GENERALITAT VALENCIANA

VNIVERSITAT DE VALÈNCIA

NEW STRATEGIES FOR THE ANALYSIS OF PESTICIDES IN AMBIENT AIR

EXPOSURE AND RISK ASSESSMENT OF AIRBORNE PESTICIDES

European Workshop on Pesticides in Ambient Air

VICENT YUSA (yusa_vic@gva.es

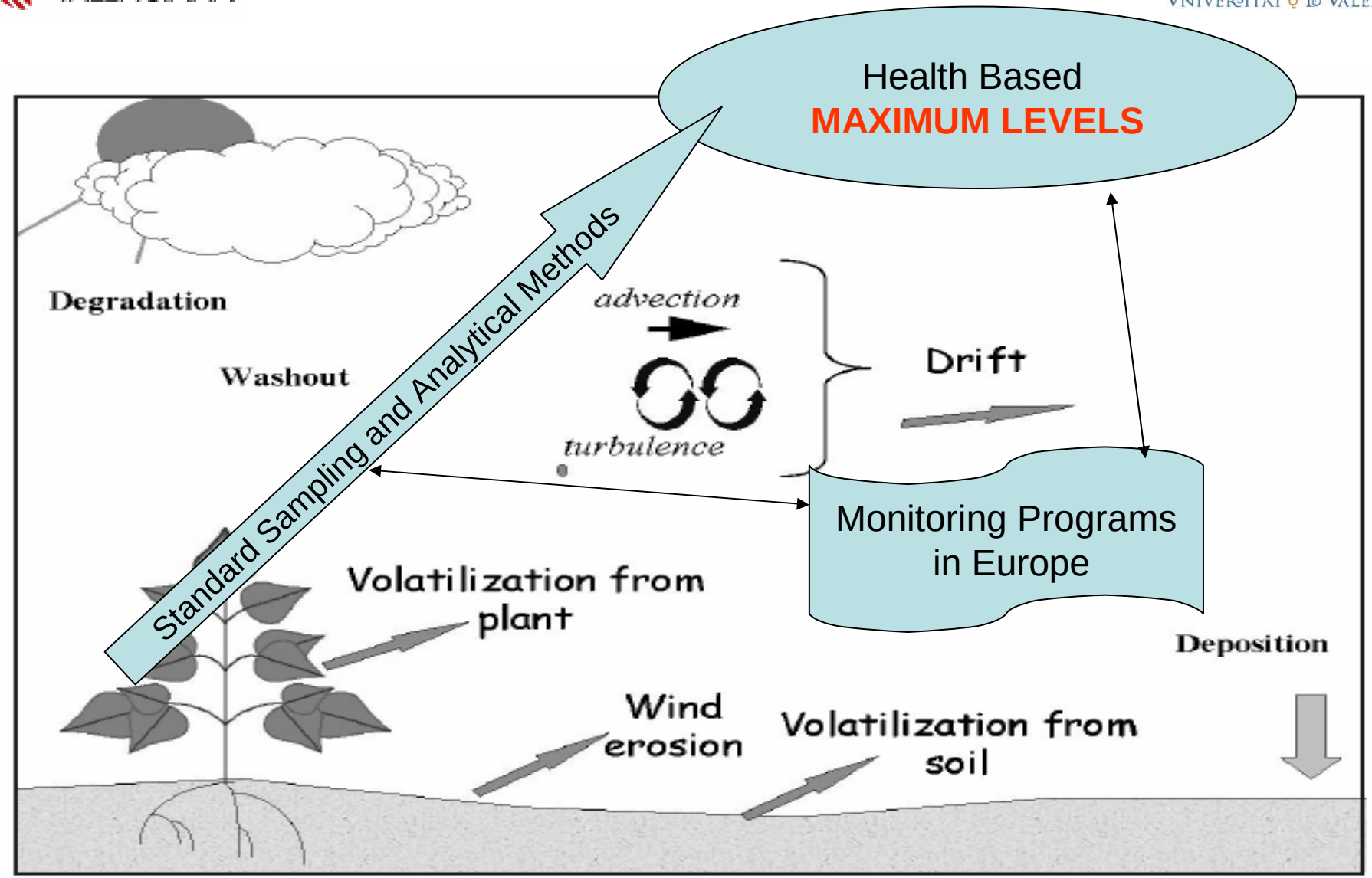

Figure 2. Processes involved in the transfer of pesticides to the atmosphere.

Carole BEDOS[1] Agronomie 22 (2002) 35–49

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HOW MANY PESTICIDES DO WE NEED TO CONTROL IN AMBIENT AIR?

Number of pesticides authorized and applied in EU (2010): ~350

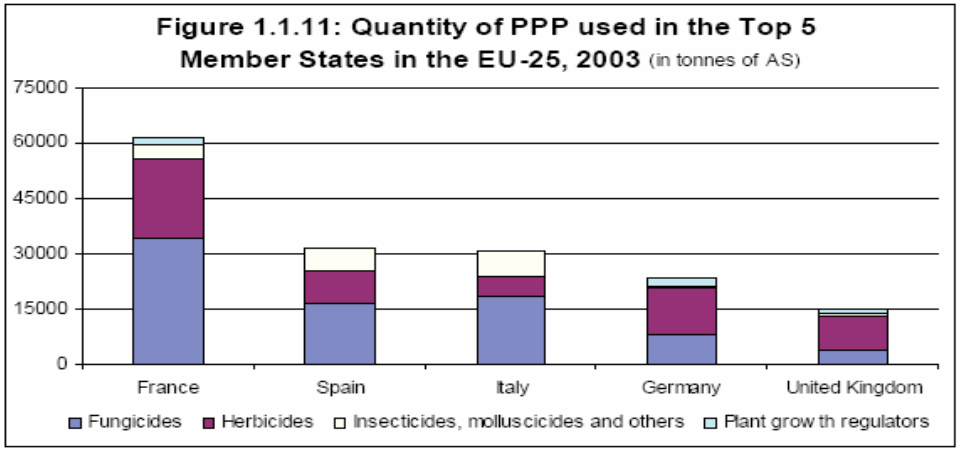
- **Herbicides: 127**
- **Fungicides: 118**
- **Insecticides: 38**
- **Others: 67**

Recommended for use (CV)	
Herbicides	12
Acaricides	10
Fungicides	60
Inecticides	45
TOTAL	**127**

eurostat

The use of plant protection prod in the European Union

Data 1992-2003



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Inventory of pesticide emissions into the air in Europe

$$ER_{ijk} = D_{ijk} + E_{app,air,ijk} + E_{crop,air,ijk}$$

Table S.3.5a Emissions of AS applied to ***vineyards*** (in kg/yr of AS) for 2000.

Active substances	AT	BE	DK	FI	FR	DE	GR	IE	IT	LU	NL	PT	ES
sulphur	134542	0	0	0	6766705	722494	859849	0	3351528	0	0	3644048	4333048
glyphosate	6940	0	0	0	0	40208	30576	0	233309	0	0	147834	142650
mancozeb	0	0	0	0	482661	0	19617	0	1092042	0	0	83873	122876
propineb	0	0	0	0	0	0	18570	0	0	0	0	108518	0
amitrol	0	0	0	0	0	0	9796	0	0	0	0	0	0
copper oxychloride	0	0	0	0	0	0	0	0	117360	0	0	0	0
copper	0	0	0	0	0	0	0	0	114236	0	0	0	0
fosetyl	0	0	0	0	531511	0	0	0	0	0	0	72656	0
1,3-dichloropropene	0	0	0	0	0	0	0	0	0	0	0	0	392079
folpet	15440	0	0	0	575090	98403	0	0	0	0	0	0	30415
metiram	0	0	0	0	314409	137008	0	0	0	0	0	0	0
tolylfluanid	0	0	0	0	0	34894	0	0	0	0	0	0	0
chlorpyrifos	0	0	0	0	0	0	0	0	0	0	0	0	0
dinocap	0	0	0	0	0	0	0	0	0	0	0	0	0
myclobutanil	0	0	0	0	0	0	0	0	0	0	0	0	0
dimethoate	0	0	0	0	0	0	0	0	0	0	0	0	0
chlorothalonil	5476	0	0	0	0	0	0	0	0	0	0	0	0
dithianon	2919	0	0	0	0	0	0	0	0	0	0	0	0

AT: Austria, **BE**: Belgium, **BG**: Bulgaria, **CZ**: Czech Republic, **DE**: Germany, **DK**: Denmark, **EE**: Estonia, **ES**: Spain, **FI**: Finland, **FR**: France, **GR**: Greece, **HU**: Hungary

130 priority substances in five major crops (cereals., maize, oilseeds, fruit threes, vineyards)

Priority score =Hazard Factor X Kg of Use

D.A. Sarigiannis

Atmospheric Environment 75 (2013) 6–14

HOW MANY METABOLITES DO WE NEED TO CONTROL IN AMBIENT AIR?

METABOLITES/DEGRADATION-TRANSFORMATION PRODUCTS/BREAKDOWN PRODUCTS

2. Relevant metabolite: a metabolite for which there is reason to assume that it has comparable intrinsic properties as the active substance in terms of its biological target activity, or that it has certain toxicological properties that are considered severe and unacceptable with regard to the decision-making criteria described in the text. Such a metabolite is therefore treated like the parent active substance in the assessment according to Annex VI, point C.2.5.1.2 of Directive 91/414/EEC. Where such a metabolite exceeds

DEGRADATION/TRANSFORMATION PRODUCTS

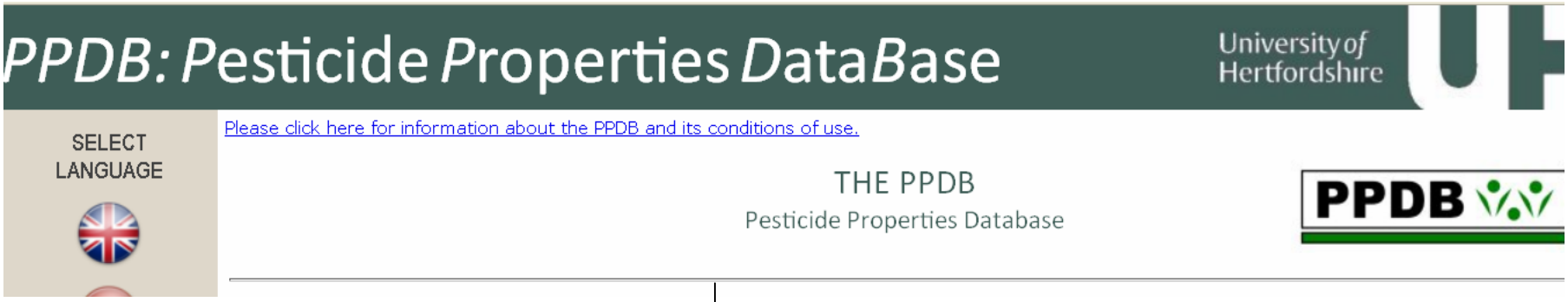
523 SOIL METABOLITES were identified from 185 CUP in UK

67 metabolites could exhibit the parent pesticidal mode of action
(57 selected for further study)

Pesticide metabolite
cis-3-chloroprop-2-enoic acid
trans-3-chloroprop-2-enoic acid
aldicarb sulfone
aldicarb sulfoxide
sulfanilamide
deethylatrazine
reference compound 10
2-aminobenzimidazole
carbofuran
carboxin sulfoxide
5-amino-4-chloropyridazin-3(2H)-one
3-carbamyl-1,2,4,5-tetrachlorobenzoic acid
3-carbamyl-2,4,5-trichlorobenzoic acid
3-cyano-6-hydroxy-2,4,5-trichlorobenzamide
R417888
3-(3-chloro-p-tolyl)-1-methylurea
cyanazine acid
AE 0542291
5-hydroxy-XDE-570
5-trifluoromethyl-pryid-2-one
FOE oxalte
thiadone
ethanol
phosphorous acid
aminomethylphosphonic acid
1-(6-chloro-pyridine-3-ylmethyl)-N-nitro guanidine
AE F145740
metsulfuron-methyl
3,5-di-iodo-4-hydroxybenzamide
3,5-di-iodo-4-hydroxybenzoic acid
3-[4-(2'-hydroxy-2'-propyl)-phenyl]-methyl urea
desmethylisoproturon
acetaldehyde
metazachlor oxalic acid
metazachlor sulfonic acid
methiocarb sulfoxide
ATSA
demethyl metoxuron
diketo metribuzin
IN-D5119
IN-D5803
dimethyloxamic acid
2,6-dinitro-3,4-xylidine
4-[(1-ethylpropyl)amino]-2-methyl-3,5-dinitro benzyl alcohol
4-[(1-ethylpropyl)amino]-3,5-dinitro-o-toluic acid
4-fluoroaniline
CGA 294849
BH518-2
BH518-4
deisopropylatrazine
methomyl
diisopropylamine
2-ethyl-7-nitro-5-(trifluoromethyl) benzimidazole

