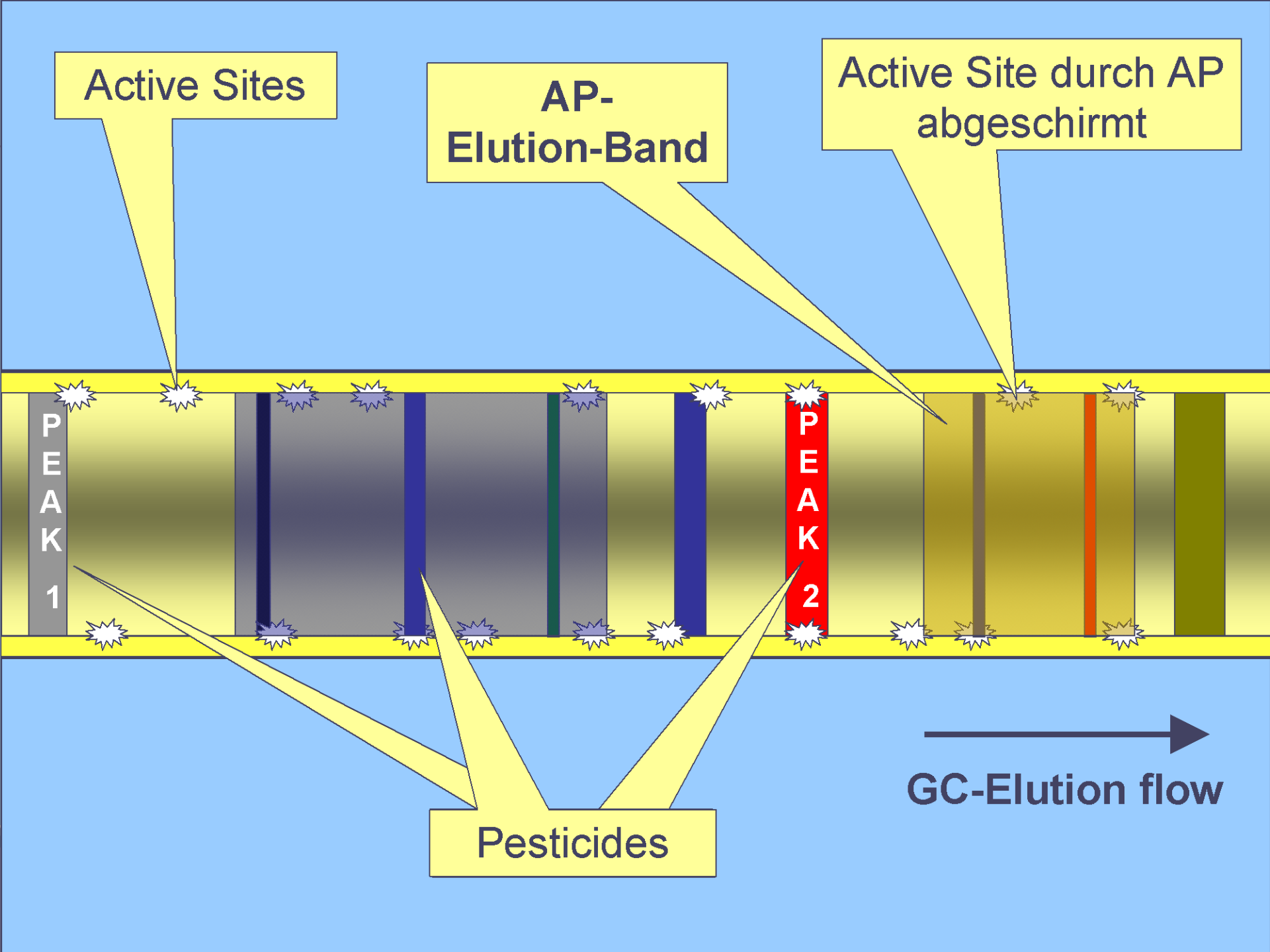


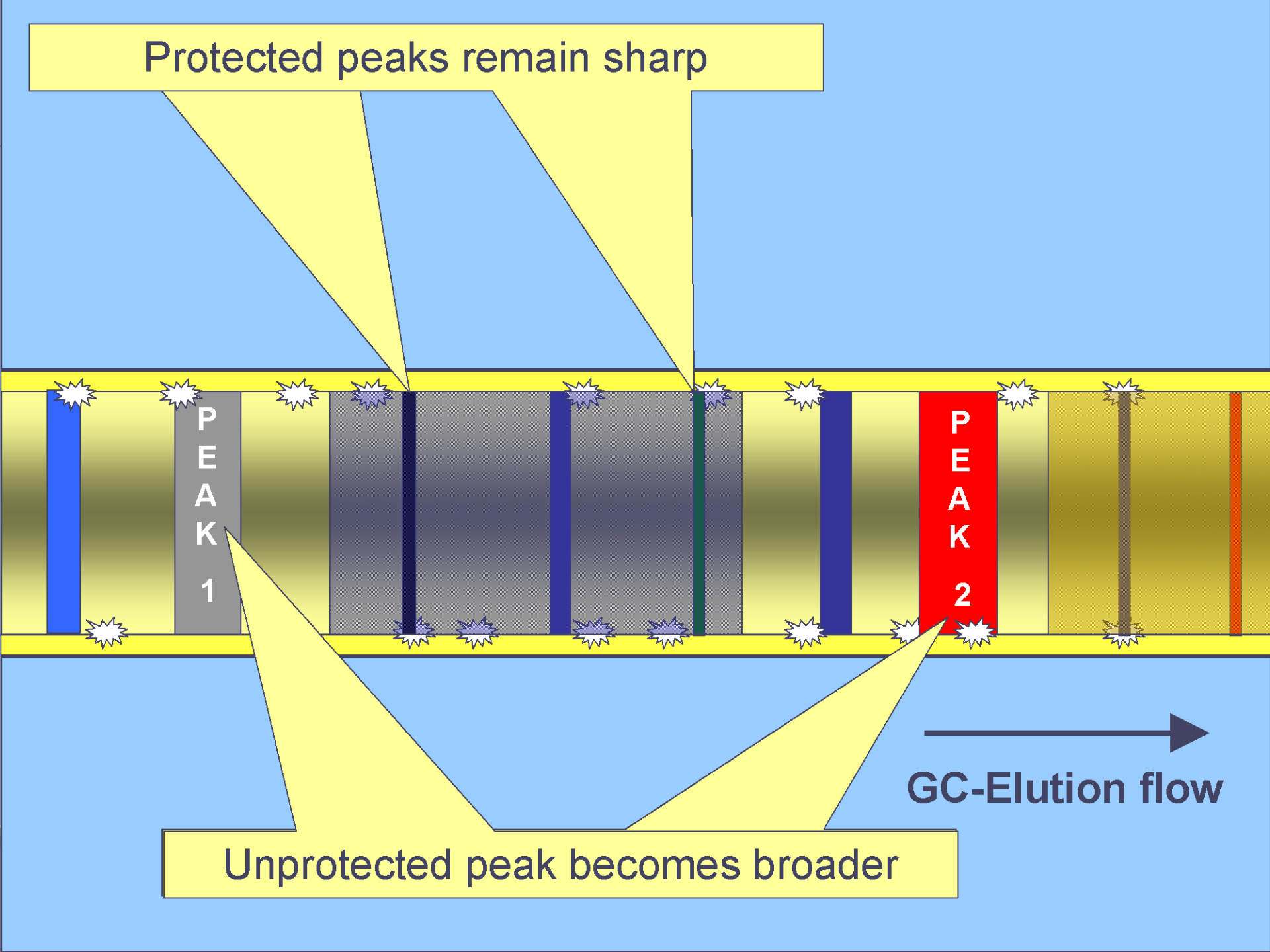
δ-Gulonolactone

Give broadly eluting peaks ⇨ **protection over a wide volatility range**

Analyte Protectants- Desirable properties

- Strong interactions with active sites (H-Bond activity)
- Simmilar volatility to analytes to be protected (so that protection extents during entire run)
- Soluble in sample extract
- Not accumulating in GC-system
- Not reactive with analytes (not inducing their degradation)
- Minimal interference with analyte detection (small m/z)
- Not deteriorating GC-column separation performance
- Cheap and not hazardous


Active Sites
AP-
Elution-Band
Active Site durch AP
abgeschirmt
PEAK 1
PEAK 2
Pesticides
GC-Elution flow

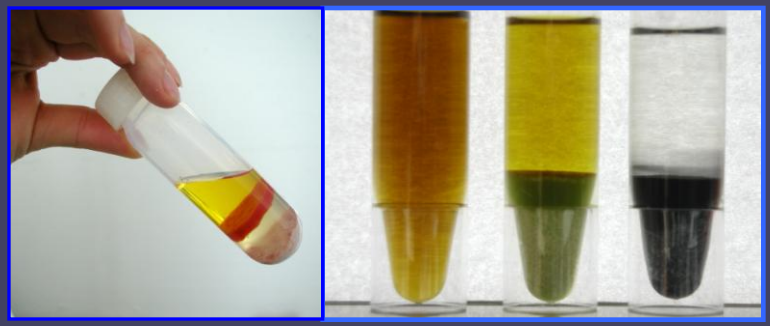

Protected peaks remain sharp
PEAK 1
PEAK 2
GC-Elution flow
Unprotected peak becomes broader

QuEChERS
New Developments

QuEChERS – Further Improvements Some Issues Addressed

- **pH-issue**
  - Stability of pH-labile Compounds
  - Recoveries of Ionizable Compounds

- **Selectivity Issue**
  - Of Extraction/partitioning
  - Of Cleanup
    - Lipids, Sugars
    - Chlorophyll, Carotenoids



- **Expanding Matrix Spectrum**
  - Fatty Commodities
  - Dry Commodities

The pH Issue

- Recoveries of Ionizable Compounds
- Stability of pH-labile Compounds
- Selectivity of Extraction (see later)