C. Sinclair et al., 2010

METABOLITES



eg. CARBENDAZIME

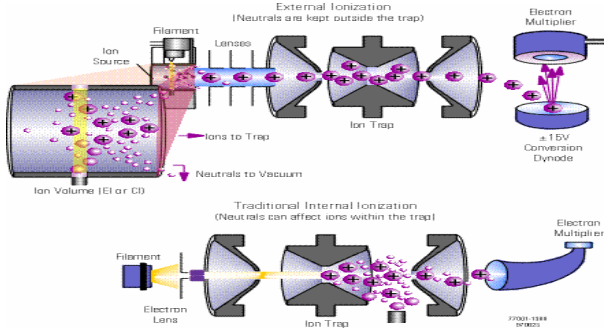
Key metabolites:

Metabolite	Formation medium	Estimated maximum occurrence fraction	91/414 relevancy
2-aminobenzimidazole	Soil	0.08	Minor fraction, Relevant

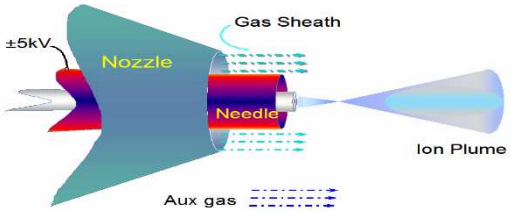
ANALYTICAL METHODS

DETERMINATION

GC-MS; GC-MS/MS



LC-MS/MS



Newly introduced pesticides are:

- **more polar**
- **less toxic**
- **more degradable in ambient air**

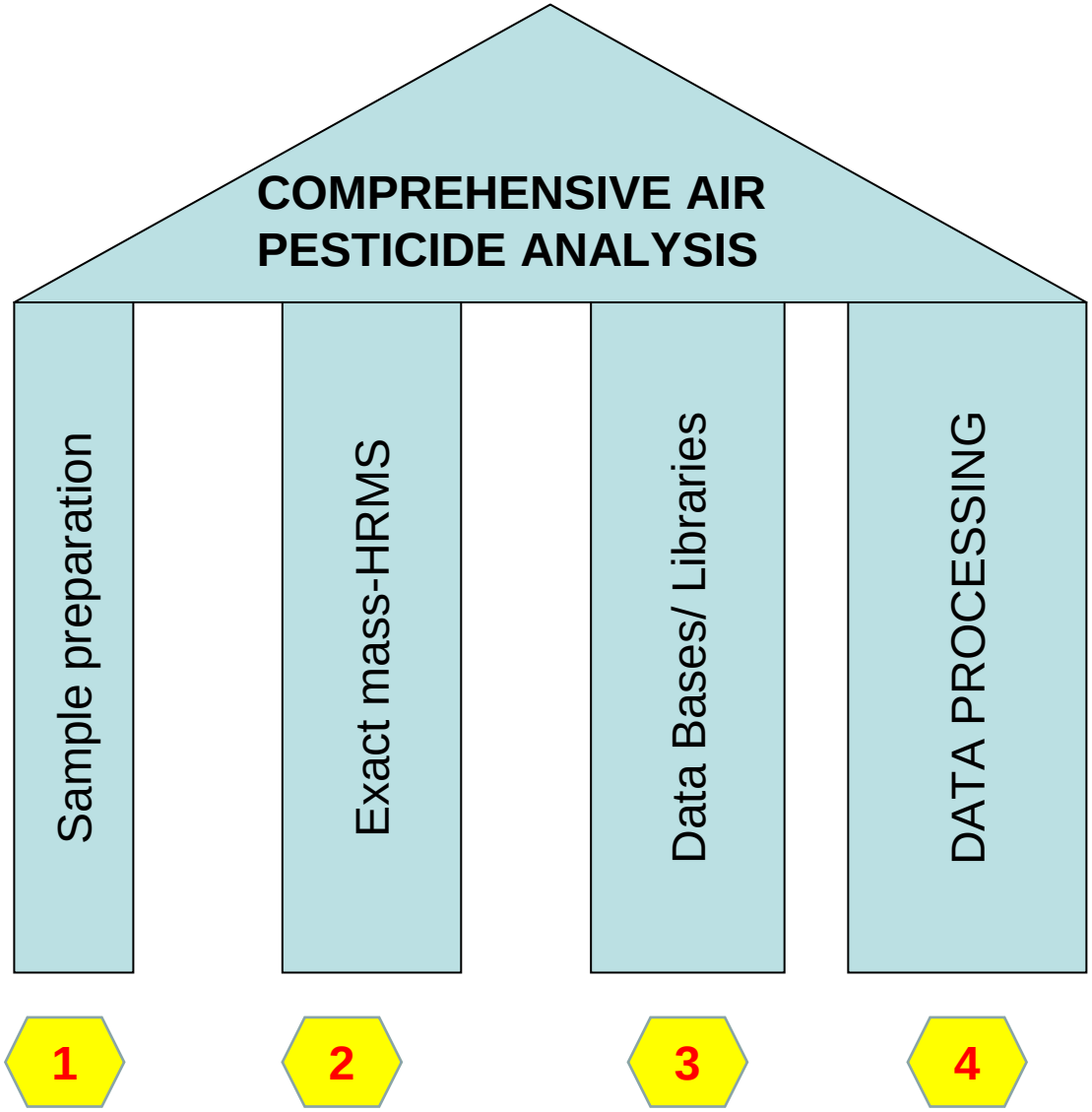
Why do we need HRMS ?

© Anton Kaufmann

Why do we need HRMS ?

© Anton Kaufmann

COMBINED TARGET/NON TARGET STRATEGIES FOR MASSIVE ANALYSIS OF PESTICIDES IN AIR USING LC-ACCURATE MASS- HRMS


COMPREHENSIVE AIR PESTICIDE ANALYSIS
Sample preparation
Exact mass-HRMS
Data Bases/ Libraries
DATA PROCESSING
1
2
3
4

GENERALITAT VALENCIANA

1

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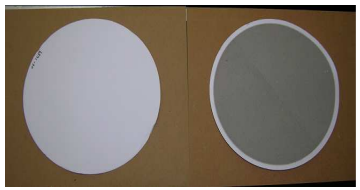
GENERIC METHOD OF EXTRACTION

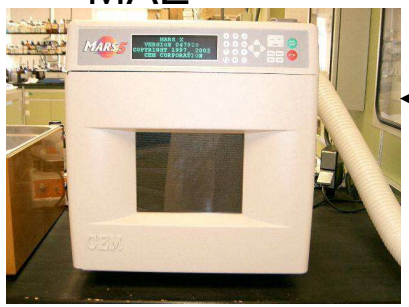
SAMPLES



filters

adsorbent

MAE



50ºC/20’
1200W
30 mL Ethyl Acetate

ASE

MATRIX EFFECT (ME) (%) = (B/A)*100 ;

B= sample solution; A= standard solution

High: ME= 23-59 %

Moderate: ME=61-80 %

Low: ME=83-90 %

2

Orbitrap Exactive

Exact mass

< 1 ppm en each scan

m/z = 200
m/z= 200.1457 // 200.1455

Resolution

1000.000 max

Dinamic Range

>10^4

Collision cell HCD

All ions fragmentation

Others

Mass range: 50-4000

Up to 10 scans/s

Polarity switching (1s)

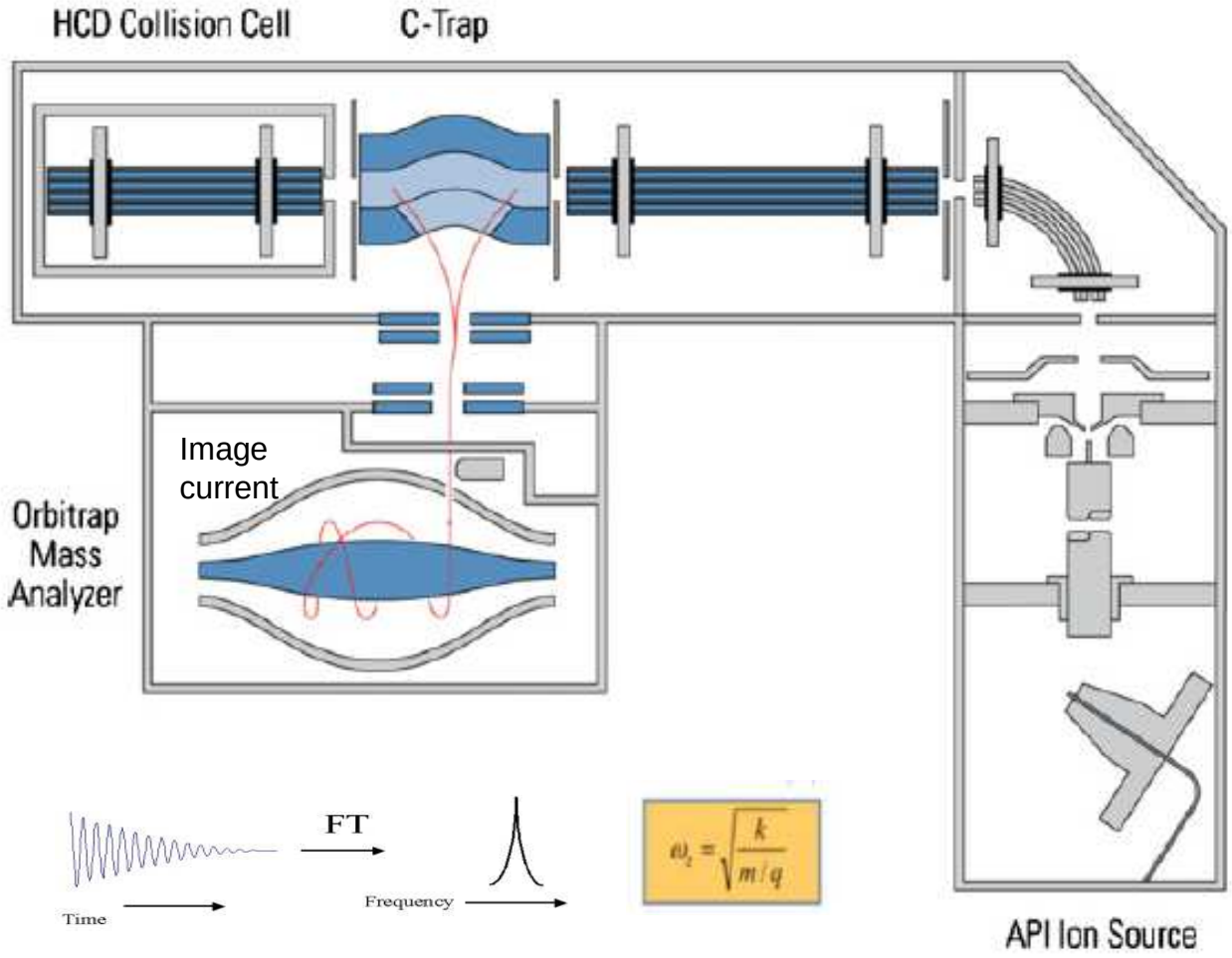


Fig. 4. Schematic layout of the Exactive™ mass spectrometer (including an optional HCD collision cell).

WHY DO I NEED HIGH RESOLUTION?