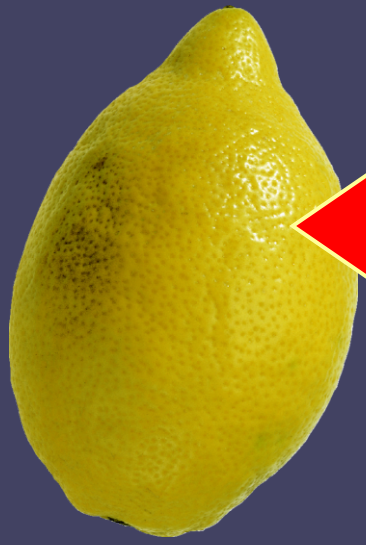
pH-issue – Ionization of Pesticides

Some pesticides get ionized at low or high pH-values

Acids: $HX \leftrightarrows H^+ + X^-$

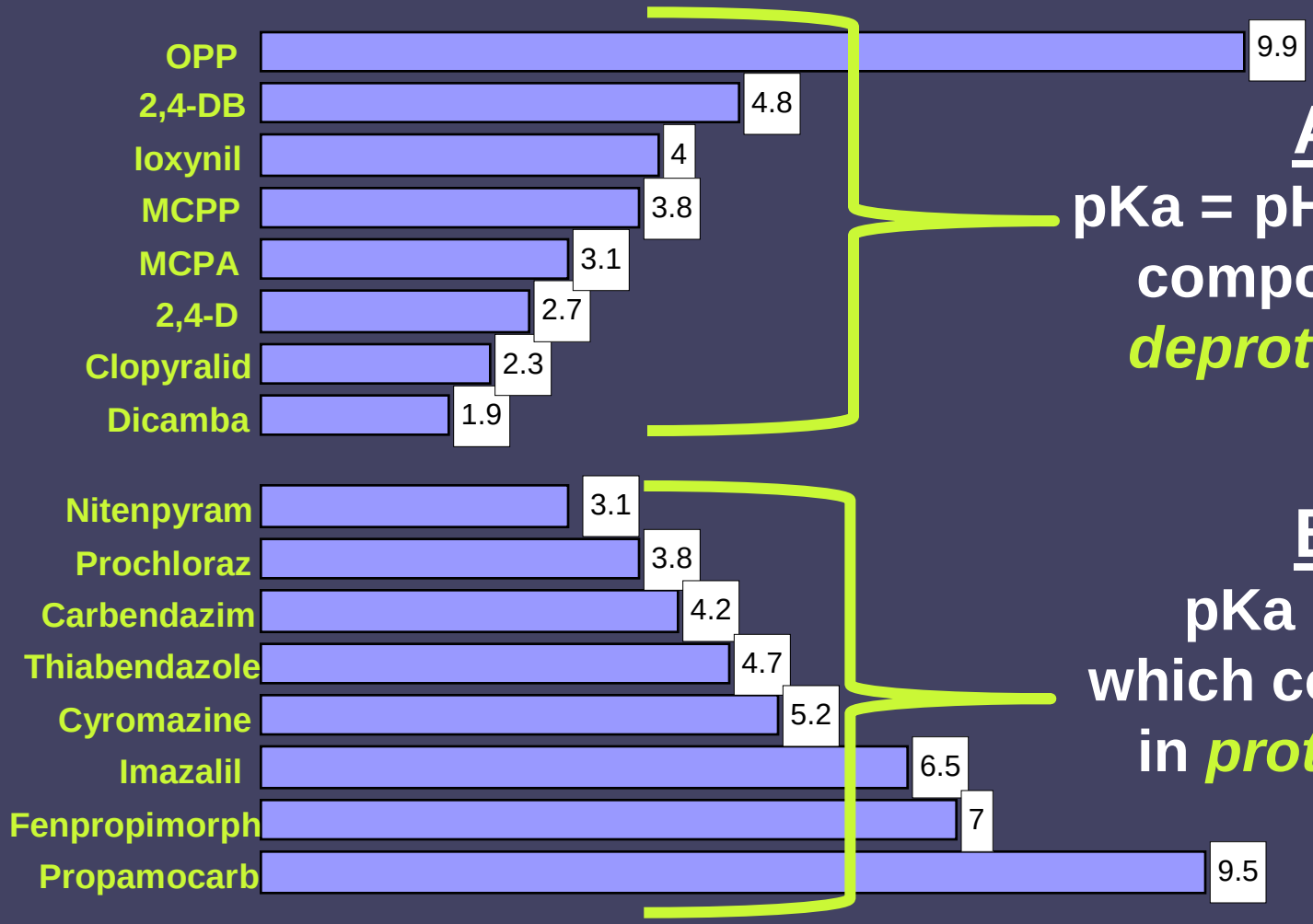
Bases: $B + H^+ \leftrightarrows BH^+$

⇨ **Ionic form prefers to stay in the water phase**



pH-Range of agricultural samples: ~2.5 – 7

Pka-Values of Acidic and Basic Pesticides



Acids

pKa = pH *above* which compounds lay in *deprotonized* form

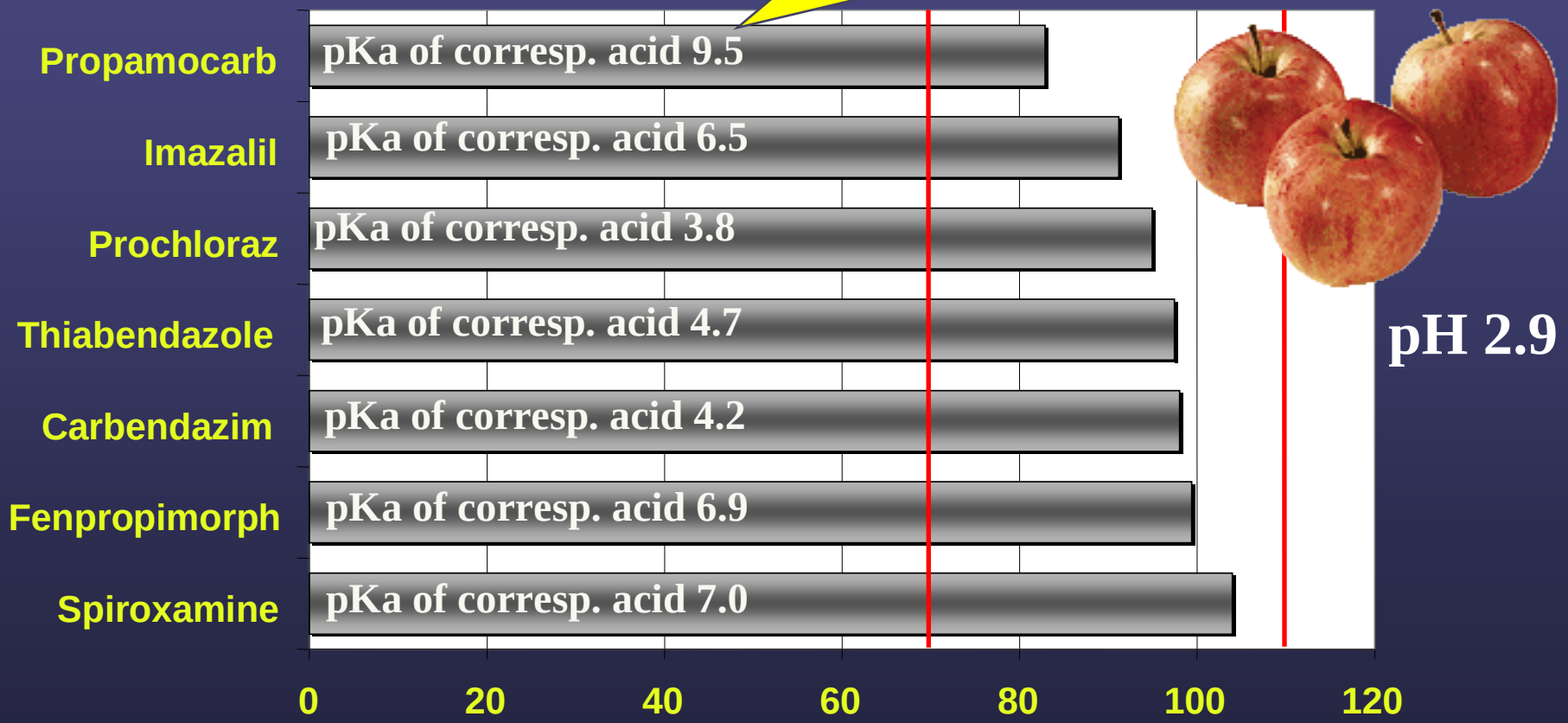
Bases

pKa = pH *below* which compounds lay in *protonized* form

In traditional methods, using non-polar solvents, pH-adjustment 1-2 units > or < PKa is recommended for quantitative recoveries

Basic Pesticides – Not affected!

pKa = pH below which the compound lays primarily in its protonized form



Commodity: Apple;
Fortif. Level: 0.1 mg/kg;
Analysis: LC-MS/MS; ESI (+)

Basic Pesticides and pH

Effect of pH on Recoveries (%)

pKa
pH below which the compound lays **predominantly** in its protonized form

Apple juice, pH adjusted with H_2SO_4	**Thiabendazole pKa = 4.7**		**Imazalil pKa = 6.3**	
	EtAc	**QuEChERS**	EtAc	**QuEChERS**
pH 3	**54**	**90**	**51**	**92**
pH 4	85	**90**	**73**	**94**
pH 5	96	**84**	84	**86**
pH 6	104	**90**	94	**90**

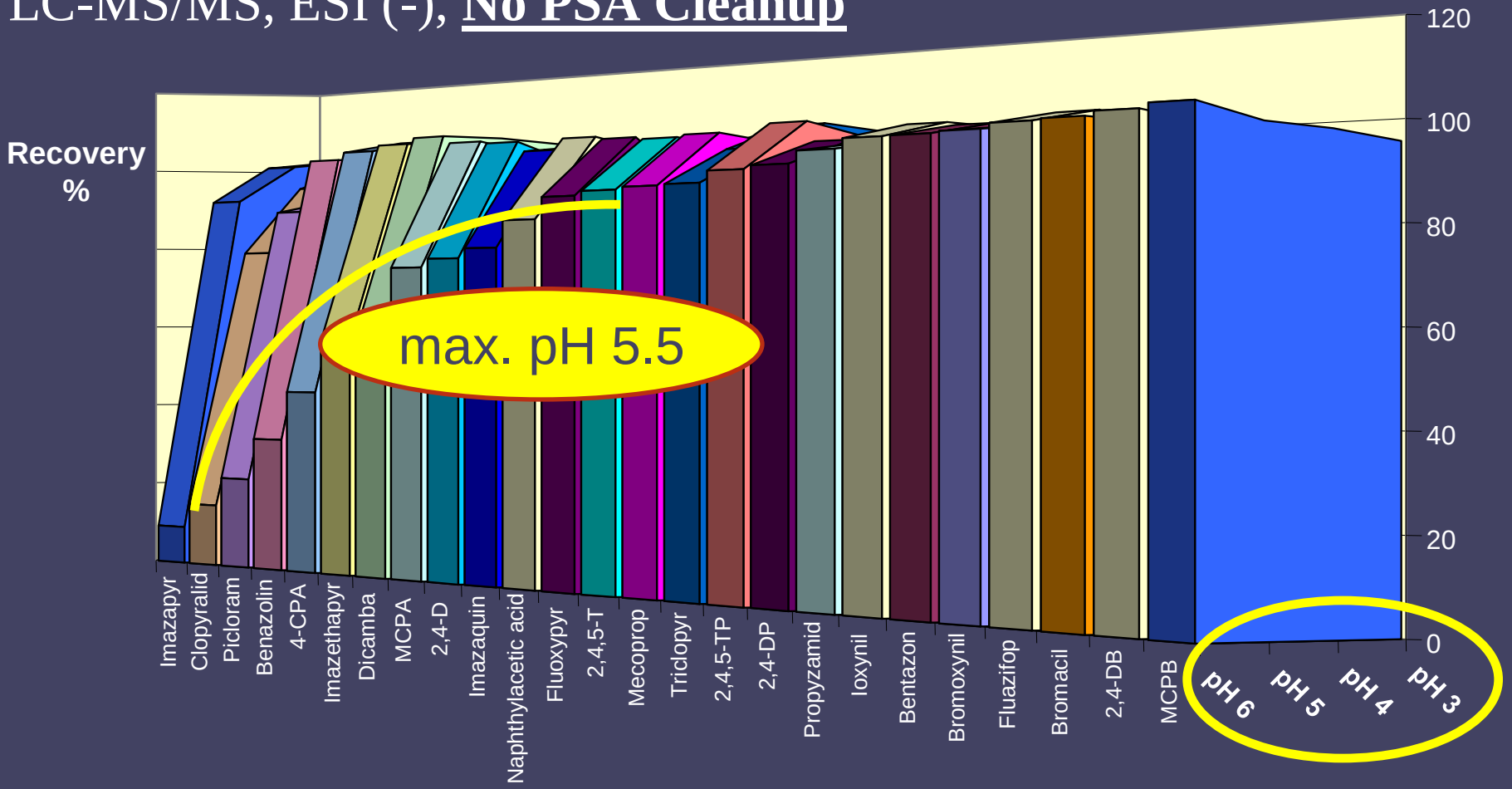
Despite theoretically unfavourable pH, the basic pesticides still prefer to partition into the MeCN phase.

Possible Reason:

After partitioning ACN still contains a considerable amount of water

Acidic Pesticides – Recovery-Drop at pH 6

LC-MS/MS, ESI (-), **No PSA Cleanup**



Lower pKa-Values ***General Trend*** Higher pKa Values

pH-Issue - Labile Compounds

Some Pesticides degrade at high or low pH-values!

- **In the sample (processing, storage)**
  - Keep low temperature

During sample preparation

- Work fast, adjust pH
- Use frozen samples for analysis

$MgSO_4$ + Water ⇨ Heat

- **In the extract during storage (1 week common)**
  - Keep low temperatures, adjust pH

SPE with PSA ⇨ Extract pH > 8

Optimal pH for QuEChERS ?

Goals:

Relevance:

- Still good recoveries for the Strongest Acids
  dicamba, 4-CPA, clopyralid… (pH < 5.5)

Extraction Step

- Still good protection for Base-Sensitives
  tolylfluanid, dichlofluanid, captan, folpet, dicofol, pyridate…
- Still good protection for Acid-Sensitives
  sulfonylureas, pymetrozine, carbosulfan, dioxacarb...

Extraction Step + Extract Storage

pH Adjustment in Extraction Step

Various Buffers tested

Compromise: **Citrate Buffer** at pH 5 to 5.5

- 4 g Magnesium sulphate anhydrous,
- 1 g Sodium chloride (still kept for better selectivity),
- 1 g Trisodium **citrate** dihydrate and
- 0.5 g Disodium hydrogen**citrate** sesquihydrate

✓ ***Good recoveries even for most acidic pesticides*** *(dicamba ....)*

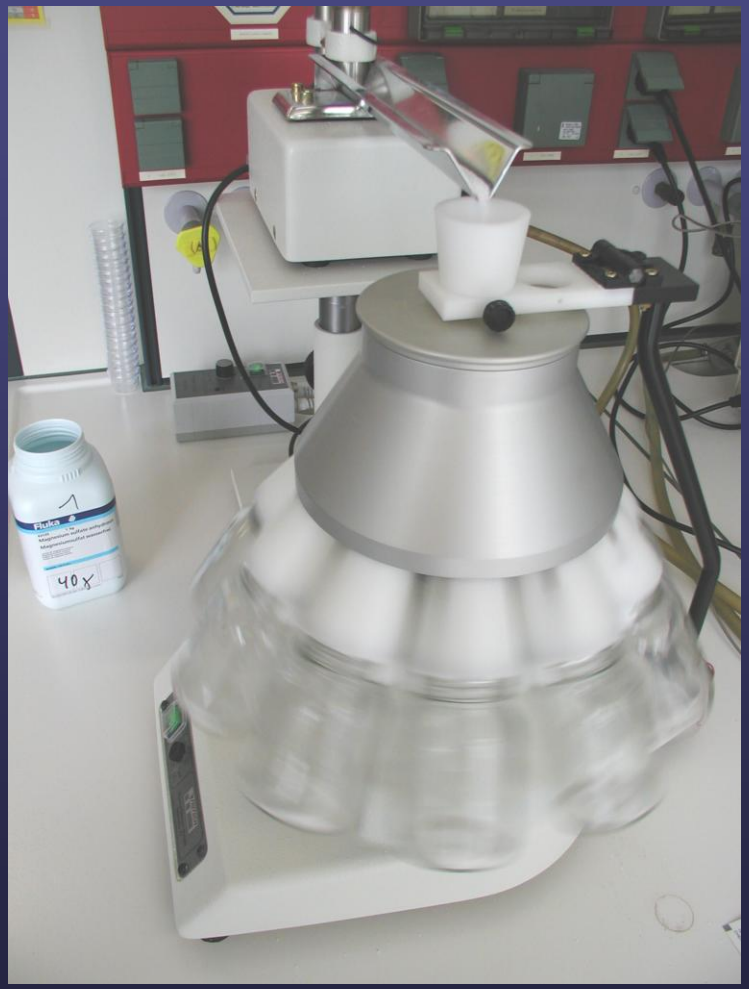
✓ ***Acceptable recoveries for base- and acid-sensitive pesticides***