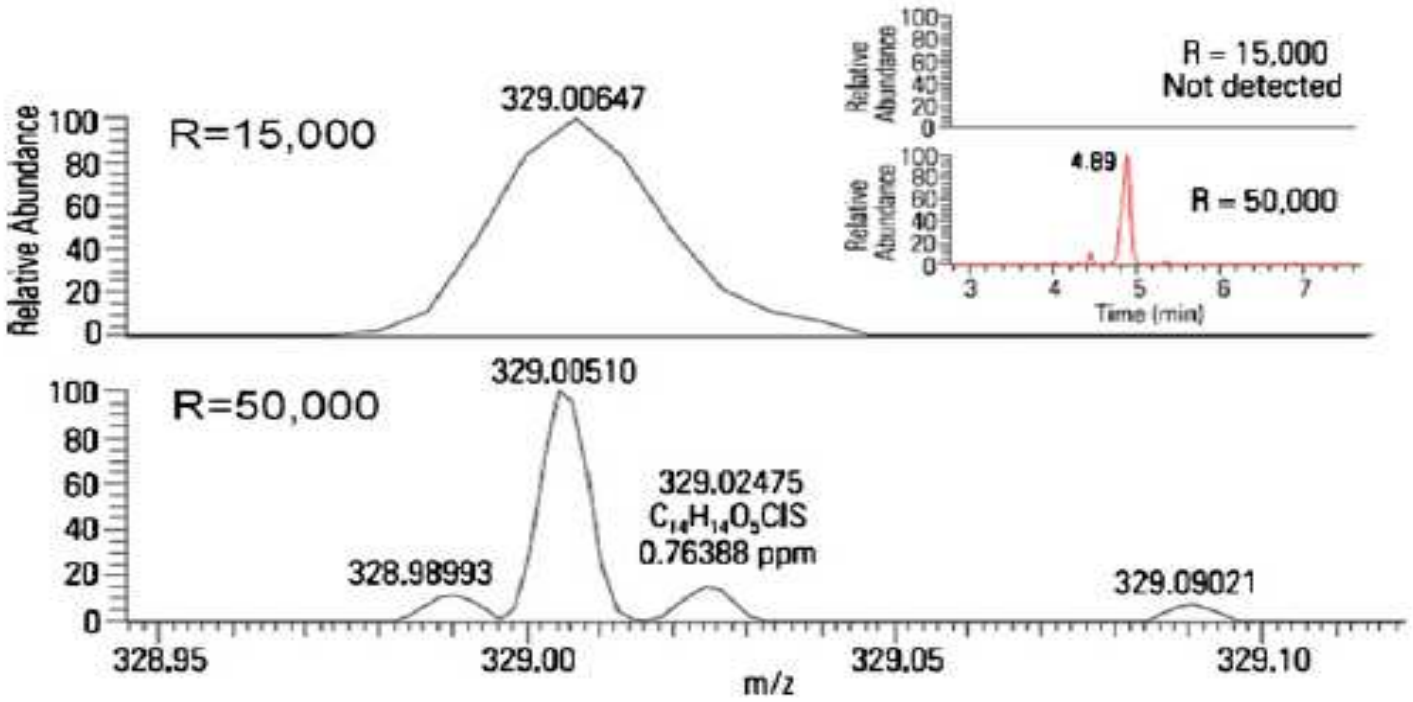
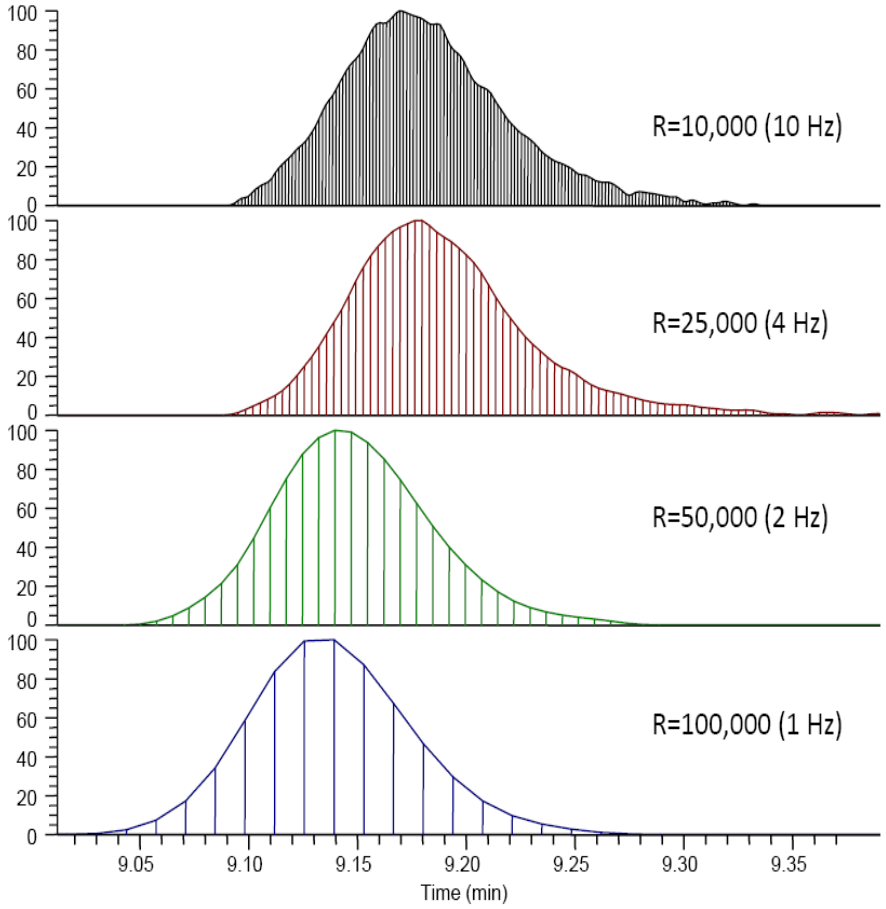


Fig. 3. High resolution prevents a false negative result. Pesticide Sulcotrion™ (*m/z* 329.02475) was measured in a mixture with other 115 pesticides and food toxins in a horse feed matrix. The mass deviation at a resolution of 15,000 is higher than 5 ppm extraction window set by the user due to the presence of an interference (top pane) giving a false negative result (insert, top trace). Sulcotrion can be detected with mass deviation of less than 1 ppm at 50,000 resolution (bottom pane) leading to a confident identification and quantitation (insert, bottom trace).
The figure courtesy of Markus Kellmann, Thermo Fisher Scientific.

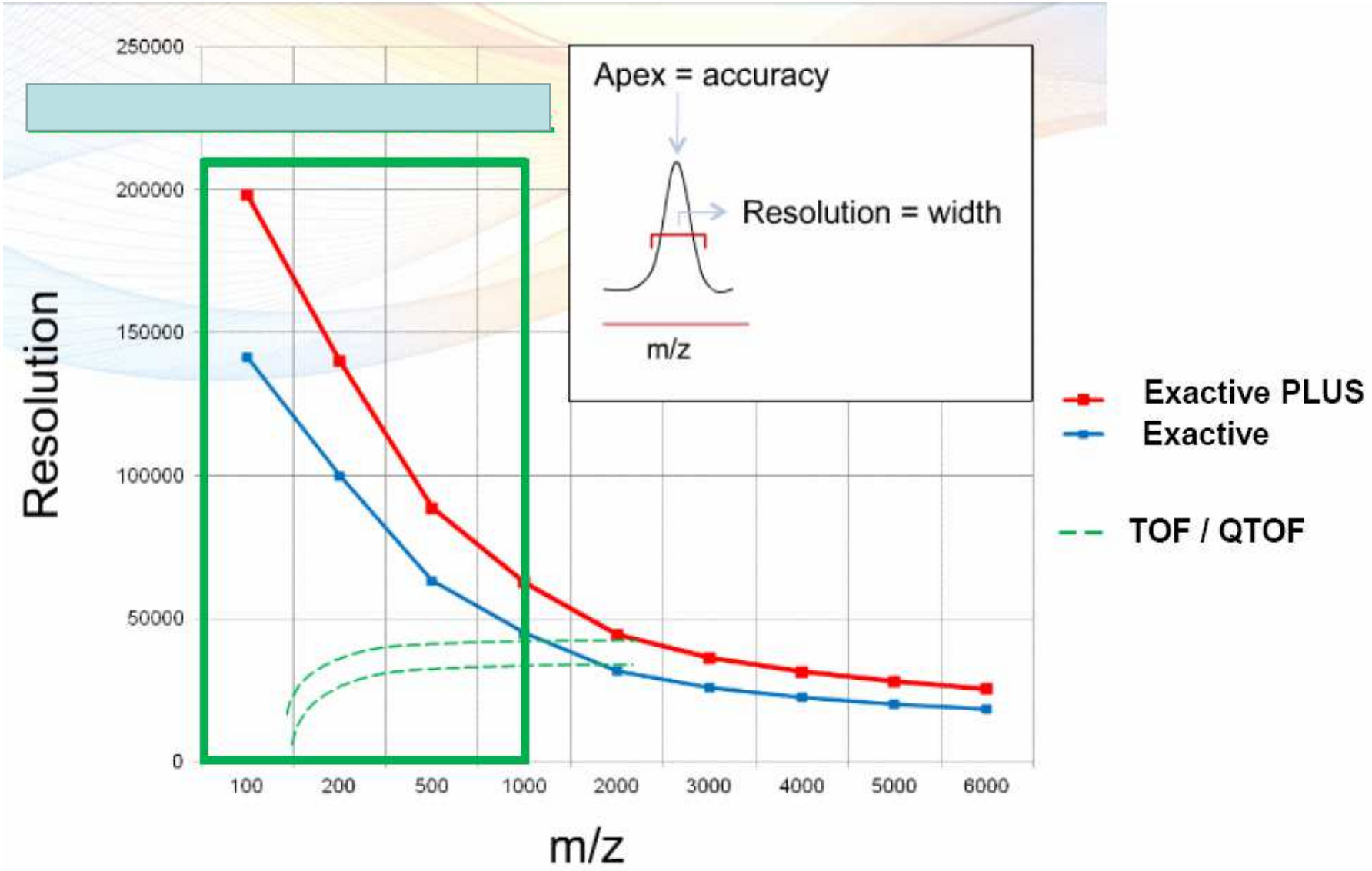
A. Makarov, M. Scigelova / J. Chromatogr. A 1217 (2010) 3938–3945

HOW MUCH RESOLUTION DO I NEED?



J Am Soc Mass Spectrom 2009, 20, 1464–1476

Orbitrap Exactive



GENERIC METHOD OF EXTRACTION

LC-HRMS

Target DB
(prioritary pesticides)

Post-run target DB
(~600 pesticides+ metabolites)

Validated quantitative method

Quantification

- Linearity
- Matrix effect
- Recovery
- RSD
- LOQ

Confirmation

- ≥ 2 diagnostic ions
- Mass accuracy < 5ppm
- t_R (±0.2 min)
- Product ion (HCD) < 5ppm
- RIA (relative isotopic abundance)
  (±30%, relative)

Screening method (without SDL)

Based on:

- Mass accuracy < 5ppm
- Product ion (HCD) < 5ppm
- RIA (relative isotopic abundance)