✓ ***Impoved Selectivity*** *(less co-extractives from* ***acidic samples****)*

✓ ***No negative effect on PSA cleanup*** *(unlike Acetate Buffer)*

Problem: Tedious Weighing of Salts…

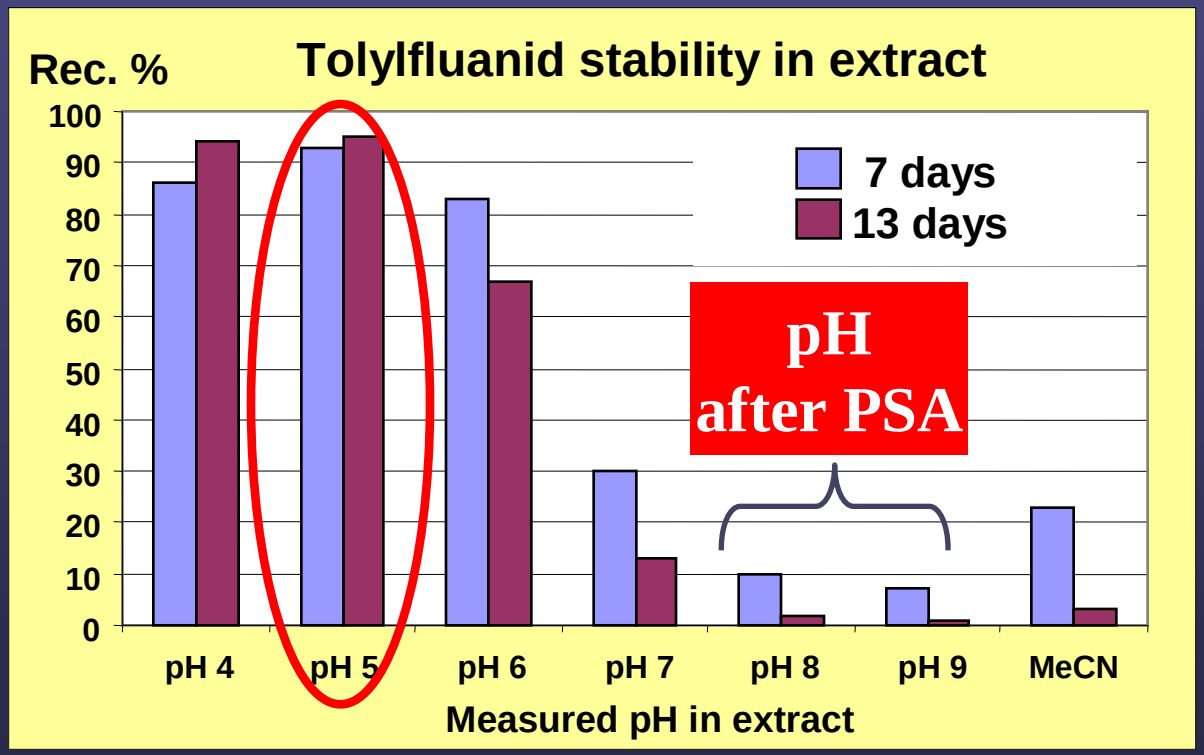
Solution: Rapid & Easy Portioning by "Sample Dividers"



- Some companies offer **ready-to-use mixtures for QuEChERS Partitioning Salts** and **Dispersive SPE Mixtures**

Need to Adjust pH in Final Extracts

Goal: Avoid degradation of Base-labile compounds in final extract.



Also the case for:
dichlofluanid,
captan,
folpet,
dicofol,
pyridate

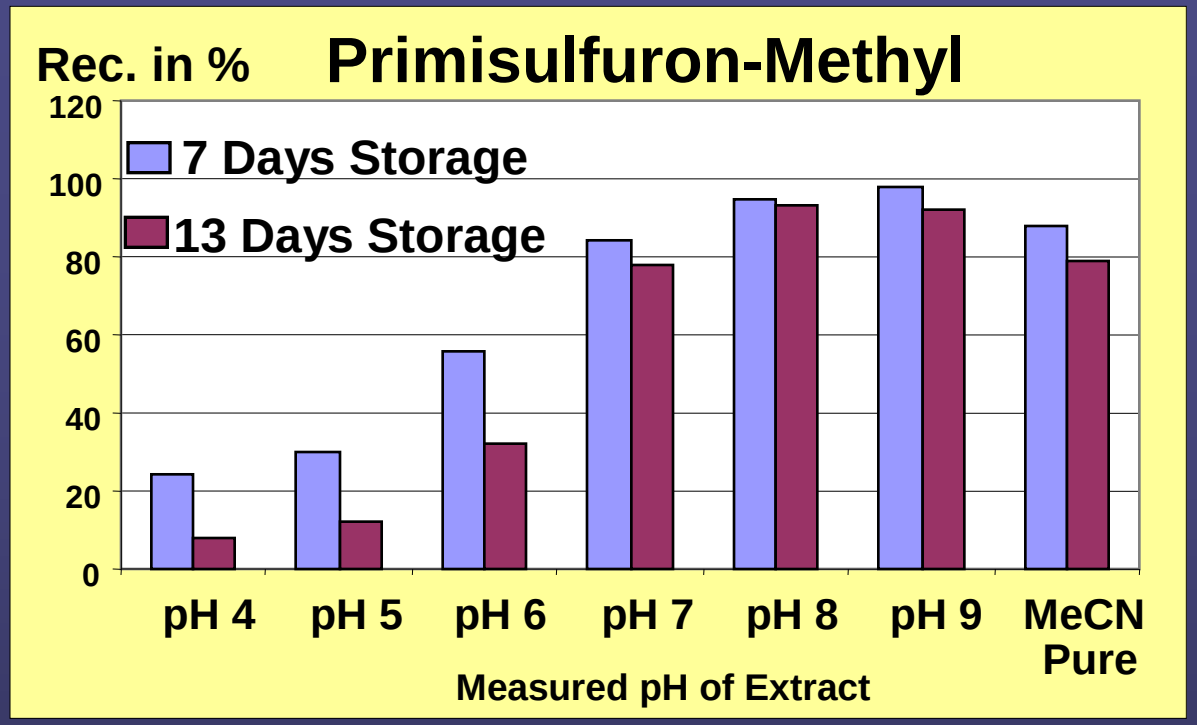
Addition of formic acid (5% in ACN):

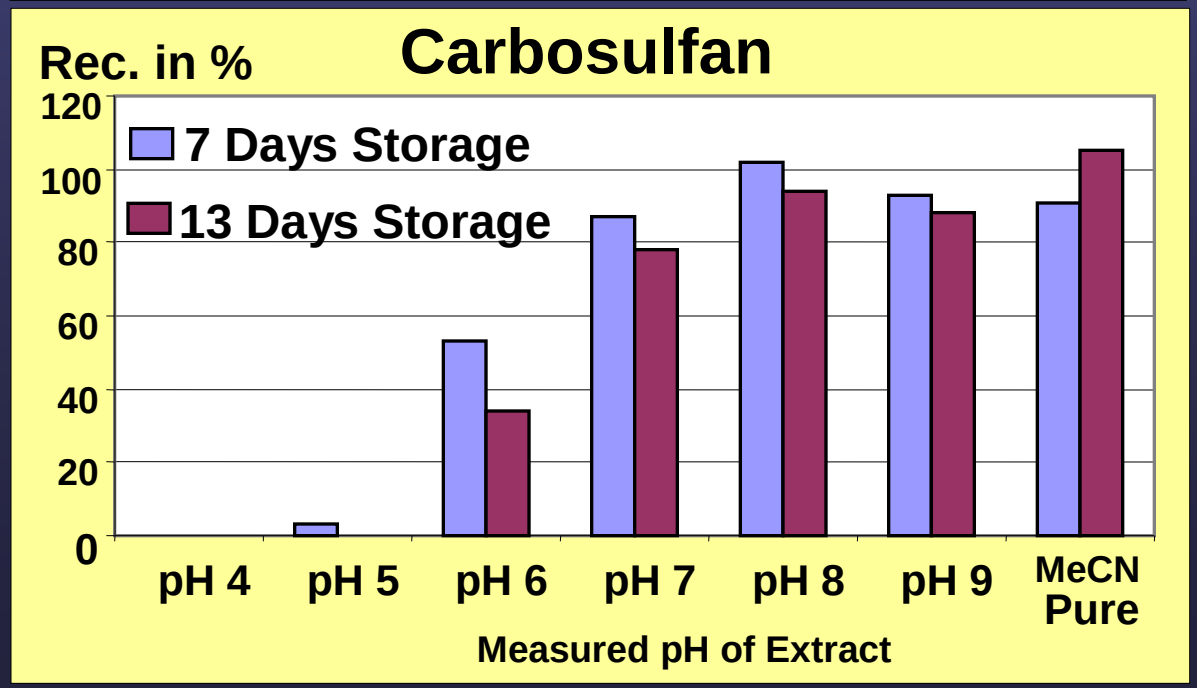
- 10 µL per mL extract brings "pH" to ~5

Sulfonylureas, Carbosulfan

acid labile...

If these compounds are included in the target spectrum use an aliquot of the final extract before acidifying

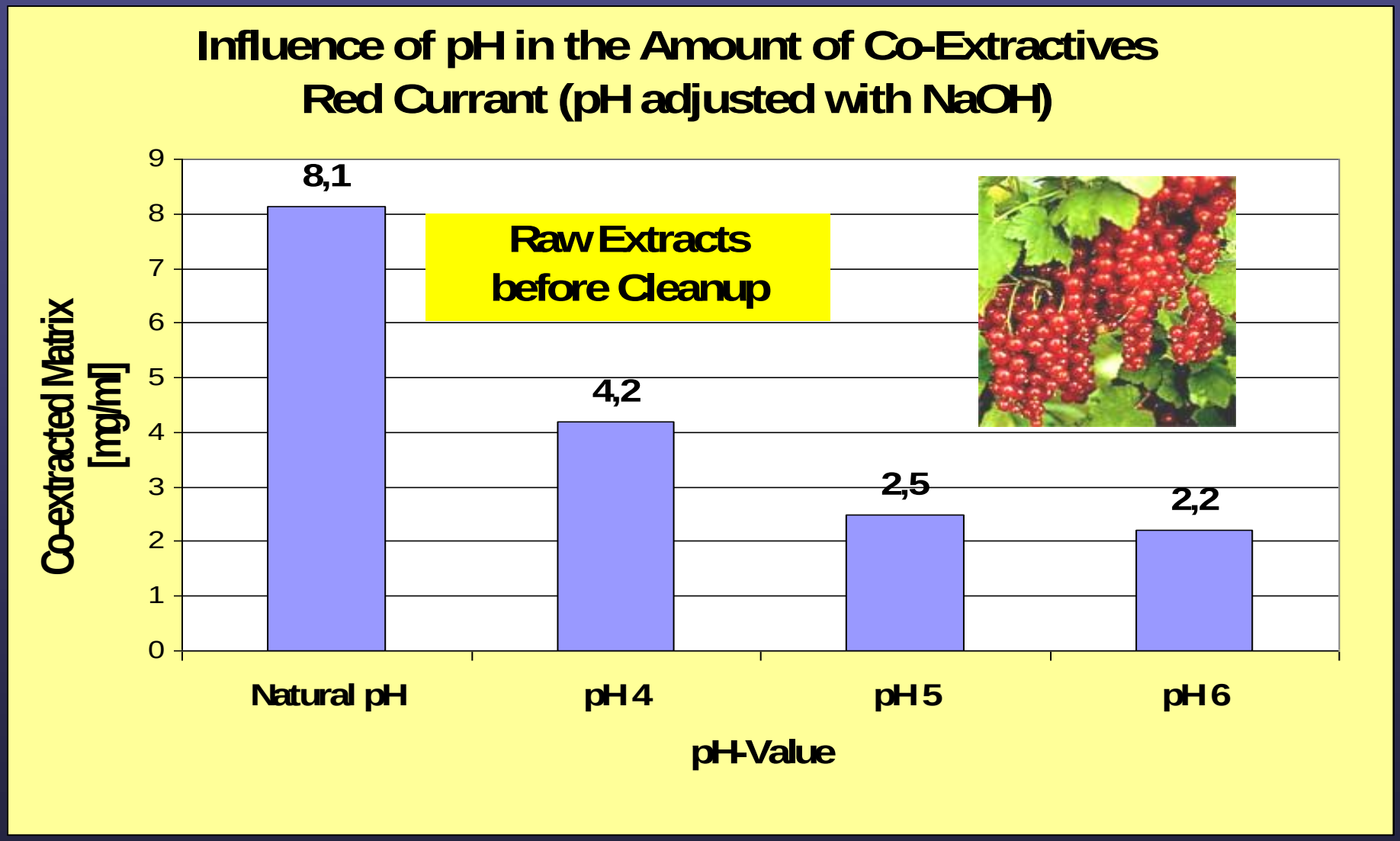




Improving Selectivity

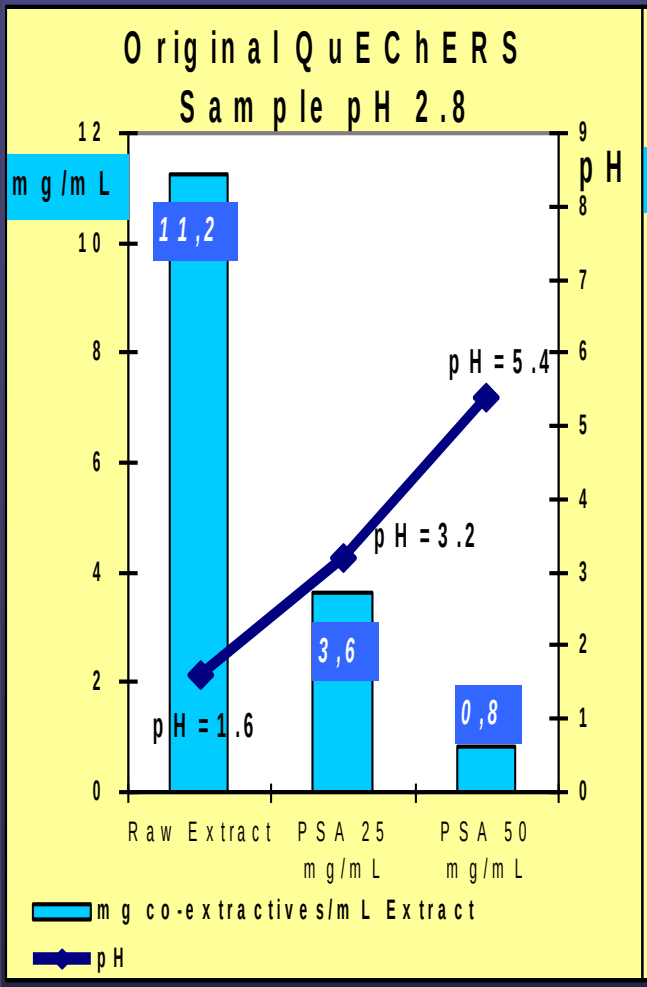
- At Extraction/partitioning Step
  - pH
  - Salts
- At Cleanup Step
  - Lipids, Sugars
  - Chlorophyll, Carotenoids

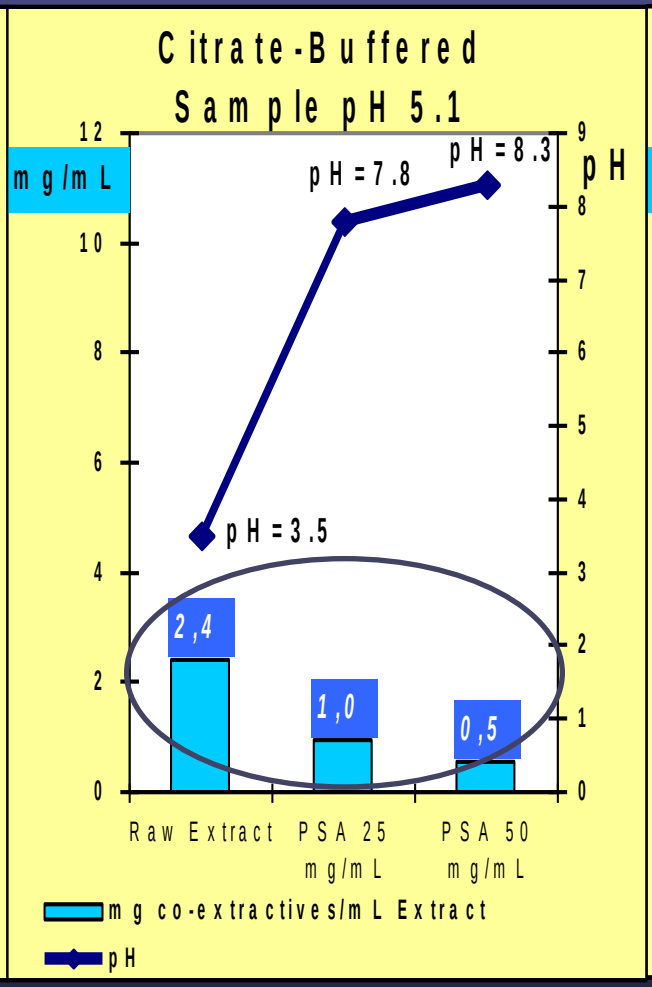
Role of pH in the Selectivity of Extraction/Partitioning

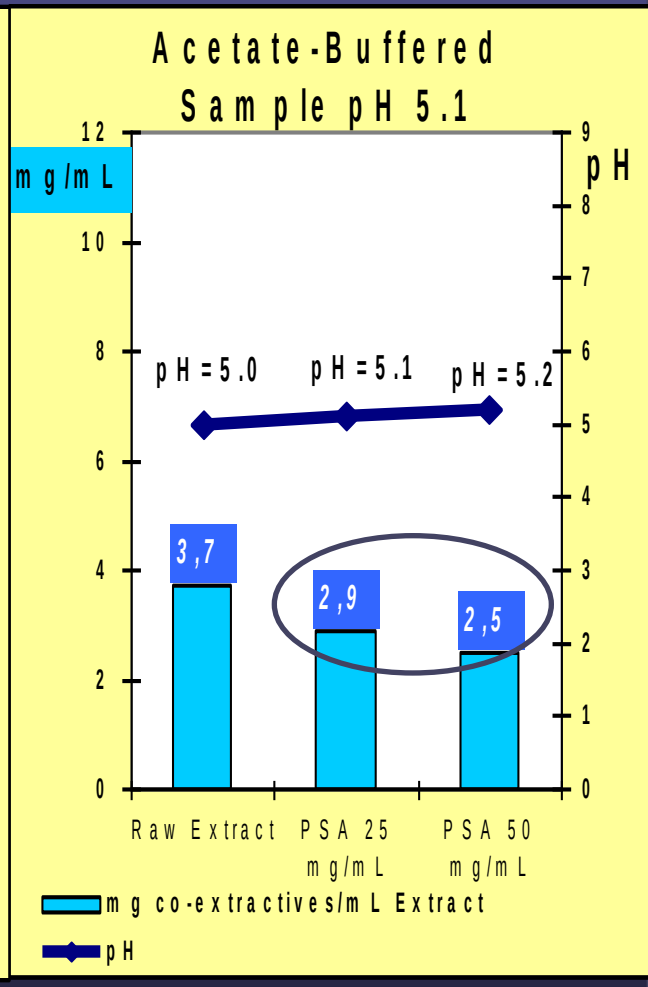


The higher the pH the less co-extractives…

Role of pH in the Selectivity of Extraction/Partitioning Step
Comparison of QuEChERS-Modifications







☺ **Buffering to pH ~5 reduces amount of acidic co-extractives**

☹ **Acetate buffer negatively affects PSA cleanup efficiency**

Selectivity of Cleanup

More than 50 SPE Sorbents and freezing-out tested!

Mainly removed:

❖ **Amino-Sorbents, Alumina:**

- Acids (including fatty acids)
- Sugars
- Pigments (Anthocyanes, some Chlorophyll)

← Losses of acidic pesticides

❖ **Carbon-based Sorbents**:

Carotinoids, Chlorophyll, Sterols

← Losses of planar pesticides

❖ **Reversed-Phase Sorbents**:

- Lipids and Waxes

← No losses observed

❖ **Freeze-out**:

- Lipids and Waxes
- Sugars

← No losses observed

Use of Carbon Sorbents

☹ PSA not satisfying when
high contents of
carotinoids or **chlorophyll**

☞ **Carbon Sorbents more Effective**

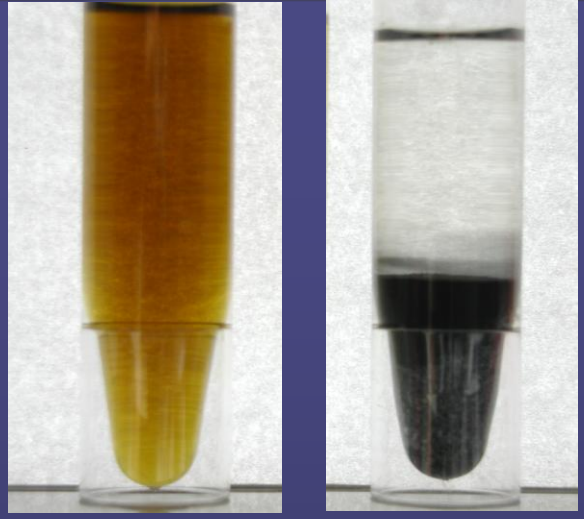
Many tested, GCB (Graphitized Carbon Black) was best in handling

- Used in combination with PSA at small amounts
- Cleanup time (shaking) extended from 30 s to 2 min

☹ Small GCB amounts are difficult to handle …

☞ Pre-mixtures GCB/$MgSO_4$ (powder) facilitate weighing

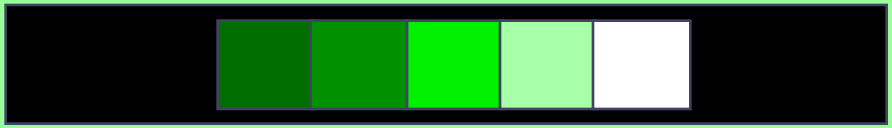
Problems with GCB:



Planar pesticides have a high affinity towards GCB
e.g. hexachlorobenzene, chlorothalonil, thiabendazole

But chlorophyll has higher affinity than all pesticides

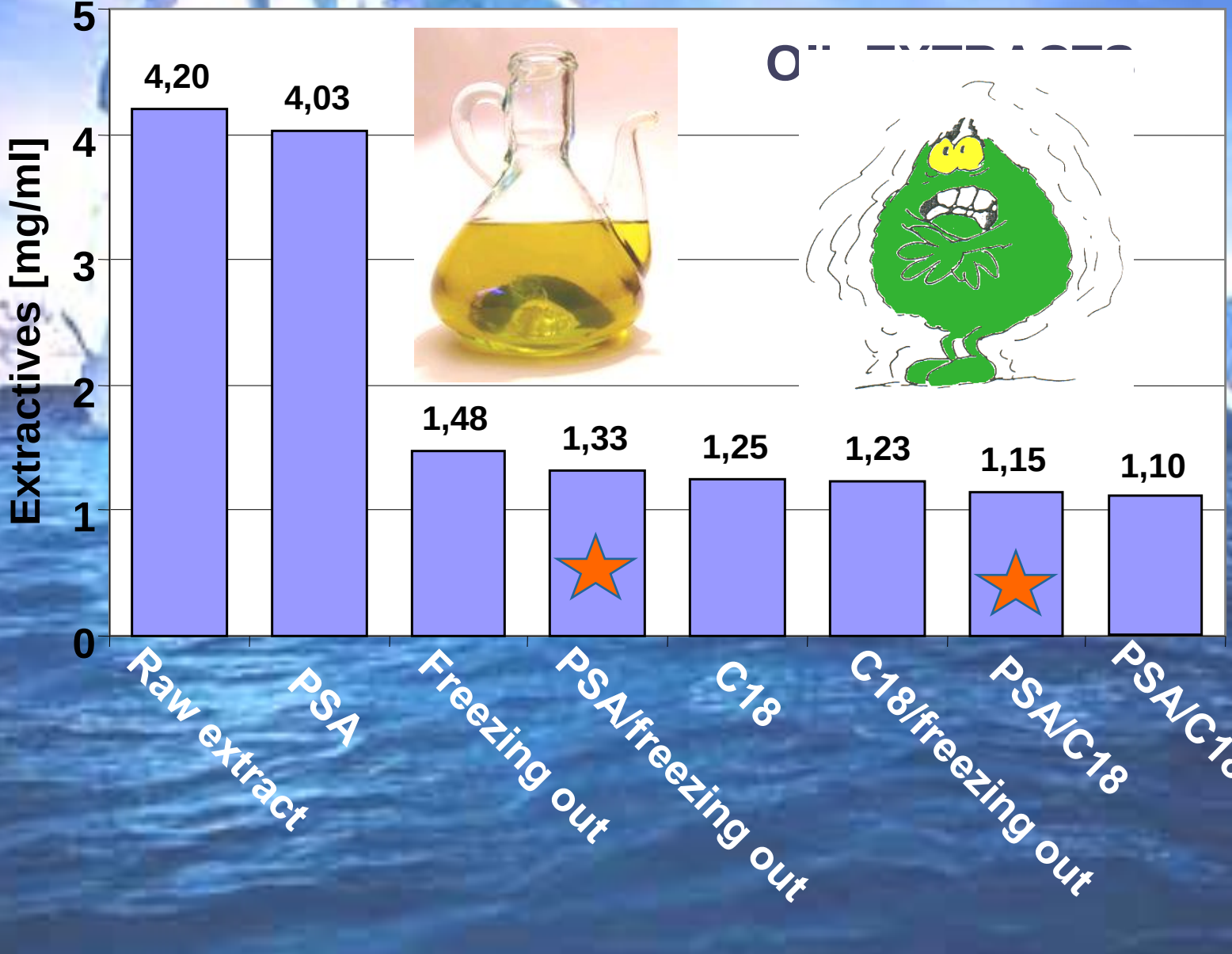
☞Final extract should remain slightly coloured!!



<u>Anthracene</u> may be used as surrogate QC standard.