SANCO/12571/2013

DATA BASE (csv) OF PESTICIDES AND OTHER SUBSTANCES

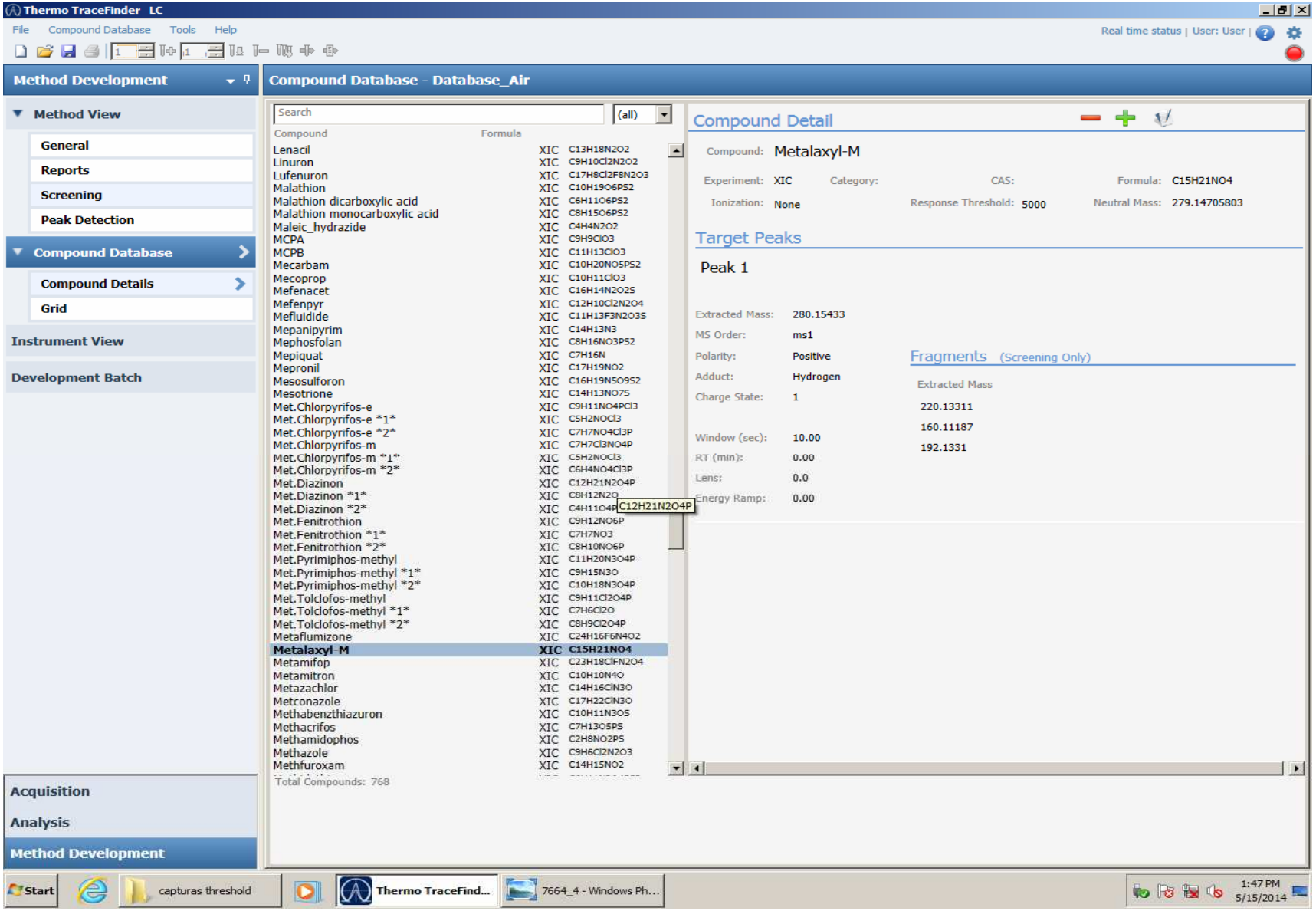
Microsoft Excel - database air33

Archivo Edición Ver Insertar Formato Herramientas Datos Ventana ?

M14

	A	B	C	D	E	F	G	H	I	J
1	TraceFinder Compound	Schema Version 1								
2				PEAK 1						
3	CompoundName	ExperimentType	ChemicalFormula	ResponseThr	ExtractedMas	Adduct	Polarity	Fragment	Fragment	Fragment
4	Bupirimate	XIC	C13H24N4O3S	5000	317.16419	M+H	+	166.0975	108.0192	210.1603
5	Buprofezin	XIC	C16H23N3OS	5000	306.16346	M+H	+	201.1056	116.0528	
6	Butachlor	XIC	C17H26ClNO2	5000	312.17248	M+H	+			
7	Butafenacil	XIC	C20H18ClF3N2O6	5000	475.08782	M+H	+			
8	Butamifos	XIC	C18H38NO3P	5000	348.26621	M+H	+			
9	Butocarboxim	XIC	C7H14N2O2S	5000	191.08487	M+H	+			
10	Butoxycarboxim	XIC	C7H14N2O4S	5000	223.0747	M+H	+	86.06	106	166
11	Butralin	XIC	C14H21N3O4	5000	296.16048	M+H	+	240.0968	222.0863	
12	Butylate	XIC	C11H23NOS	5000	218.15731	M+H	+			
13	Cadusafos	XIC	C10H23O2PS2	5000	271.09498	M+H	+	158.9698	96.9508	130.9385
14	Cafenstrole	XIC	C16H22N4O3S	5000	351.14854	M+H	+			
15	Captafol	XIC	C10H9Cl4NO2S	5000	347.91809	M+H	+			
16	Captan	XIC	C9H8Cl3NO2S	5000	299.94141	M+H	+			
17	Carbaryl	XIC	C12H11NO2	5000	202.08625	M+H	+	145.0647	117.0699	155.0491
18	Carbendazim	XIC	C9H9N3O2	5000	192.07675	M+H	+	160.0511		
19	Carbetamide	XIC	C12H16N2O3	5000	237.12337	M+H	+	120.0444	118.0863	100.0757
20	Carbofuran	XIC	C20H32N2O3S	5000	381.22064	M+H	+	165.091	123.0441	
21	Carbophenothion	XIC	C11H16ClO2PS3	5000	342.98113	M+H	+			
22	Carbosulfan	XIC	C20H32N2O3S	5000	381.22064	M+H	+	118.0685	128.1434	
23	Carboxin	XIC	C12H13NO2S	5000	236.07398	M+H	+	143.0161		
24	Carfentrazone-ethyl	XIC	C15H14Cl2F3N3O3	5000	412.04371	M+H	+	316.0028	345.9956	
25	Carpropamide	XIC	C15H18Cl3NO	5000	334.05267	M+H	+	139.0324		
26	Cartap	XIC	C13H7Br2N3O6	5000	238.06785	M+H	+			
27	Carvone	XIC	C10H14O	5000	151.11175	M+H	+	109	123	
28	Chinomethionat	XIC	C10H6N2OS2	5000	234.99943	M+H	+	207.0043	163.0325	
29	Chloralose	XIC	C8H11Cl3O6	5000	308.9694	M+H	+			
30	Chloramben	XIC	C7H5Cl2NO2	5000	205.97701	M+H	+			
31	Chloramben *1*	XIC	C7H5Cl2NO2	5000	205.97701	M+H	+			
32	Chlorantraniliprole	XIC	C18H14BrCl2N5O2	5000	481.97807	M+H	+	285.9179		
33	Chlorbenthiazone	XIC	C8H6ClNOS	5000	199.99314	M+H	+			
34	Chlorbromuron	XIC	C9H10BrClN2O2	5000	292.96869	M+H	+			
35	Chlorbromuron *1*	XIC	C9H10BrClN2O2	5000	292.9687	M+H	+			
36	Chlorbufam	XIC	C11H10ClNO2	5000	224.04728	M+H	+			
37	Chlordane	XIC	C10H6Cl8	5000	406.80805	M+H	+			
38	Chlorfenapyr	XIC	C15H11BrClF3N2O	5000	406.97681	M+H	+			
39	Chlorfenprop	XIC	C9H8Cl2O2	5000	218.99741	M+H	+			
40	Chlorfenvinphos	XIC	C12H14Cl3O4P	5000	358.9768	M+H	+			
41	Chlorfluazuron	XIC	C20H9Cl3F5N3O3	5000	539.97024	M+H	+	158.0412	141.0146	
42	Chlorflurenol	XIC	C14H9ClO3	5000	261.0313	M+H	+			
43	Chloridazon	XIC	C10H8ClN3O	5000	222.04287	M+H	+	104.0495		
44	Chlormephos	XIC	C5H12ClO2PS2	5000	234.97776	M+H	+			

DATA BASE OF PESTICIDES AND OTHER SUBSTANCES



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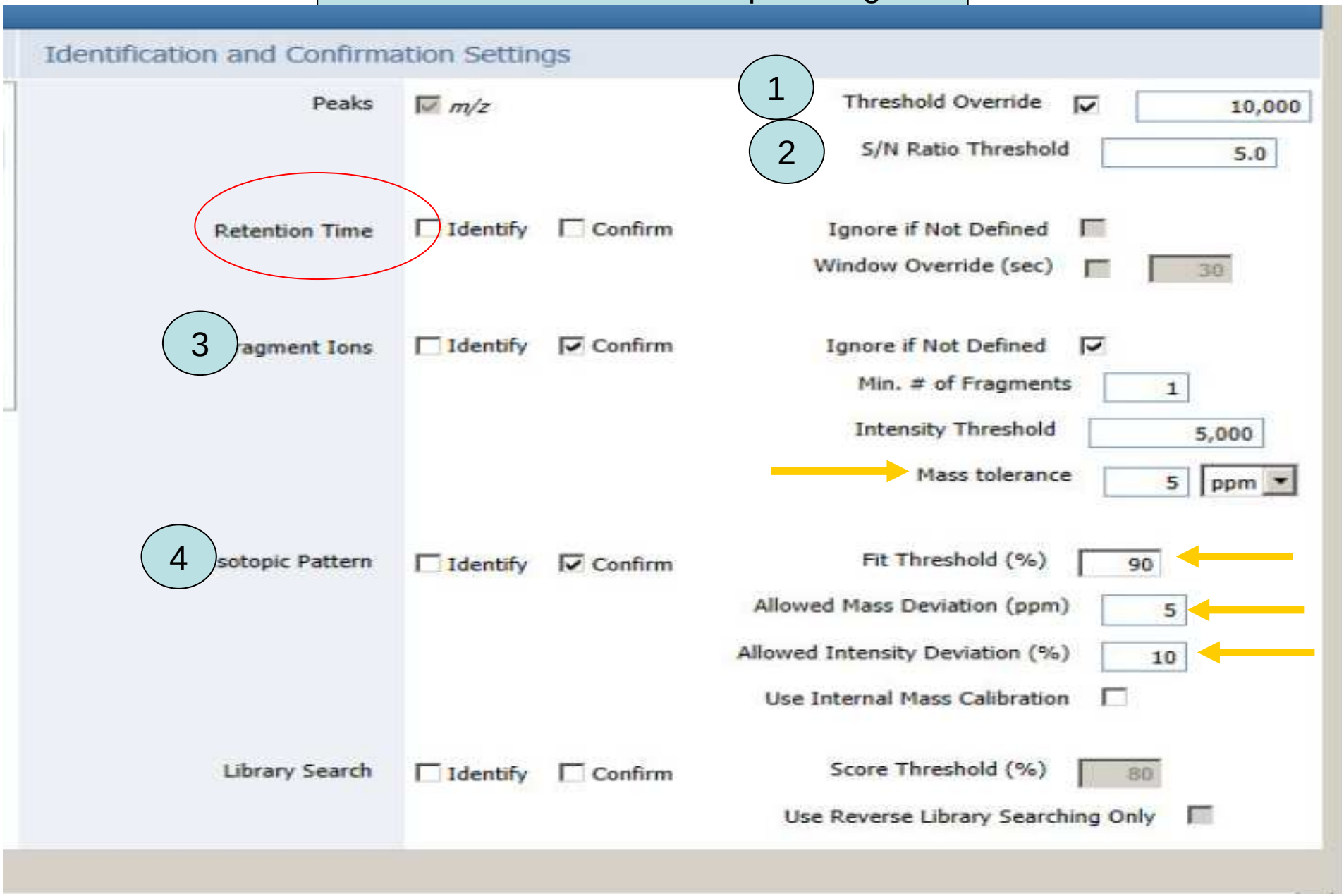
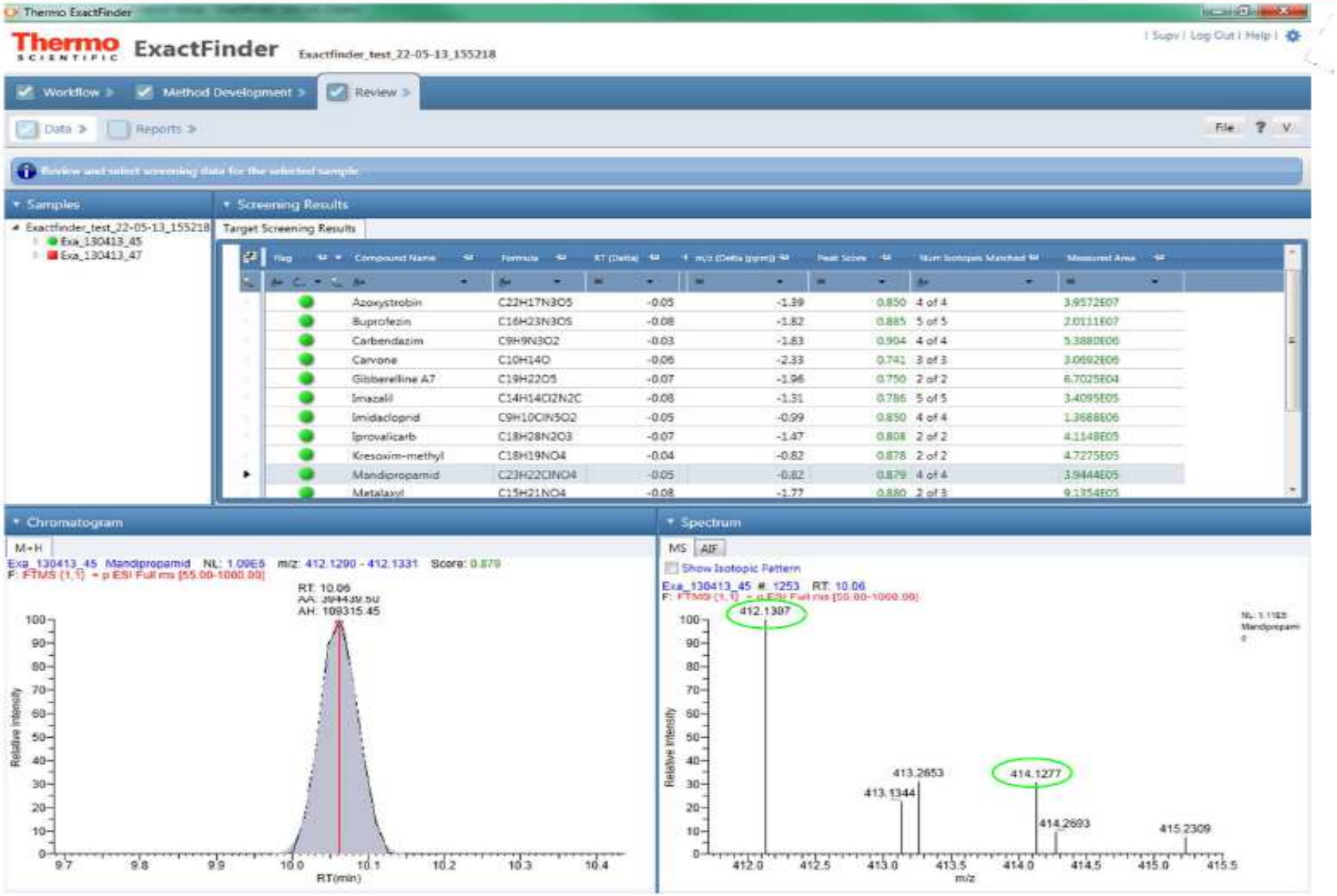
DETECTION CRITERIA post-target