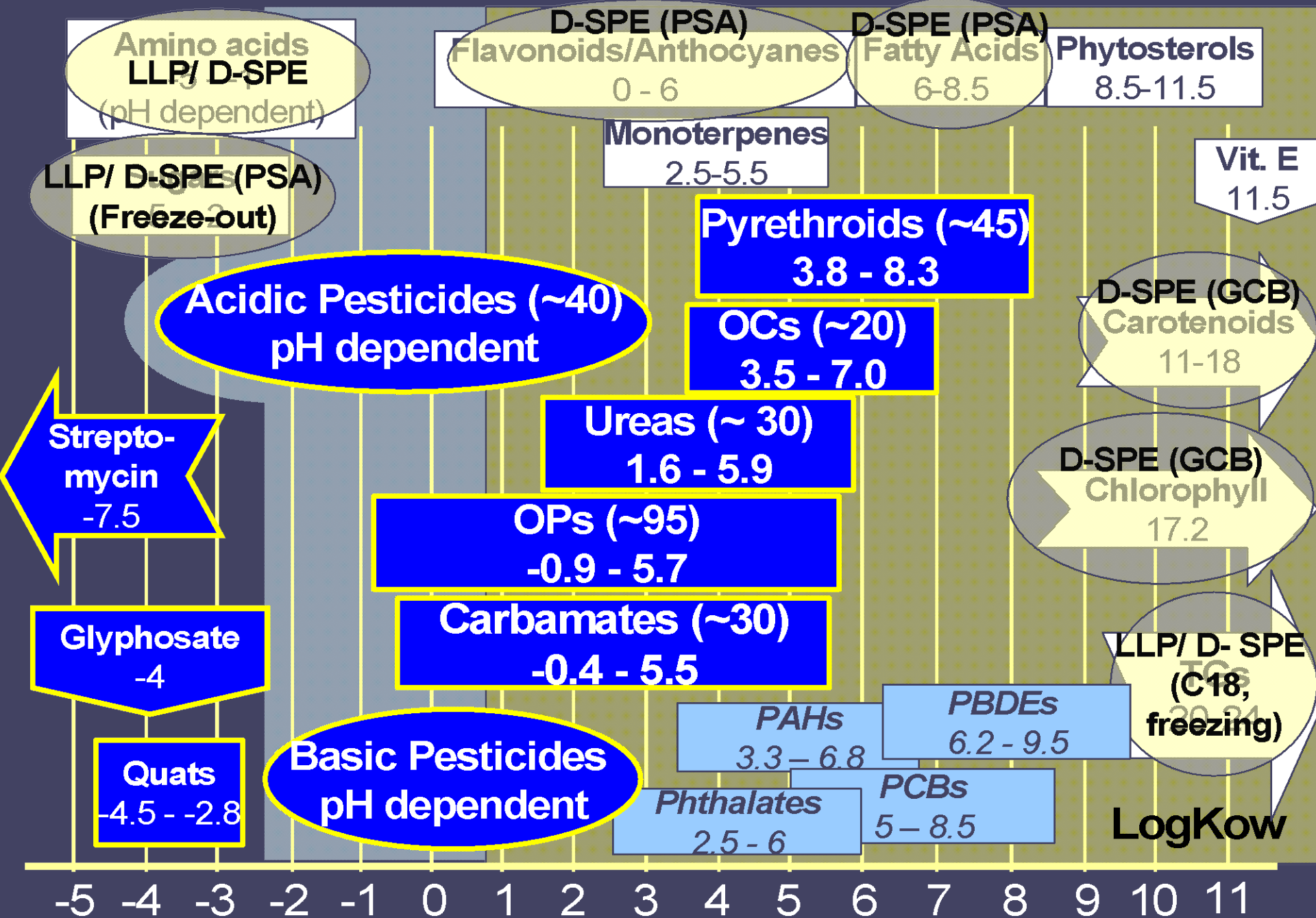
Recoveries > 70% will indicate that no unacceptable losses of pesticides have occurred.

Removal of co-extracted lipids by C18 or freezing out



Removal of co-extractives from Whole-Wheat flour

Scope and Pereformance of QuEChERS



QuEChERS- Multiresidue-Method

Weigh 10 g of Frozen Sample

Add 10 mL Acetonitrile

Add ISTD-Solution

Shake

Add 4 g $MgSO_4$ / 1 g NaCl / Citrate Buffer (pH 5-5.5)

Shake & Centrifuge

Mix an Aliquot w. $MgSO_4$ & Sorbents, freeze-out

Shake & Centrifuge

Acidify extract to pH ~5
to protect base-sensitive pesticides

Optionally: Add other "Analyte Protectants"

Changes introduced to the method.

The method will become official CEN method

Optionally:
Acidic Pest. by LC-MS/MS

Optionally:
SUs by LC-MS/MS

Multiresidue Analysis
by GC-MS, LC-MS ...

Broaden matrix spectrum

Dry commodities (cereals, dried fruits)

Fatty Commodities

Broaden matrix spectrum – Dry Commodities

E.g. cereals, dried fruits

- **Water-Addition prior to extraction**
  - **to weaken interactions of pesticides with matrix and to ensure adequate partitioning.**

→ Sample amount is reduced and water is brought to 10 mL

→ Co-extracted fat removed by freezing out or C18, if necessary....

Dry Commodities

Sample type	Weigh	Water	Annotation
Fruit/Vegetables (water >80 %)	10 g	-	
Fruit/Vegetables (water 30-80 %)	10 g	X g	X = 10 g – water amount in 10 g sample
Cereals	5 g	10 g	
Dried fruits	5 g	8.5 g	Add water to comminute, weigh 13.5 g of homogenate
Honey	5 g	10 g	
Spices	2 g	10 g	

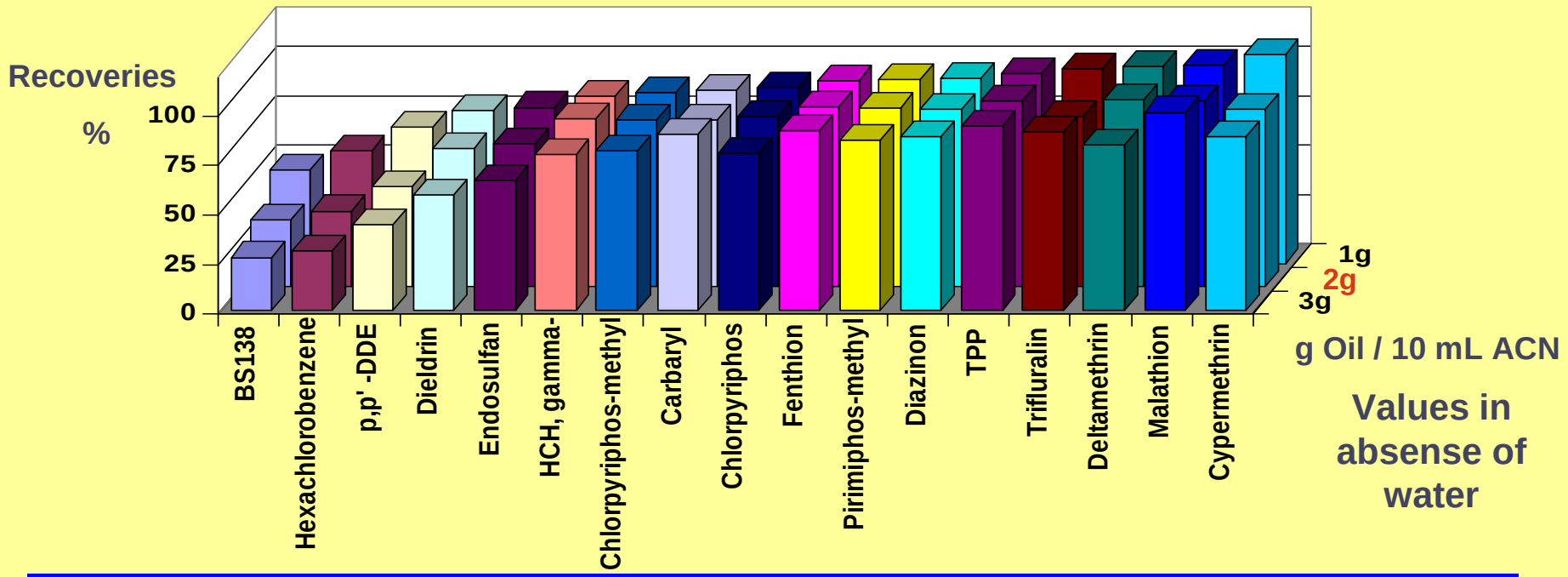
Broaden matrix spectrum – Fatty commodities

Commodities with a high lipid load, such as avocados or plant oils can be employed.

Problems:

- Co-extracted lipids should be removed prior to GC-analysis
- Highly non-polar pesticides may give recoveries < 70% (e.g. HCB and DDT)
- Accessibility of residues may be limited (Ultra Turrax)

Recoveries of pesticides in high fat samples



PCB 138 or 153 may be used as surrogate QC standards

Rec. > 70% will indicate that no unacceptable pesticide losses occurred

The **tolerable lipid-amount** depends on the selection of pesticides to be covered

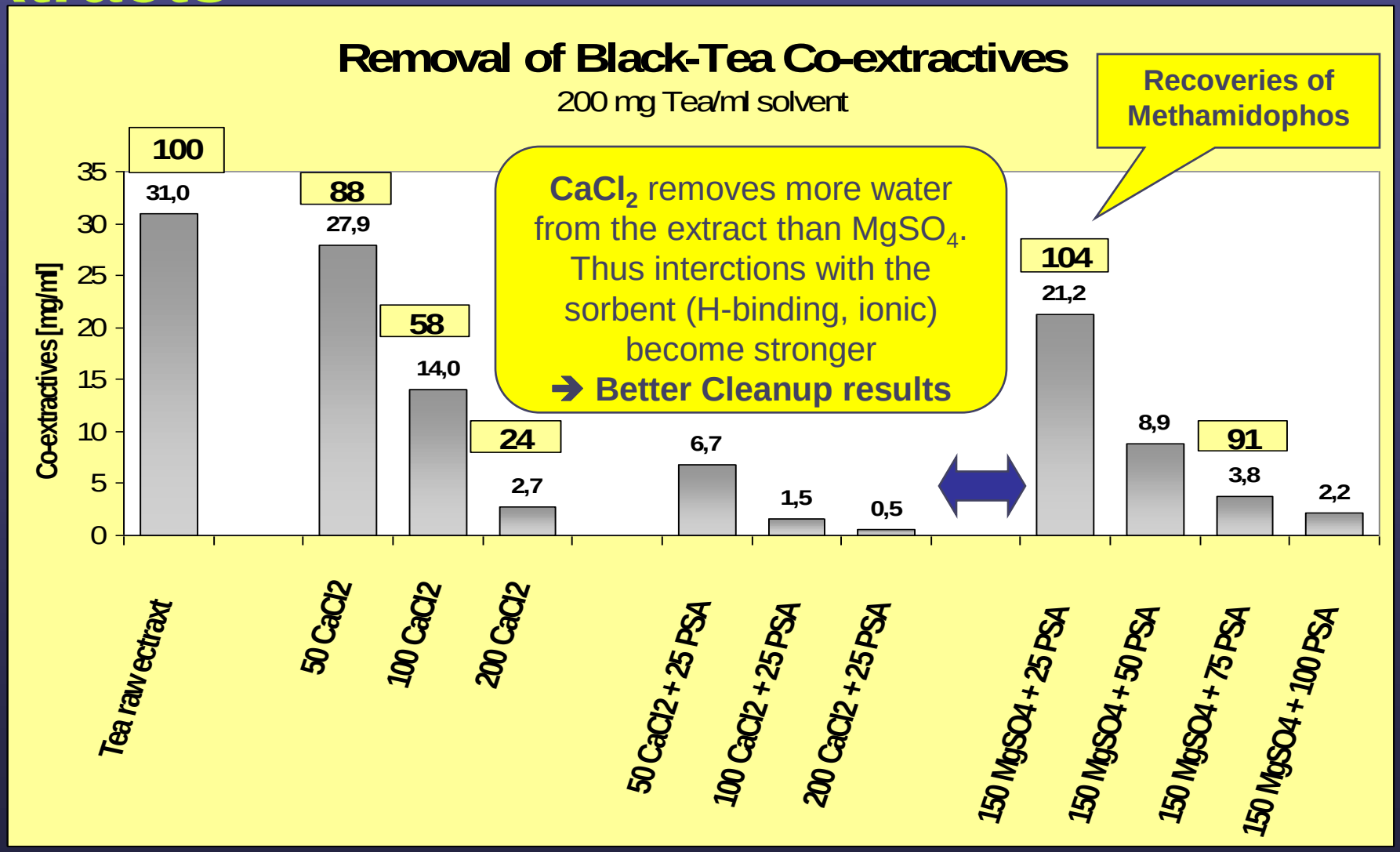
e.g. for HCB 0.4 g lipids are still OK (>70% rec.), for DDE 1 g, for Endosulfane 5 g

(NOTE: In presence of water (ternary system) values are different, less lipid is tolerable)

Compromise for Oil samples: 2 g oil + 10 mL ACN

- HCB and DDE give recoveries <70%…
- but equilibrium is defined and recovery-correction is justified

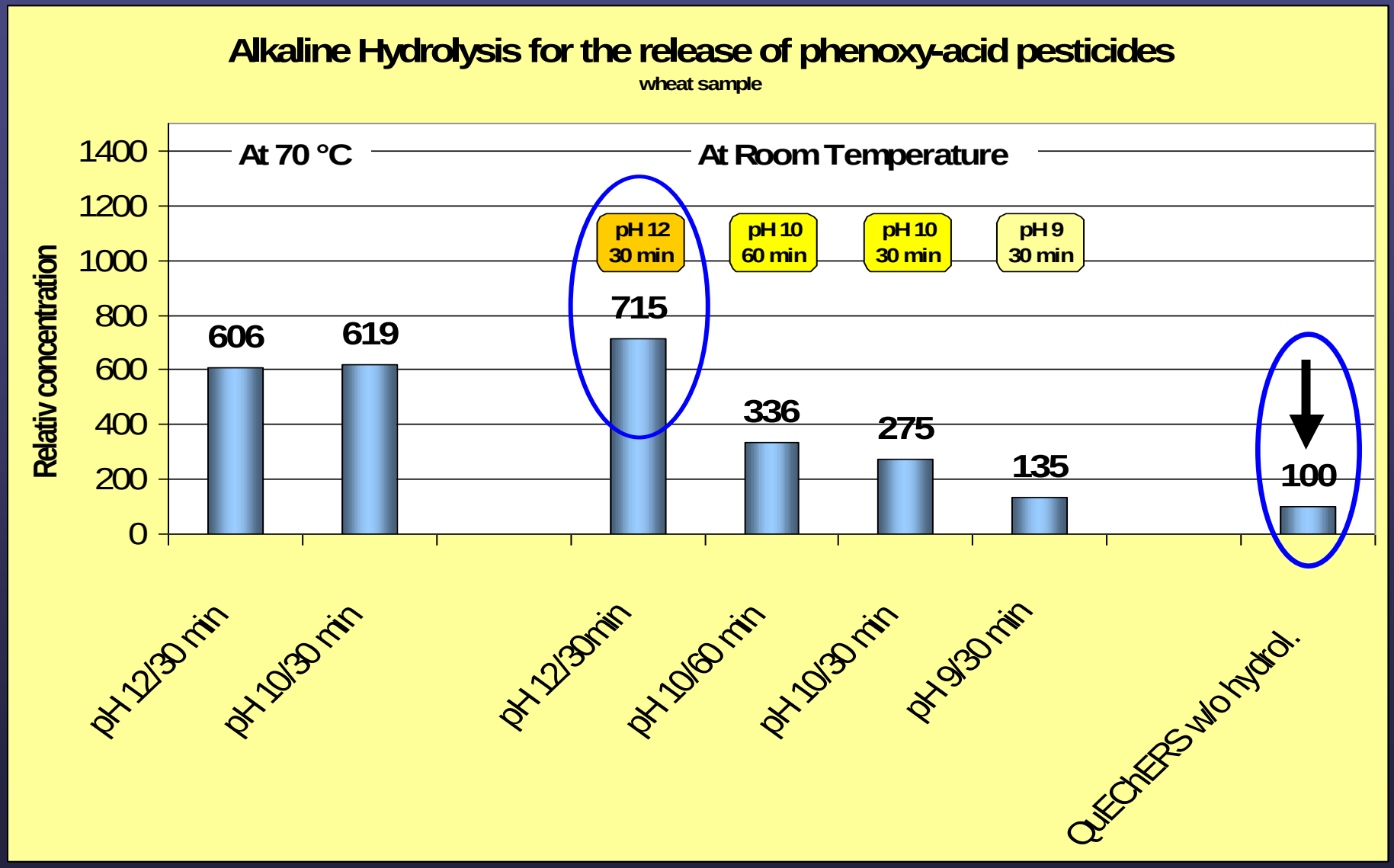
Cleanup of Fermented Tea extracts



Problem with $CaCl_2$: recoveries of polar pesticides drop

➔ if polar pesticides are not of interest $CaCl_2$ / PSA is a serious cleanup option

Release of covalently bound phenoxy-acids by alkaline cleavage



Impact of QuEChERS-Implementation

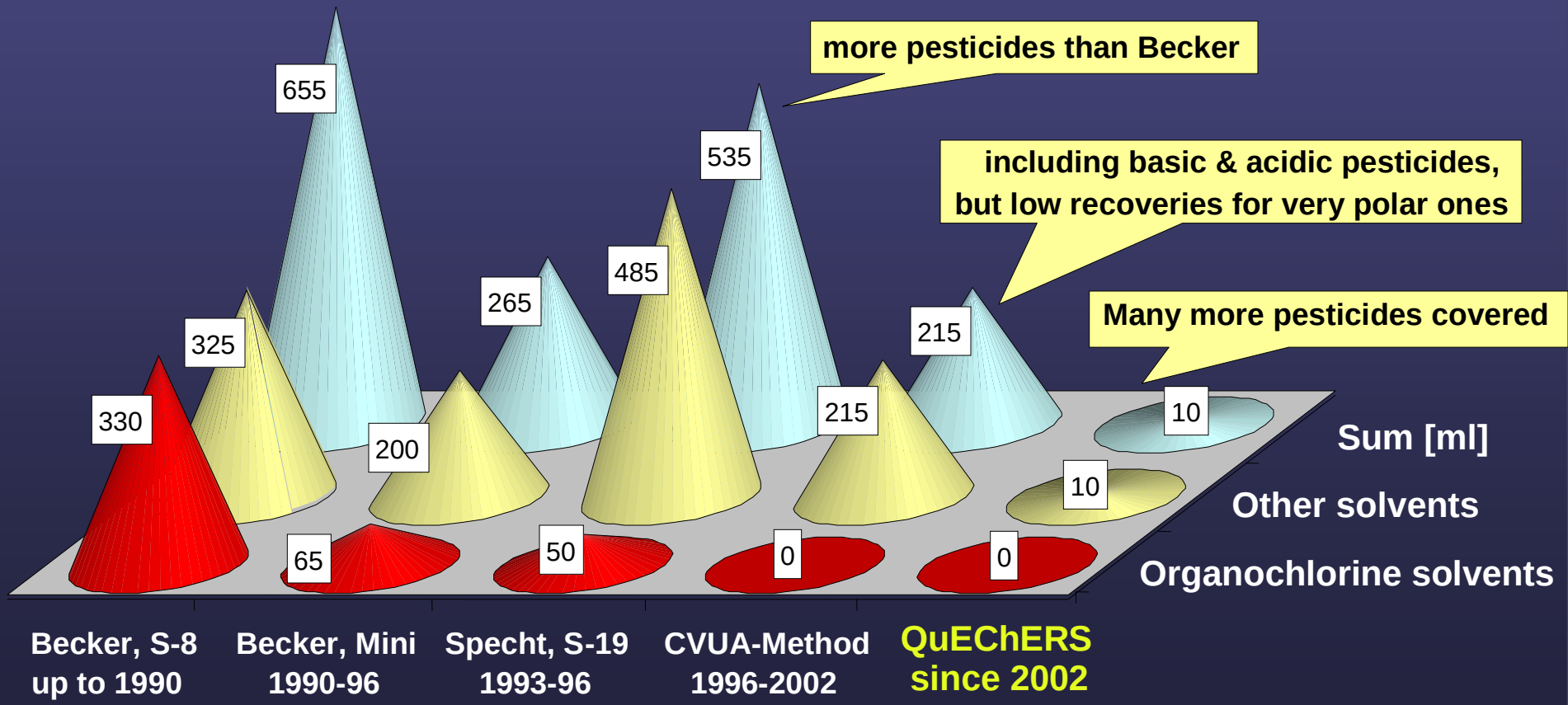
- ✓ More time for instrumental analysis
- ✓ More time for QA/QC (incl. validation)
- ✓ Broader analyte spectrum
- ✓ Higher sample throughput and turnaround time
- ✓ Less solvent consumption
- ✓ Less lab space needed (hoods are empty)
- ✓ Sample preparation more pleasant


People in the Lab
are more HAPPY!!

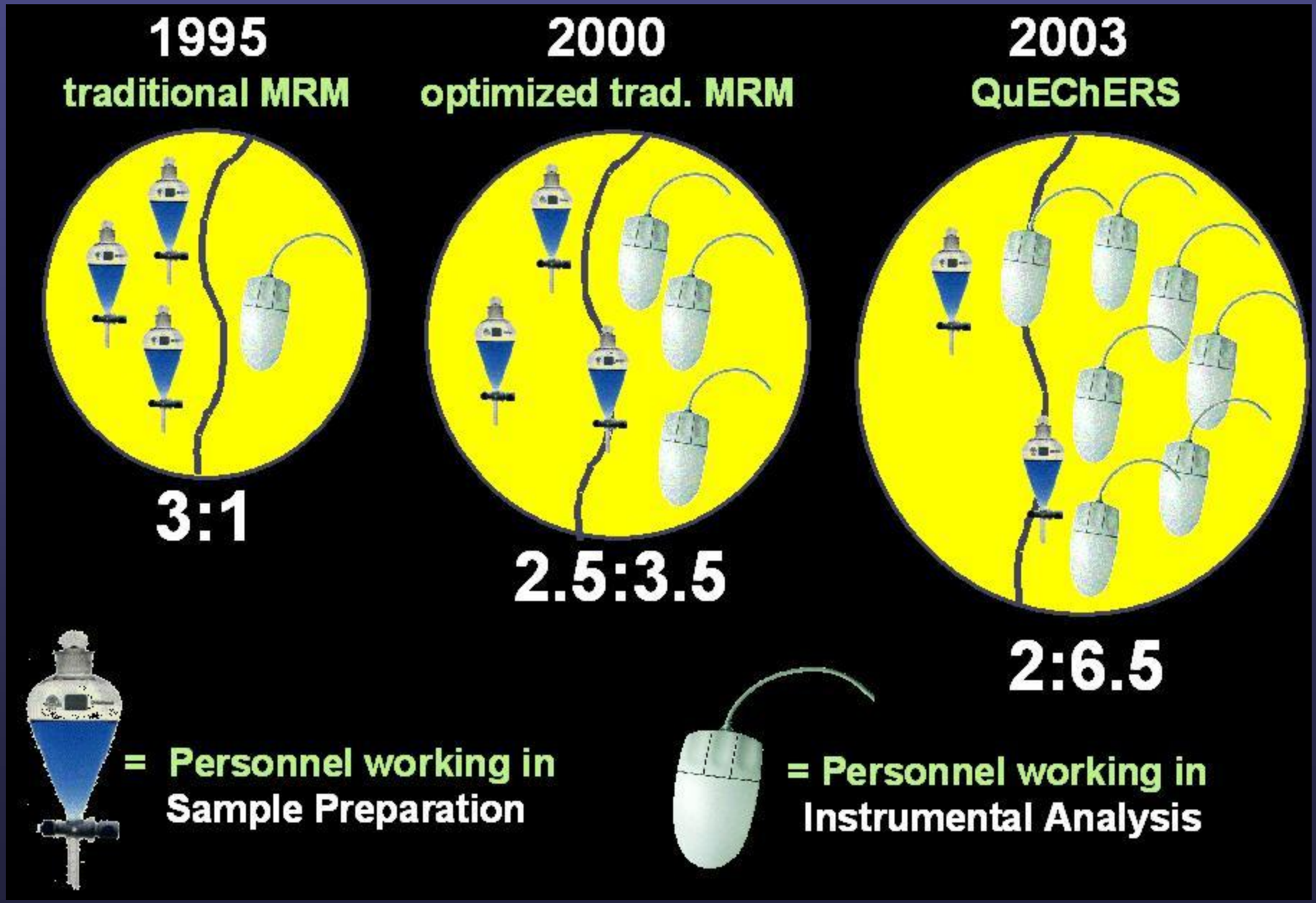
Reduction of Solvent Consumption

15.000 € savings in 1 year just for solvent !! (for ca. 2000 samples)

mL Solvent/Sample

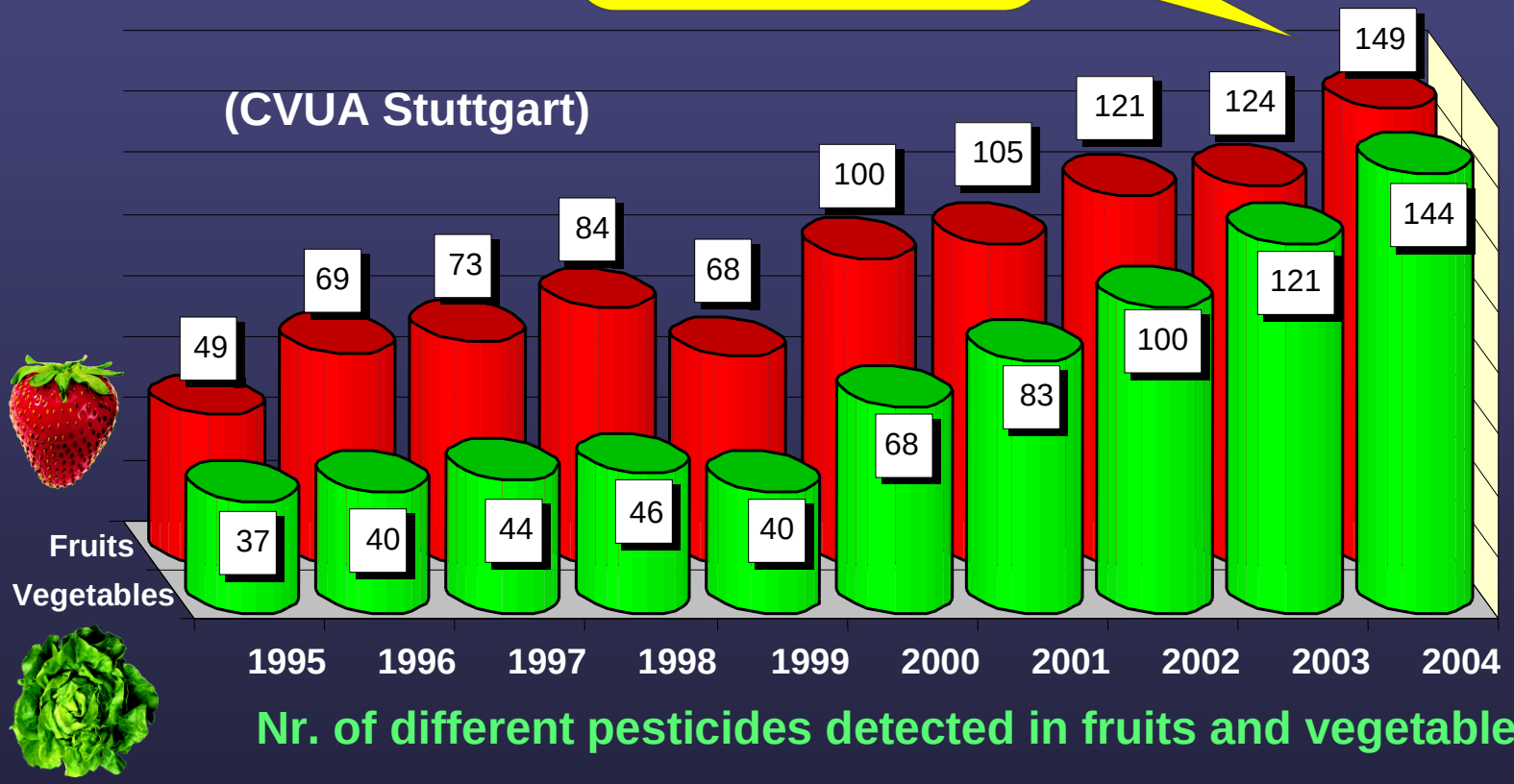


"Mass Migration" of Personnel



more findings...