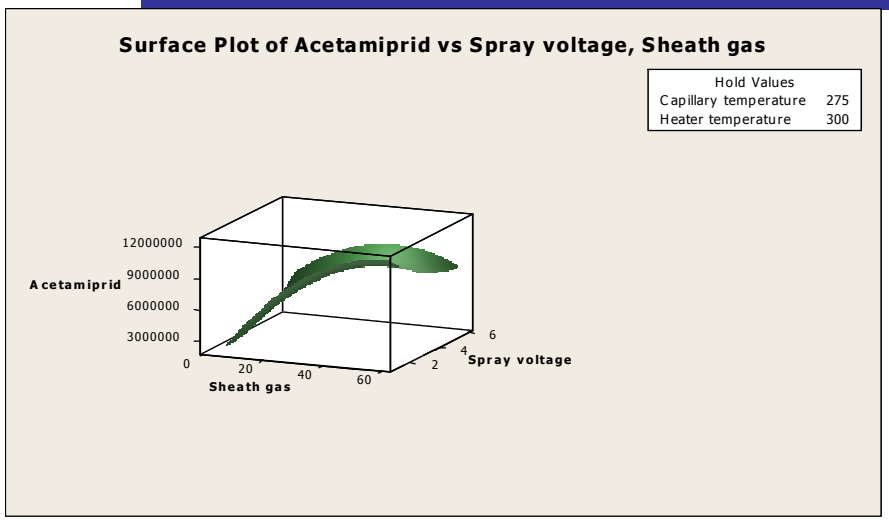


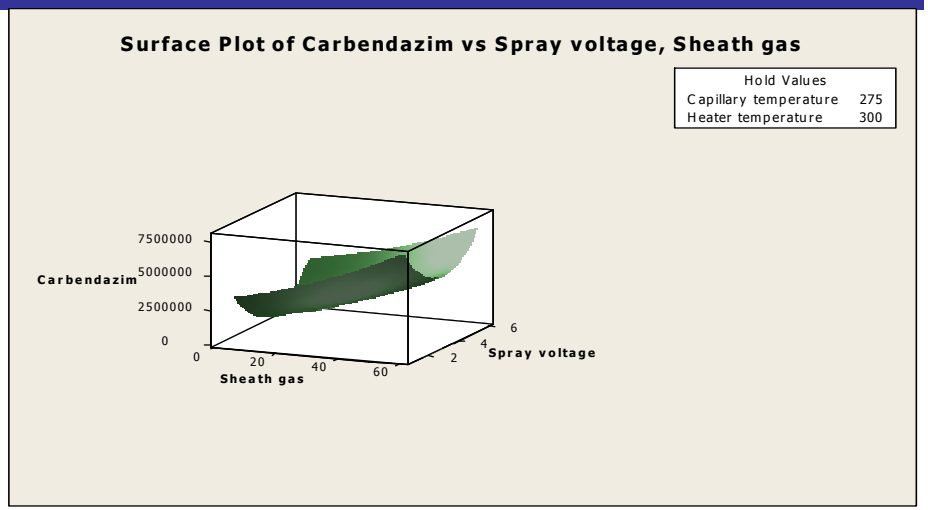
Result review



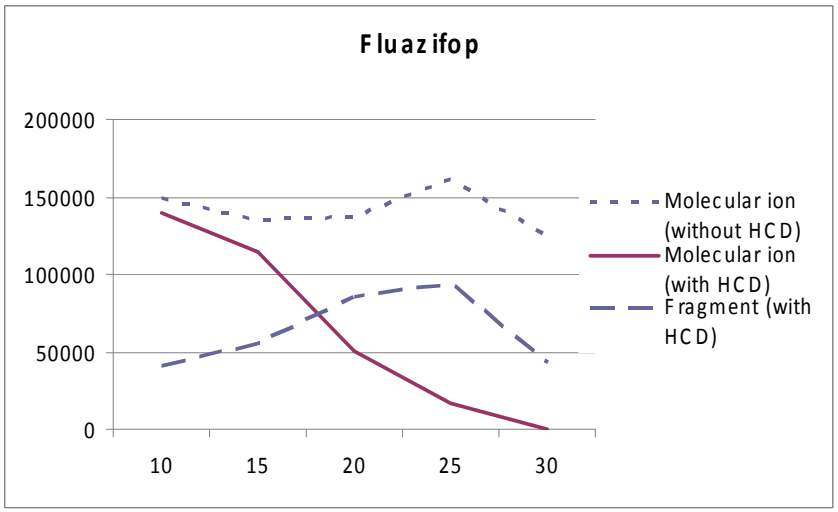
METHOD DEVELOPMENT

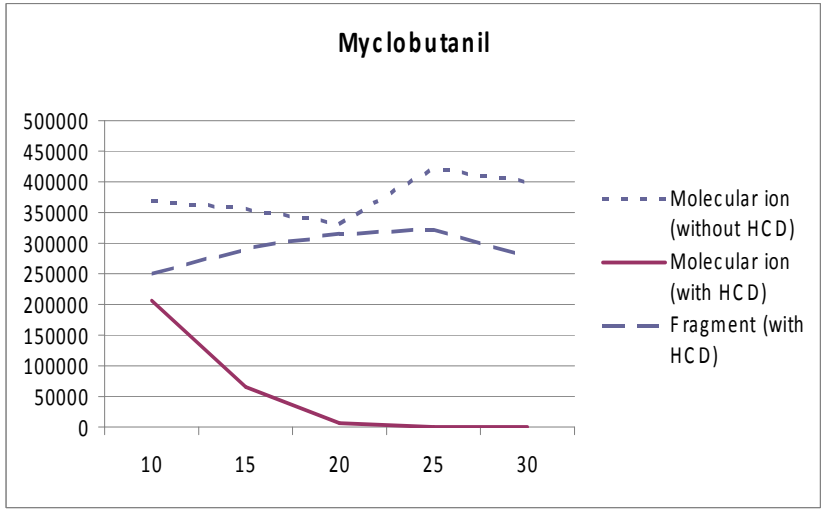
OPTIMIZATION OF ION SOURCE SETTINGS (DoE)





STUDY OF HCD FRAGMENTATION





PERFORMACE OF THE QUANTITATIVE METHOD

Linearity : 5 and 100 ng mL^{-1} in vial

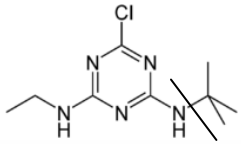
Matrix effect: 23-90% → (matrix matched calibration)

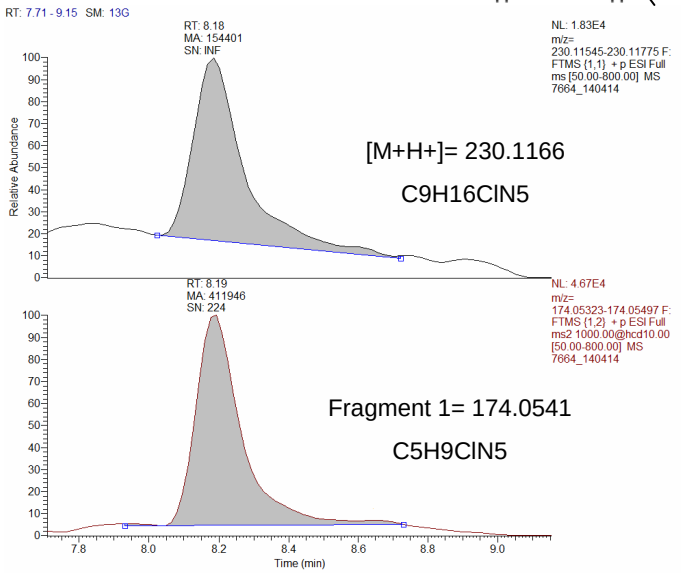
Recovery: 73-113 % (20ng/filter)

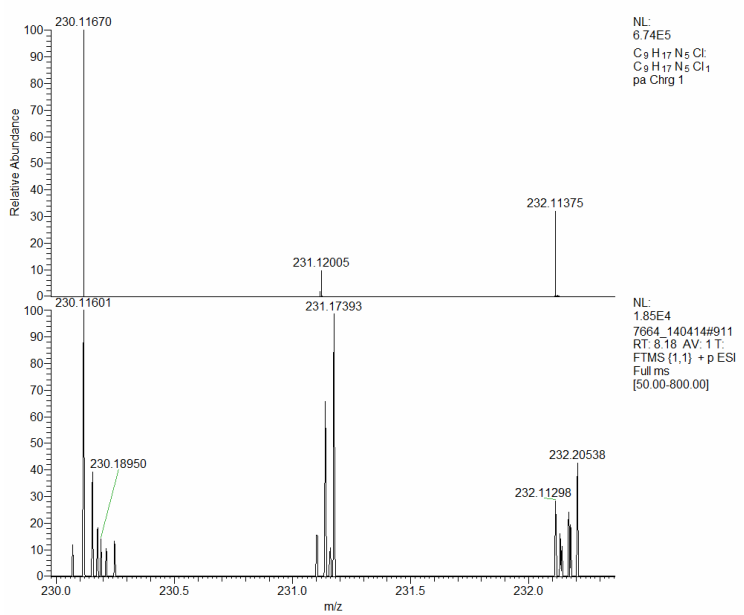
RSD: 6-25% (20ng/filter)

LOQ: 6.5 pg/m^3 -75 pg/m^3 (when 760 m^3 where collected)