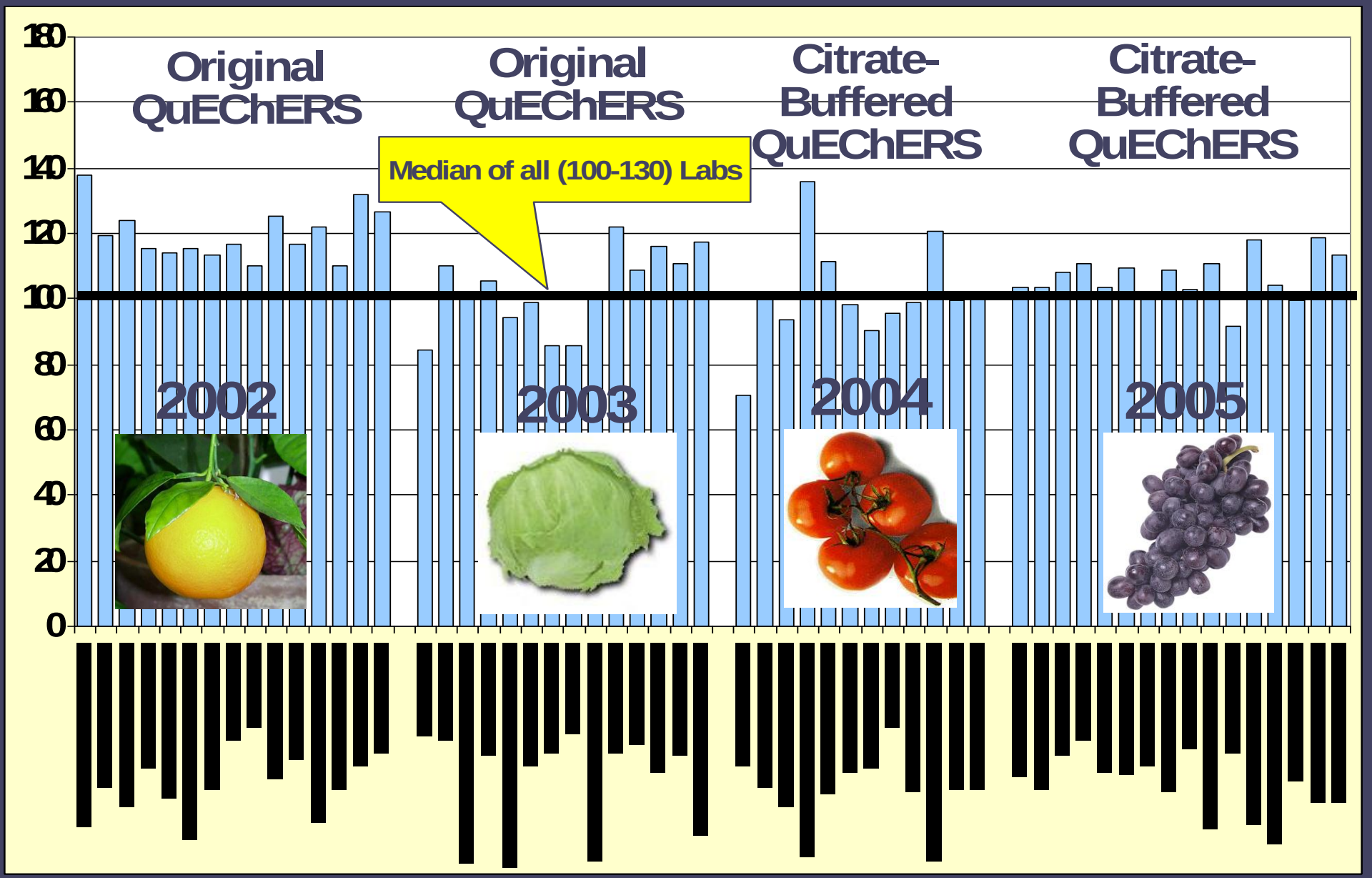
In 2004:
200 Pesticides
in Total



Nr. of different pesticides detected in fruits and vegetables

Participation in EU-Proficiency Tests using the QuEChERS-Method

Using QuEChERS in EU - Proficiency Tests



Using QuEChERS in EU - Proficiency Tests

Unknown Pesticides and Unknown concentrations

Participants: 100-130 EU-Official Labs

Results:

- ✓ **All 57 identified (100%)**
  - ✓ **95% (54/57): within +/-30% from median concentration**
  - ✓ **82% (47/57): within +/-20%**
  - ✓ **53% (30/57): within +/-10%**
- ✓ **On average +8% above the median**

2002

2003

2004

2005

QuEChERS
Inter-Laboratory Validation Studies

GC-MS and LC-MS/MS Inter-Laboratory Validation Study (GDCh)

Pesticide Name		Mean Recovery						RSD (%)						Nr. of Laboratories reported results (n=5 each)					
		Apple 0,25	Apple 0,025	Lettuce 0,25	Lettuce 0,025	Orange 0,25	Orange 0,025	Apple 0,25	Apple 0,025	Lettuce 0,25	Lettuce 0,025	Orange 0,25	Orange 0,025	Apple 0,25	Apple 0,025	Lettuce 0,25	Lettuce 0,025	Orange 0,25	Orange 0,025
Azoxystrobin	GC	97%	104%	105%	105%	108%	112%	4%	9%	7%	1%	14%	7%	3	3	3	2	3	3
Chlorpyrifos	GC	104%	104%	105%	104%	104%	105%	3%	7%	5%	6%	5%	6%	9	9	8	8	8	8
Cyprodinil	GC	101%	101%	103%	102%	100%	95%	3%	6%	7%	7%	5%	8%	7	7	6	6	6	6
Dimethoat	GC	97%	103%	109%	97%	99%	94%	8%	13%	8%	14%	21%	7%	4	4	3	2	3	3
Fenhexamid	GC	83%	86%	78%	79%	90%	90%	9%	11%	15%	20%	12%	8%	5	5	3	2	4	4
Imazalil	GC	102%	100%	101%	104%	102%	98%	8%	4%	7%	8%	8%	4%	5	4	4	3	4	4
Kresoxim-methyl	GC	104%	102%	105%	108%	104%	103%	6%	9%	6%	4%	8%	7%	8	8	7	7	7	7
lambda-Cyhalothrin	GC	105%	124%	101%	115%	105%	119%	8%	14%	7%	13%	10%	15%	9	9	8	7	8	8
Metalaxyl	GC		100%	106%	105%	99%	100%	7%	9%	5%	11%	6%	2%	7	7	6	6	6	5
Myclobutanil	GC		106%	104%	106%	103%	101%	7%	13%	5%	4%	6%	4%	8	8	7	7	7	7
Penconazol	GC		107%	103%	104%	103%	102%	2%	12%	5%	4%	5%	4%	8	8	7	7	7	7
Pirimicarb	GC		97%	104%	104%	97%	98%	6%	4%	3%	4%	5%	3%	6	6	5	5	5	5
Procymidon	GC		104%	10															8
Propyzamid	GC	106%	105%	10															7
Pyridaben	GC	105%	104%	10															7
Pyrimethanil	GC	101%	102%	10															7
Quinoxyfen	GC	102%	102%	10															7
Tetradifon	GC	104%	102%	10															8
Thiabendazol	GC	87%	88%	9															2
Mean Recovery GC		**99%**	**102%**	**10**															
Acetamiprid	LC (+)	99%	99%	9															9
Azoxystrobin	LC (+)	102%	101%	10															9
Carbendazim	LC (+)	92%	92%	9															9
Dimethoat	LC (+)	97%	99%	9															9
Fenhexamid	LC (+)	79%	78%	7															9
Cyprodinil	LC (+)	100%	98%	9															7
Imazalil	LC (+)	97%	100%	9															9
Kresoxim-Methyl	LC (+)		%	10															4
Metalaxyl	LC (+)		%	10															8
Methamidophos	LC (+)		%	8															9
Methiocarb	LC (+)		%	10															9
Myclobutanil	LC (+)	100%	101%	97%	96%	101%	101%	6%	2%	4%	5%	5%	4%	5	5	5	5	5	5
Penconazole	LC (+)	100%	102%	98%	98%	100%	97%	3%	5%	3%	5%	6%	5%	6	6	6	6	6	6
Pirimicarb	LC (+)	97%	99%	94%	95%	99%	96%	4%	5%	5%	8%	3%	3%	7	7	7	7	7	7
Propamocarb	LC (+)	83%	84%	86%	83%	80%	79%	8%	5%	9%	11%	6%	7%	9	9	9	9	9	9
Propyzamid	LC (+)	99%	100%	102%	98%	102%	96%	5%	3%	6%	4%	1%	7%	4	4	4	4	4	4
Pyridaben	LC (+)	101%	103%	100%	97%	104%	110%	3%	5%	4%	8%	9%	7%	4	4	4	4	4	4
Pyrimethanil	LC (+)	99%	102%	95%	96%	100%	98%	3%	3%	4%	9%	5%	7%	7	7	7	7	7	7
Quinoxyfen	LC (+)	99%	97%	93%	98%	103%	100%	9%	1%	4%	5%	14%	1%	2	3	3	3	3	2
Tebufenozide	LC (+)	103%	101%	101%	98%	102%	99%	5%	4%	5%	5%	5%	8%	9	9	9	9	9	9
Thiabendazole	LC (+)	93%	94%	89%	87%	91%	87%	5%	7%	8%	9%	6%	9%	9	9	9	9	9	9
Mean recovery LC (+)		**96%**	**97%**	**95%**	**94%**	**97%**	**94%**	**5%**	**5%**	**6%**	**7%**	**5%**	**7%**						
2,4-D	LC (-)		%	102%	98%	100%	96%	2%	4%	10%	13%	7%	2%	4	4	4	4	4	4
Bromoxynil	LC (-)		%	105%	101%	98%	97%	1%	3%	8%	5%	5%	4%	4	4	4	4	4	4
Fludioxonil	LC (-)		%	108%	103%	101%	98%	3%	2%	8%	7%	4%	2%	4	5	4	4	5	4
Lufenuron	LC (-)		%	106%	105%	101%	107%	5%	7%	5%	12%	4%	7%	3	3	3	3	4	3
Mean recovery All		**101%**	**100%**	**105%**	**102%**	**100%**	**100%**	**3%**	**4%**	**8%**	**9%**	**5%**	**3%**						

GC

LC (+)

LC (-)

Mean	Orange 0.25	Orange 0.025	Apple 0.25	Apple 0.025	Lettuce 0.25	Lettuce 0.025
Rec.:	101%	100%	105%	102%	100%	100%
RSD:	3%	4%	8%	9%	5%	3%

LC-MS/MS Inter-Laboratory Validation Study (BLAPS-Working Group)

Pesticides	Mean Recovery								RSD (%)								Nr. of Laboratories that reported results (n=5 each)							
	Cucumber 0,1	Cucumber 0,01	Lemon 0,1	Lemon 0,01	Wheat Flour 0,1	Wheat Flour 0,01	Raisins 0,1	Raisins 0,01	Cucumber 0,1	Cucumber 0,01	Lemon 0,1	Lemon 0,01	Wheat Flour 0,1	Wheat Flour 0,01	Raisins 0,1	Raisins 0,01	Cucumber 0,1	Cucumber 0,01	Lemon 0,1	Lemon 0,01	Wheat Flour 0,1	Wheat Flour 0,01	Raisins 0,1	Raisins 0,01
Pesticides that showed generally good recoveries and precision																								
3,4,5-Trimethacarb	100%	98%	100%	99%	100%	101%	99%	101%	4%	6%	11%	7%	4%	9%	4%	4%	5	5	5	5	5	5	5	4
Acephate	88%	92%	89%	81%	83%	83%	83%	82%	8%	12%	10%	21%	6%	12%	7%	5%	4	4	4	4	4	4	4	3
Aldicarb	92%	97%	101%	96%	98%	97%	95%	102%	12%	5%	5%	4%	3%	12%	7%	8%	5	5	5	5	5	5	5	4
Azoxystrobin	100%	96%	97%	101%	99%	98%	100%	100%	5%	4%	4%	6%	1%	6%	4%	4%	5	5	5	5	5	5	5	4
Bendiocarb	98%	100%	105%	98%	99%	102%	100%	98%	5%	7%	4%	4%	3%	4%	3%	8%	5	5	5	5	5	5	5	4
Butocarboxim	89%	97%	99%	103%	93%	93%	96%	96%	20%	13%	6%	14%	10%	16%	3%	11%	5	4	5	5	5	5	5	4
Carbaryl	101%	100%	100%	103%	100%	103%	99%	99%	4%	8%	6%	4%	6%	10%	5%	7%	5	5	5	5	5	5	5	4
Carbendazim	94%	95%	91%	90%	88%	89%	85%	86%	4%	5%	5%	5%	3%	4%	8%	9%	5	5	5	5	5	5	5	3
Carbofuran																								
Cyprodinil																								
Dimethoat																								
Fenhexamid																								
Fenoxycarb																								
Fenpropimorph																								
Flufenoxuron																								
Imazalil																								
Imidacloprid																								
Indoxacarb																								
Iprovalicarb																								
Isoproturon																								
Linuron																								
Metalaxyl																								
Methamidophos																								
Methiocarb																								
Methomyl	99%	103%	97%	98%	95%	101%	93%	97%	3%	11%	6%	10%	9%	6%	4%	2%	5	5	5	5	5	5	5	3
Methoxyfenozid	98%	96%	100%	101%	102%	103%	98%	103%	5%	8%	4%	1%	4%	10%	5%	3%	5	5	5	5	5	5	5	4
Metolachlor	101%	97%	104%	102%	100%	104%	101%	101%	2%	5%	6%	5%	2%	6%	3%	7%	5	5	5	5	5	5	5	4
Monocrotophos	96%	93%	97%	95%	93%	97%	94%	92%	3%	7%	9%	6%	5%	6%	6%	3%	5	5	5	5	5	5	5	3
Oxamyl	100%	94%	96%	95%	98%	96%	96%	102%	4%	13%	5%	12%	7%	18%	5%	10%	5	5	5	5	5	5	5	3
Oxydemeton-methyl	94%	97%	95%	89%	91%	95%	88%	86%	5%	11%	8%	12%	4%	13%	6%	4%	5	5	5	5	5	5	5	4
Picoxystrobin	100%	98%	102%	101%	100%	103%	99%	104%	3%	6%	6%	3%	3%	8%	4%	5%	5	5	5	5	5	5	5	4
Pirimicarb	92%	93%	97%	95%	98%	100%	93%	95%	10%	7%	3%	5%	2%	5%	4%	6%	5	5	5	5	5	5	5	4
Promecarb	99%	96%						6%	5%	5%	5%	10%	2%	8%	2%	7%	5	5	5	5	5	5	5	4
Propamocarb	94%	104%						1%	10%	16%	6%	14%	6%	4%	9%	10%	5	5	5	5	5	4	5	4
Propoxur	100%	102%						9%	5%	3%	2%	3%	3%	4%	5%	8%	5	5	5	5	5	5	5	4
Pyraclostrobin	99%	100%						1%	5%	5%	4%	7%	5%	8%	4%	2%	5	5	5	5	5	5	5	4
Pyrimethanil	98%	96%						5%	3%	5%	4%	6%	2%	6%	4%	2%	5	5	5	5	5	5	5	4
Spiroxamine	96%	95%						9%	6%	8%	5%	2%	4%	6%	5%	3%	5	5	5	5	5	5	5	4
Tebuconazol	99%	99%						1%	4%	6%	4%	7%	7%	13%	6%	2%	5	5	5	5	5	5	5	4
Tebufenozid	103%	101%						6%	4%	5%	4%	5%	3%	5%	5%	3%	5	5	5	5	5	5	5	4
Thiabendazol	94%	99%						9%	6%	4%	4%	11%	4%	6%	13%	4%	5	5	5	5	5	5	5	4
Thiacloprid	101%	95%						0%	4%	6%	11%	3%	4%	7%	4%	4%	5	5	5	5	5	5	5	4
Thiofanox	89%	88%						%	31%	13%	14%	6%	7%	5%	9%	10%	5	5	5	5	5	4	5	3
Vamidothion	98%	96%	99%	99%	97%	96%	94%	91%	5%	6%	11%	7%	4%	3%	8%	6%	5	5	5	5	5	5	5	4
Mean Recovery	**97%**	**97%**	**98%**	**98%**	**96%**	**98%**	**95%**	**97%**	**7%**	**8%**	**7%**	**7%**	**5%**	**8%**	**6%**	**7%**								
2 Compounds that partly gave insufficient recovery due to degradation																								
Ethiofencarb	59%	67%	95%	95%	93%	90%	87%	90%	45%	29%	5%	9%	8%	20%	18%	15%	5	5	5	4	5	4	5	4
Pymetrozin	67%	69%	44%	44%								0%	10%	8%	41%	47%	5	5	5	3	5	4	5	3
4 sulfonyl-urea pesticides (SUs) that s... procedure was not followed in detail... final extract													ethod ... fied											
Cinosulfuron	62%	63%	70%	72%								3%	60%	75%	52%	67%	5	5	5	5	5	4	5	4
Metsulfuron-methyl	63%	64%	61%	65%								8%	59%	82%	41%	54%	5	5	5	4	5	5	5	4
Prosulfuron	71%	69%	69%	74%								5%	43%	51%	48%	61%	5	5	5	5	5	5	5	4
Thifensulfuron-methyl	65%	65%	61%	61%	41%	35%	44%	40%	34%	30%	36%	41%	71%	85%	68%	68%	5	5	5	5	5	5	5	4
Mean Recovery SUs	**65%**	**65%**	**65%**	**68%**	**44%**	**41%**	**46%**	**47%**	**31%**	**28%**	**33%**	**31%**	**58%**	**73%**	**52%**	**63%**								

Mean	Cucumber 0.1	Cucumber 0.01	Lemon 0.1	Lemon 0.01	Wheat 0.1	Wheat 0.01	Raisins 0.1	Raisins 0.01
Rec.:	97%	97%	98%	98%	96%	98%	95%	97%
RSD	7%	8%	7%	7%	5%	8%	6%	7%

Ethiofencarb was oxidized in cucumber

SUs degraded in acidified extract

LC-MS/MS Inter-Laboratory Validation Study (BLAPS-Working Group II)

Pesticides	Mean Recovery								RSD (%)								Nr. of Laboratories reported results (n=5 each)							
	Cucumber 0.1	Cucumber 0.01	Lemon 0.1	Lemon 0.01	Wheat Flour 0.1	Wheat Flour 0.01	Raisins 0.1	Raisins 0.01	Cucumber 0.1	Cucumber 0.01	Lemon 0.1	Lemon 0.01	Wheat Flour 0.1	Wheat Flour 0.01	Raisins 0.1	Raisins 0.01	Cucumber 0.1	Cucumber 0.01	Lemon 0.1	Lemon 0.01	Wheat Flour 0.1	Wheat Flour 0.01	Raisins 0.1	Raisins 0.01
Acetamiprid	101%	96%	101%	97%	97%	99%	98%	94%	3%	3%	5%	10%	2%	3%	5%	9%	5	5	4	4	4	4	5	5
Avermectin B1a	94%	96%	99%	82%	95%	97%	105%	101%	17%	6%	7%	19%	7%	11%	3%	9%	4	3	3	3	4	3	4	2
Avermectin B1b	99%	104%	113%	102%	102%	108%	95%	110%	4%	10%	19%	18%	6%	3%	5%	10%	3	3	2	2	3	2	3	2
Boscalid	102%	100%	100%	102%	97%	101%	100%	97%	2%	6%	3%	3%	9%	6%	13%	8%	5	5	4	4	4	4	5	4
Buprofezin																								
Chloridazon																								
Clofentezin																								
Cymoxanil																								
Cyproconazol																								
Demeton-S-met																								
Difenoconazol																								
Dimethachlor																								
Dimethomorph																								
Diniconazol																								
Epoxiconazol																								
Ethoprophos																								
Famoxadon																								
Fenarimol																								
Fenazaquin																								
Fenpropidin																								
Fenpyroximat	102%	99%	98%	95%	102%	102%	97%	96%	2%	5%	9%	10%	2%	10%	12%	6%	5	5	4	4	4	4	5	5
Fenthion	91%	86%	98%	88%	93%	89%	97%	97%	12%	21%	3%	13%	11%	4%	11%	8%	3	3	3	3	3	3	4	4
Flufenacet	102%	99%	100%	96%					2%	3%	3%	13%	1%	3%	6%	4%	5	5	4	4	4	4	5	5
Flurtamon	103%	100%	100%	97%					2%	4%	2%	7%	4%	6%	7%	6%	5	5	4	4	4	4	5	5
Flusilazol	104%	99%	101%	97%					4%	3%	1%	7%	1%	6%	6%	5%	5	5	4	4	4	4	5	5
Hexaconazol	101%	95%	98%	95%					3%	6%	2%	12%	6%	5%	7%	3%	5	5	4	4	4	4	5	5
Hexythiazox	103%	95%	98%	94%	101%	104%	97%	100%	4%	5%	5%	12%	3%	16%	11%	3%	5	5	4	4	4	4	5	5
Mepanipyrim	101%	93%	102%	91%	103%	100%	101%	98%	2%	5%	2%	8%	13%	7%	5%	6%	5	5	4	4	4	4	5	5
Metobromuro					98%	103%	104%	97%	2%	4%	1%	17%	7%	6%	7%	7%	5	5	4	4	4	4	5	5
Omethoat					84%	89%	85%	80%	6%	11%	6%	4%	15%	10%	9%	15%	5	4	4	3	4	3	5	5
Pirimiphos-m					106%	108%	100%	95%	3%	8%	3%	9%	10%	6%	7%	9%	5	5	4	4	4	4	5	5
Profenofos					102%	107%	100%	97%	2%	21%	4%	11%	2%	20%	8%	9%	5	5	4	4	4	4	5	5
Prometryn					101%	100%	100%	95%	2%	3%	2%	6%	1%	5%	4%	7%	5	5	4	4	4	4	5	5
Propargit					103%	105%	98%	98%	3%	4%	6%	9%	4%	6%	12%	6%	5	5	4	4	4	4	5	5
Propiconazol					100%	99%	100%	91%	3%	7%	3%	7%	3%	6%	6%	7%	5	5	4	3	4	4	5	4
Propyzamid															7%	6%	5	5	4	4	4	4	5	5
Pyrifenox	102%	97%	98%	96%											6%	4%	5	5	4	4	4	2	5	4
Pyriproxyfen	102%	102%	98%	96%											11%	7%	5	5	4	4	4	4	5	5
Quinoxyfen	101%	100%	96%	92%											13%	8%	5	5	4	4	4	4	5	5
Spinosyn A	100%	98%	100%	93%											4%	9%	5	5	4	4	4	4	5	5
Spinosyn D	98%	90%	100%	91%											5%	17%	5	5	4	4	4	4	5	5
Tebufenpyrad	102%	100%	96%	102%											5%	15%	5	5	4	4	4	4	5	5
Tetraconazol	102%	94%	100%	95%											4%	4%	5	5	4	4	4	4	5	5
Thiamethoxam	101%	95%	94%	87%	97%	95%	97%	90%	2%	5%	9%	7%	7%	5%	4%	8%	5	4	4	3	4	4	5	4
Triadimefon	103%	101%	99%	98%	95%	102%	103%	96%	4%	5%	1%	11%	14%	4%	7%	6%	5	5	4	4	4	4	5	5
Trifloxystrobin	102%	104%	99%	96%	107%	108%	102%	101%	2%	15%	4%	10%	6%	9%	6%	4%	5	5	4	4	4	4	5	5
Mean Recovery	101%	98%	99%	94%	100%	101%	100%	97%	3%	8%	7%	8%	4%	9%	5%	11%								
Carboxin	62%	49%	97%	95%	97%	99%	95%	86%	48%	74%	3%	3%	2%	4%	4%	12%	4	4	4	4	3	4	5	4
Cyromazin	41%	49%	40%	28%	22%	25%	13%	21%	23%	38%	12%	13%	40%	3%	54%	100%	4	3	2	2	3	2	3	2
Metosulam	73%	73%	80%	74%	17%	#DIV/0!	47%	#DIV/0!	46%	23%	40%	#DIV/0!	442%	#DIV/0!	70%	#DIV/0!	4	2	3	1	3	0	3	0
Quinmerac	31%	23%	23%	12%	8%	14%	17%	9%	80%	81%	53%	139%	77%	23%	48%	125%	4	4	3	2	4	3	3	3
Thiodicarb	99%	298%	98%	273%	73%	296%	98%	302%	3%	30%	2%	26%	25%	41%	7%	43%	5	3	4	3	4	4	5	4
Thiophanat-methyl	76%	1469%	99%	1989%	94%	573%	101%	2050%	37%	#DIV/0!	4%	#DIV/0!	3%	122%	9%	#DIV/0!	3	1	3	1	4	2	5	1

Mean	Cucumber 0.1	Cucumber 0.01	Lemon 0.1	Lemon 0.01	Wheat 0.1	Wheat 0.01	Raisins 0.1	Raisins 0.01
Rec.:	101%	98%	99%	94%	100%	101%	100%	97%
RSD	3%	8%	7%	8%	4%	9%	6%	11%

Oxidation

Acids, lost in PSA cleanup

Degraded in the standard solution provided

QuEChERS- Multiresidue-Method

Advantages

- ✓ **Rapid** (8 Samples in Less Than 30 min)
- ✓ **Simple** (No Laborious Steps, Minimal Sources of Errors)
- ✓ **Cheap** (~1 € Sample Prep. Materials for 1 mL Extract)
- ✓ **Low Solvent Consumption** (10 mL Acetonitrile)
- ✓ **Practically no Glassware Needed**
- ✓ **Wide Pesticide Range** (Polar, pH-Dependent Compounds)
- ✓ **Extract in Acetonitrile** (GC- and LC-Amenable)

QuEChERS.com

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QuEChERS

Since its introduction, the QuEChERS method [1] has been readily accepted by many pesticide residue analysts. Some modifications to the original QuEChERS method had to be introduced to ensure efficient extraction of pH dependent compounds (e.g. phenoxyalcanoic acids), to minimize degradation of susceptible compounds (e.g. base and acid labile pesticides) and to expand the spectrum of matrices covered.

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....the modified QuEChERS method including all presented modifications and a lot of background information is available via the internet, as well as the validation data.

QuEChERS has been used in several EU Proficiency Tests by the CVUA Stuttgart.

Participation in Proficiency Tests using the QuEChERS-Method (PDF 129 KB; opens in a new window)

F

www.quechers.com

Validation of a Simple and Rapid Multiresidue Method an its Implementation in Routine Pesticide Analysis (ZIP 1220 KB; cont.

[1] M. Anastassiades, S.J. Lehotay, D. Stajnbaher and F.J. Schenck, J AOAC Int 86 (2003) 412.

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