TERBUTHYLAZINE

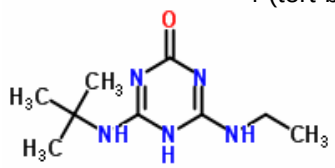


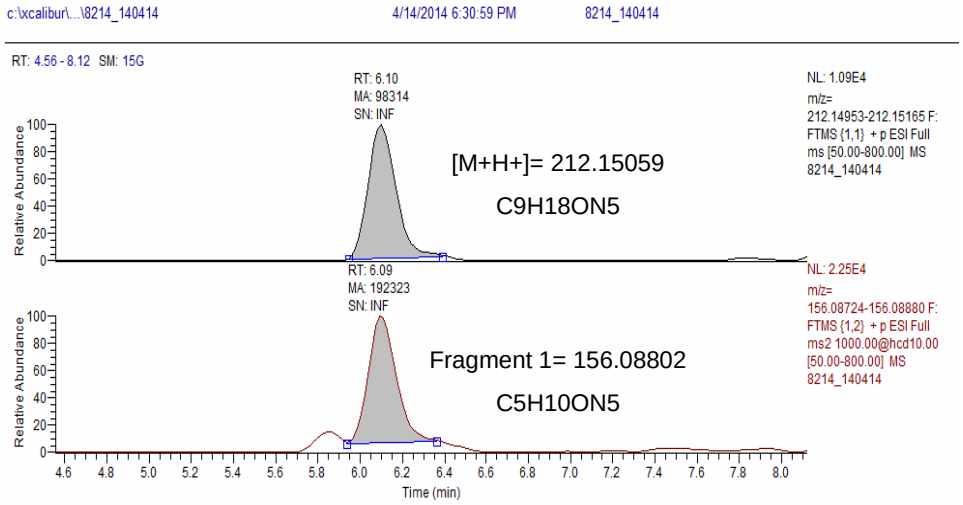


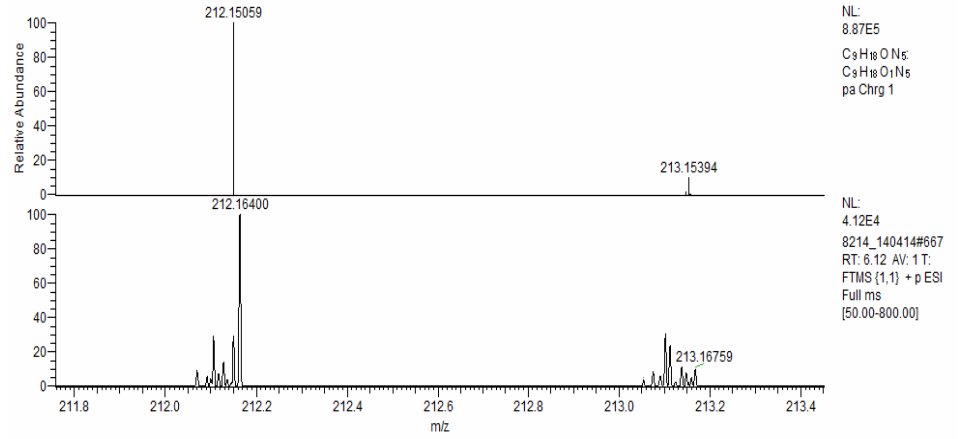


hydroxy-terbuthylazine

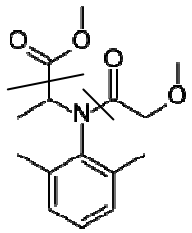
4-(tert-butylamino)-6-(ethylamino)-1,3,5-triazin-2(1H)-one

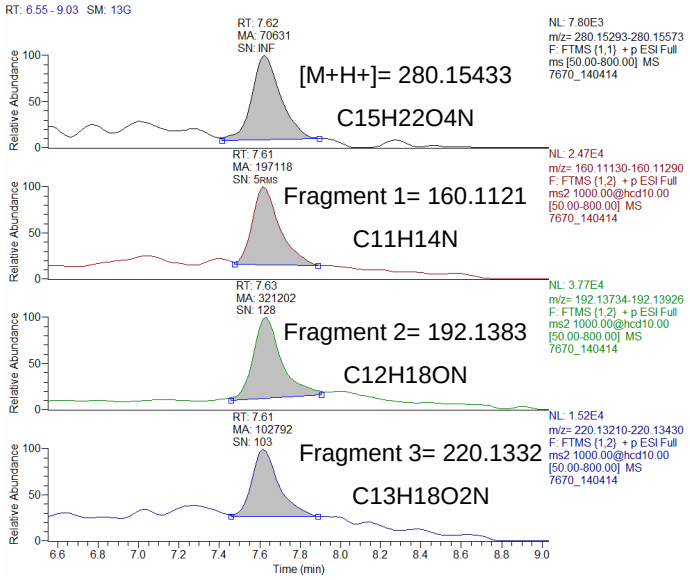


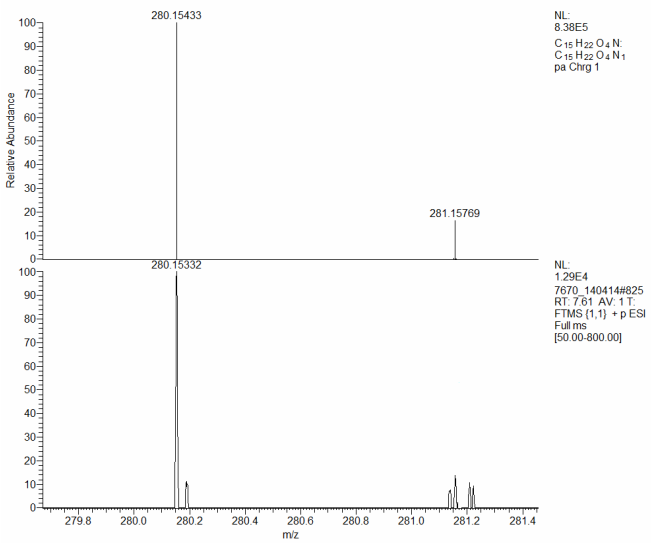




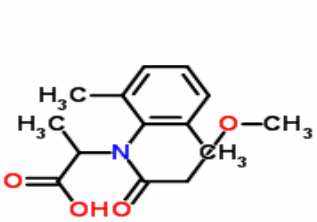
METALAXYL

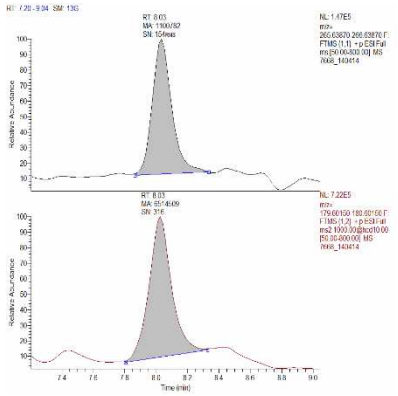






N-(2,6-dimethylphenyl)-N-(methoxyacetyl)alanine



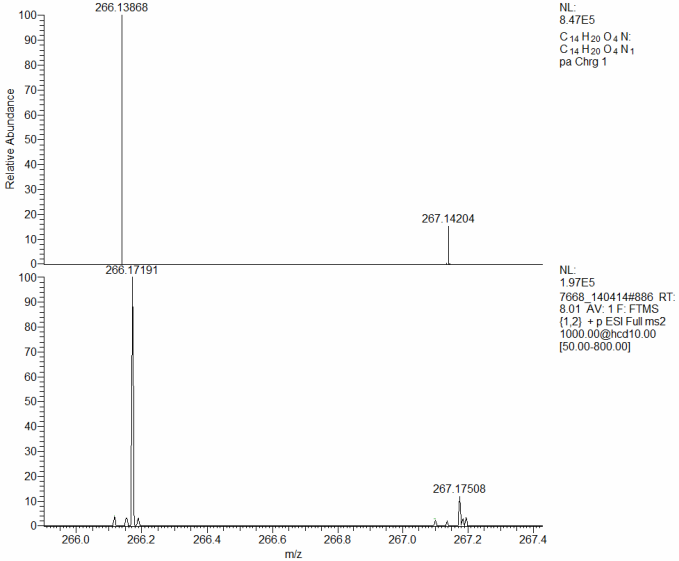


[M+H+]= 266.1387

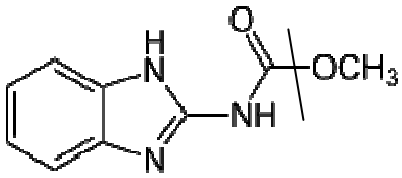
C14H20O4N

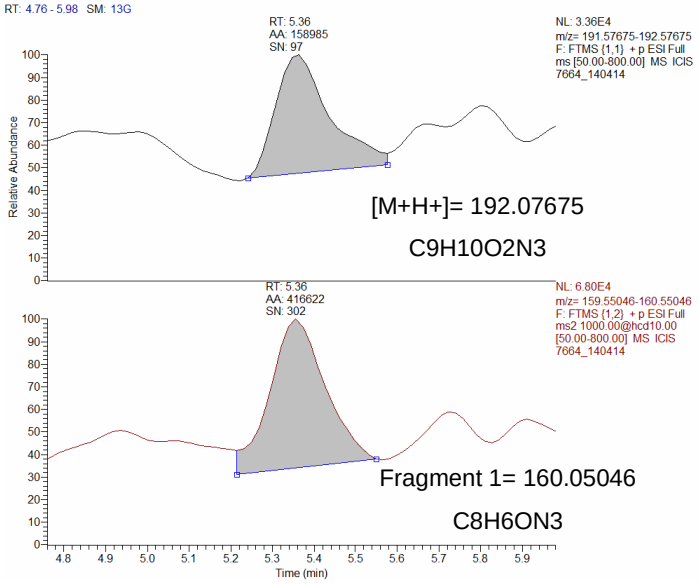
Fragment 1= 180.1015

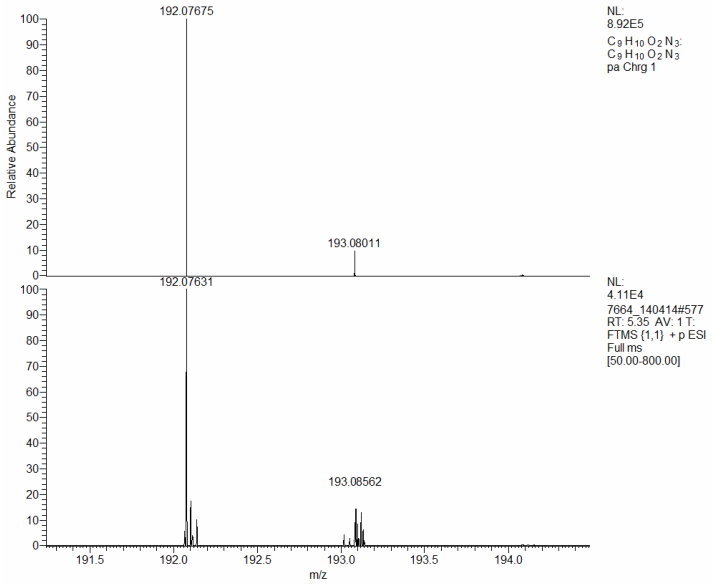
C10H12O3



CARBENDAZIM

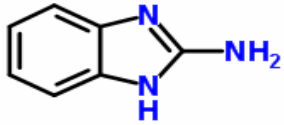


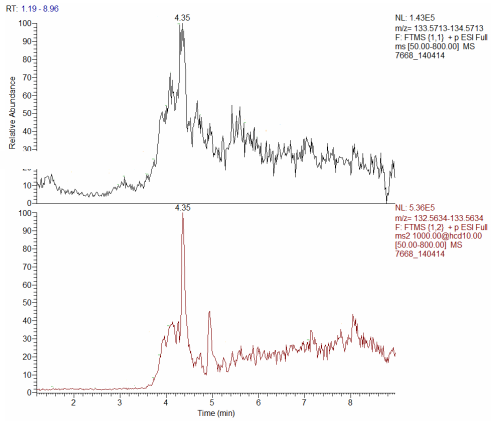




2-AMINOBENZIMIDAZOLE

1H-Benzimidazol-2-amine



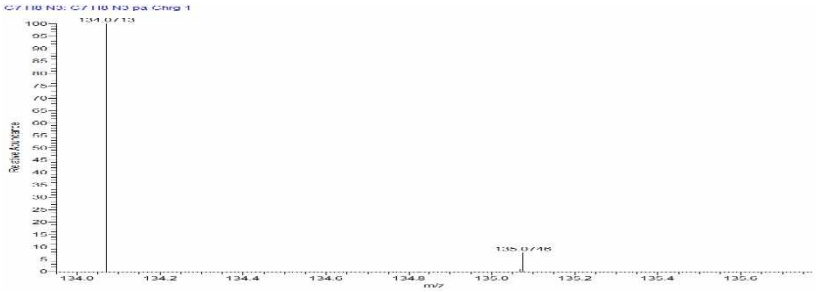


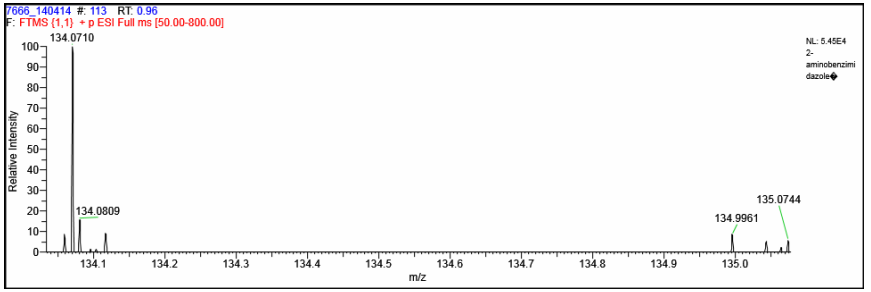
[M+H+]= 134.0713

C7H8N3

Fragment 1= 133.0634

C7H7N3





TAKE HOME MESSAGES

GENERIC SAMPLE PREP. SUITED FOR MANY PESTICIDES/TOXICANTS

LC-MASS ACCURACY-HRMS READY TO REPLACE MANY QqQ METHODS

COMBINE TARGET/ POST-TARGET STRATEGIES FOR MASSIVE DETECTION OF PESTICIDES AND METABOLITES IN AMBIENT AIR

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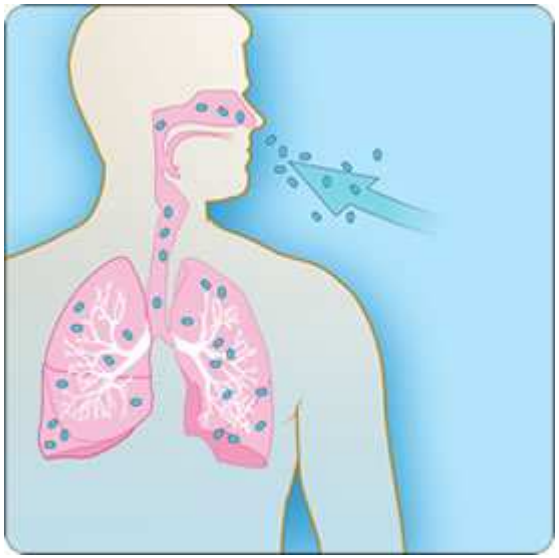

EXPOSURE AND RISK ASSESSMENT OF AIRBORNE PESTICIDES

HAZARD QUOTIENT (HQ)

$$HQ = DIE_i / HBRV_i$$

HBRVs: AOAEL, ADI, ARfD

Hazard Quotients (HQ) as a risk descriptor

$HBRV_i$ is the Health Based Reference Value for i

HQ < 1 ➔ ACCEPTABLE RISK

DAILY INHALATION EXPOSURE (DIE)

$$DIE\ (\mu g/kg/day) = \Sigma(C \times IR_{inh} \times ED)/ BW$$

C is the total (particle+ gas phases) concentration of each pesticide in the air (ngm^{-3}), IR_{inh} is the inhalation rate per hour (m^3/h), ED is the exposure duration (h) to air and BW is body weight of the subject (kg).

USEPA / WHO

METHODS FOR CUMULATING THE TOXICITY OF COMPOUNDS

HAZARD INDEX (HI)

The hazard index (HI) is the sum of the hazard quotients (HQ), i.e. the ratio between exposure and the respective RV for each compound in the CAG.

$$HI = \frac{Exp_1}{RV_1} + \frac{Exp_2}{RV_2} + \frac{Exp_3}{RV_3} + \cdots$$

RVs: Reference Values: (AOAEL, ADI, ARfD,)

HI < 1 ➔ ACCEPTABLE RISK

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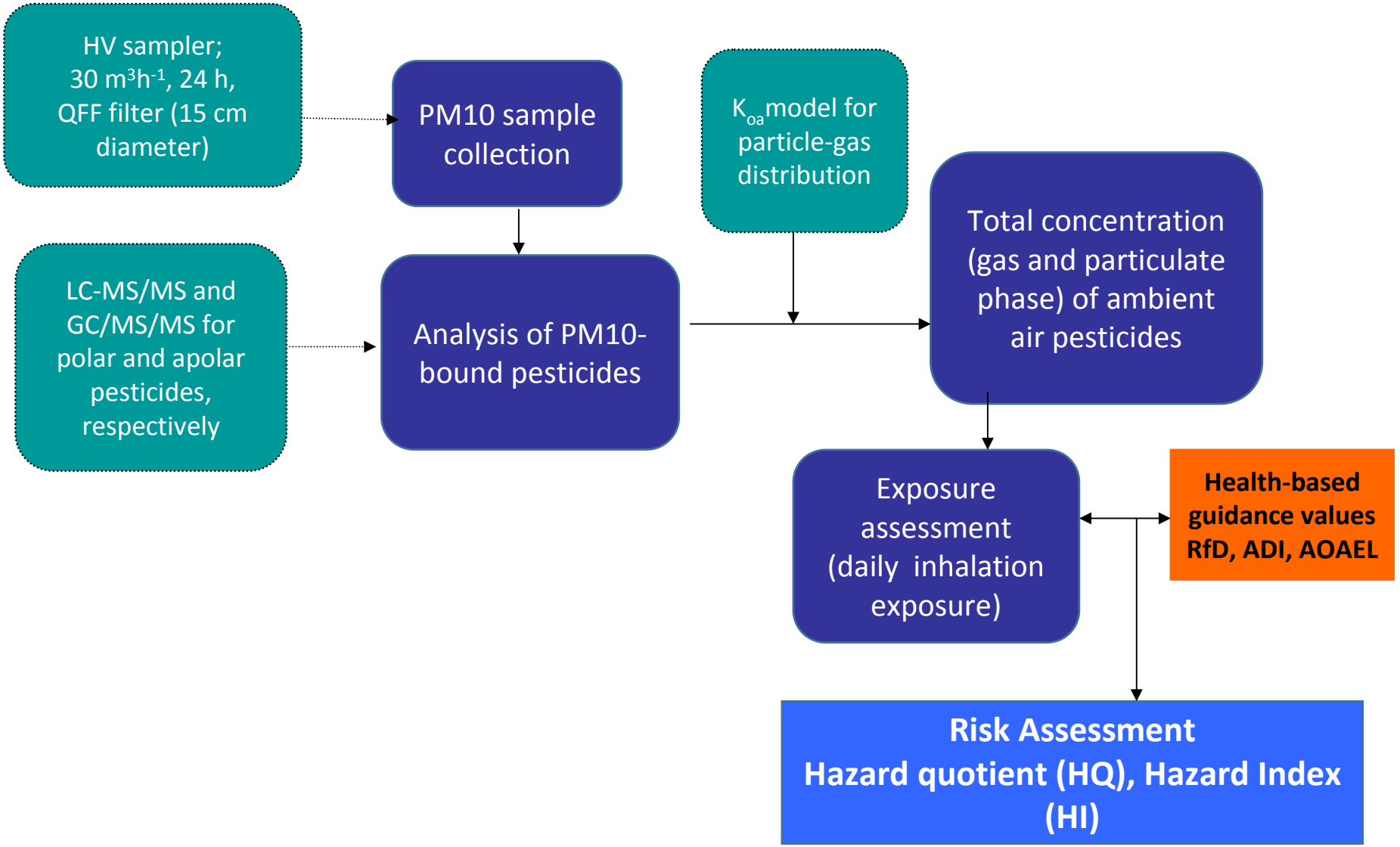
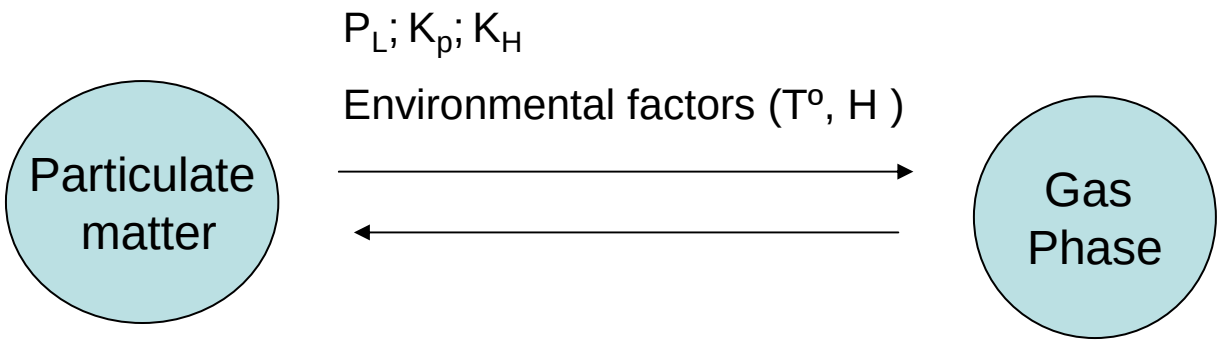
SCREENING RISK ASSESSMENT OF ATMOSPHERIC PESTICIDES



G/P PARTITIONING



K_{oa} ABSORTION MODEL (Finnizio et al.1887; Harner and Bieldman, 1998, Sofouglu, 2004)

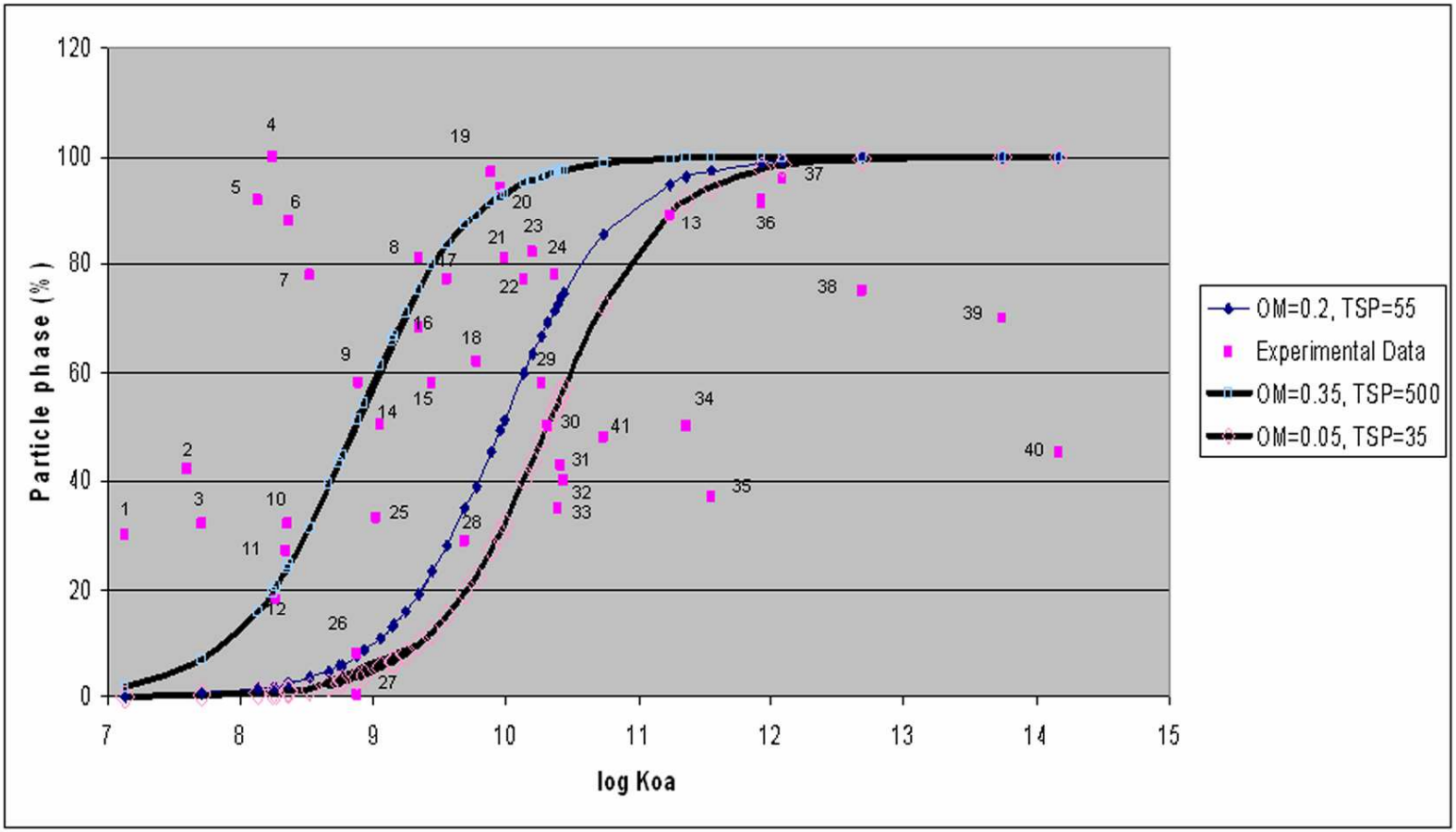
$$ø = (K_pC_{TSP})/(1 + KpC_{TSP})$$

Where *ø* is the particulate percentage (fraction of the compound in the particle phase), C_{TSP} is the concentration of total suspended particles in the air (µg m-3), and **K_p is the gas/particle partition coefficient.**

$$\log K_p = \log K_{oa} + \log f_{OM} - 11.91$$

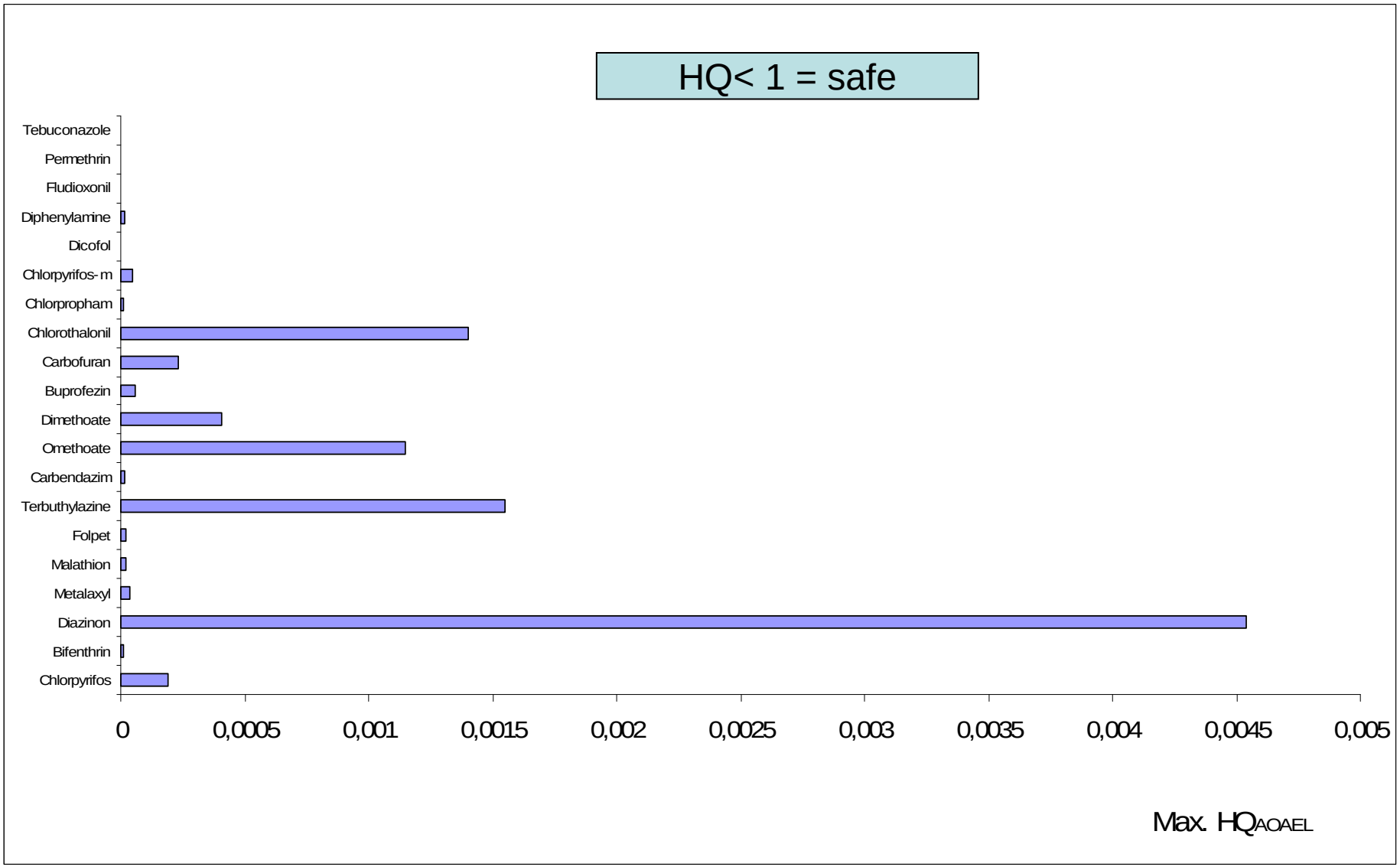
Where $\log K_{oa}$ is the octanol-air partitioning coefficient, and $\log f_{OM}$ is the fraction of organic matter.

G/P PARTITIONING

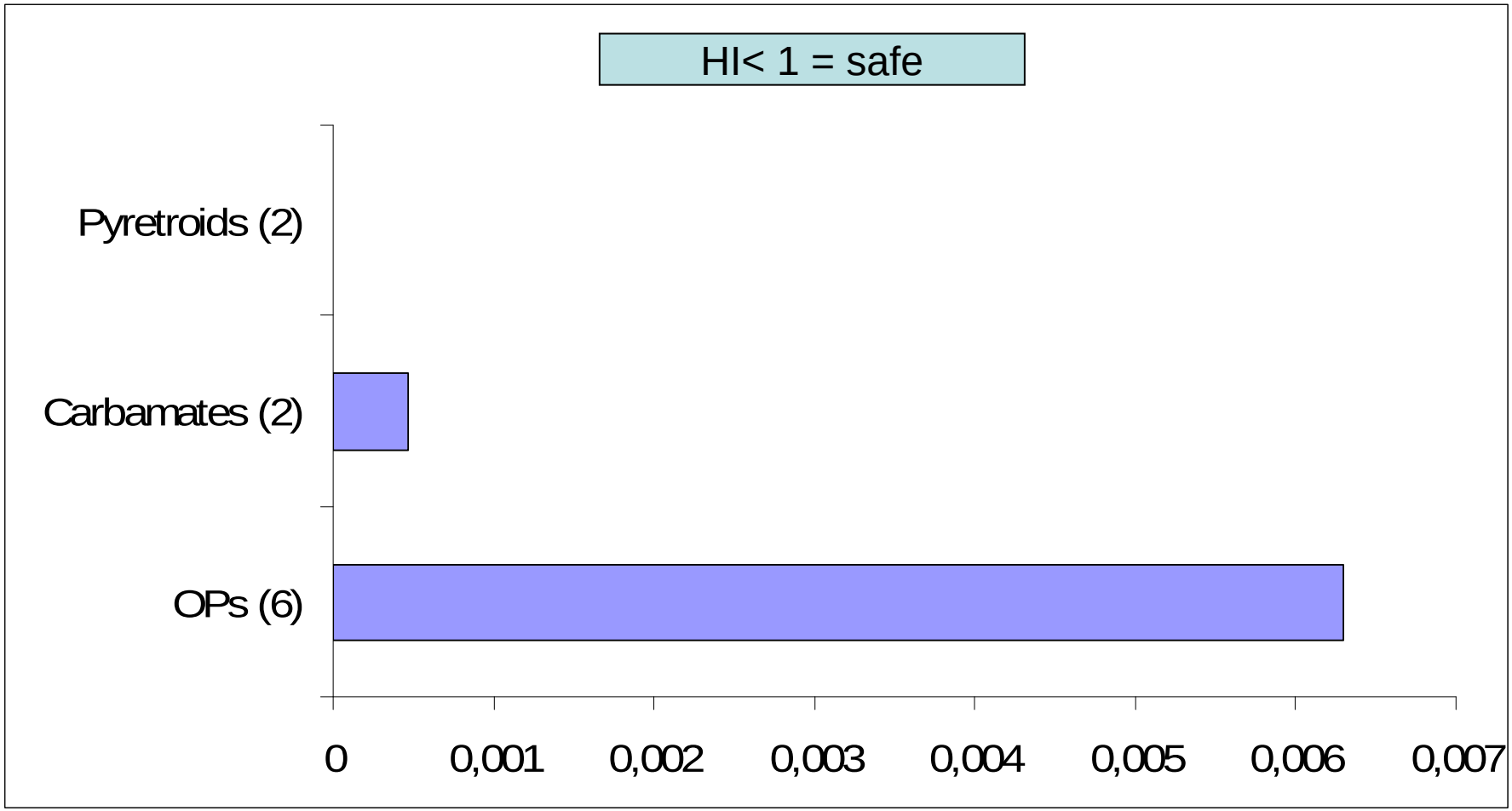


HAZARD QUOTIENT

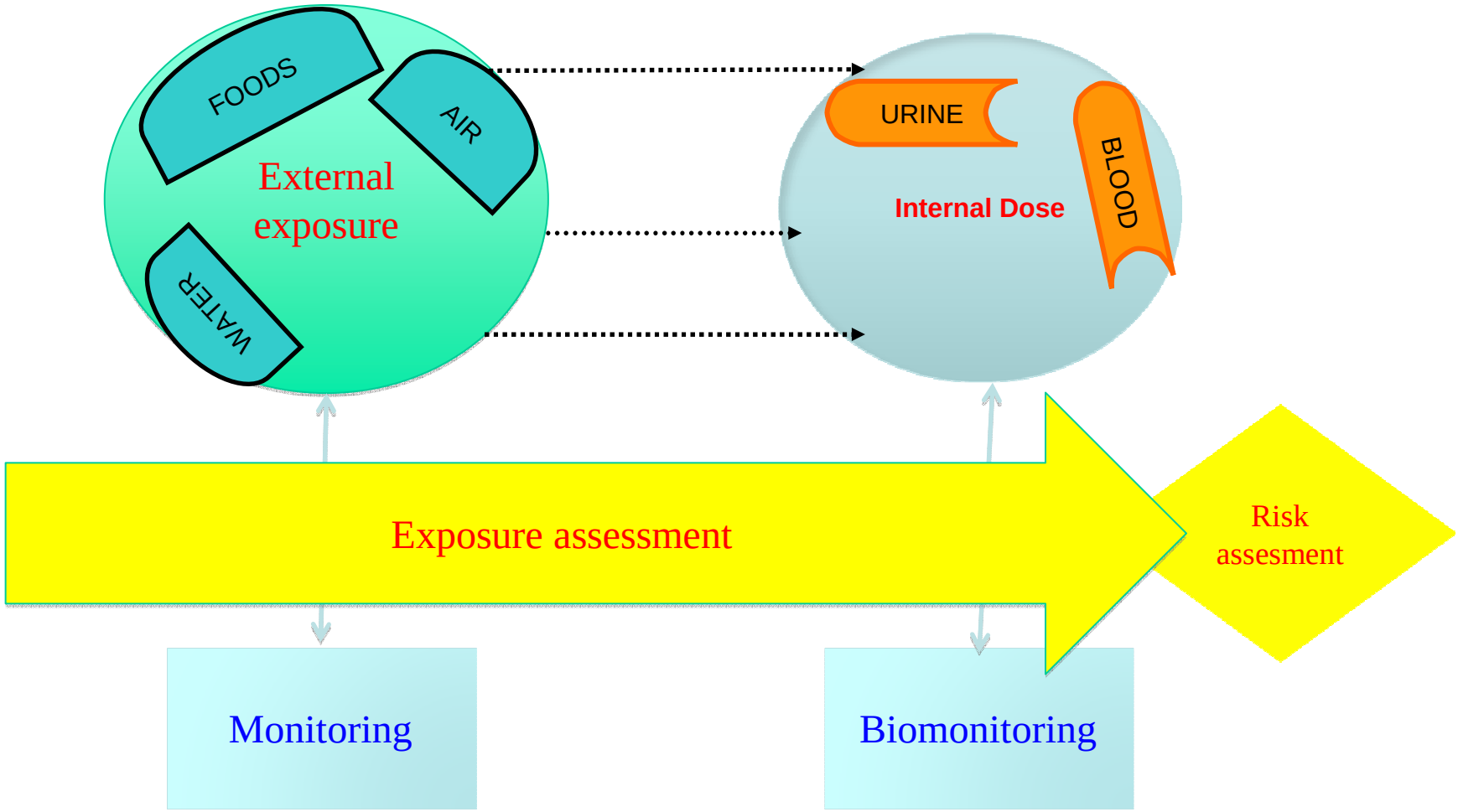
HQ< 1 = safe



HAZARD INDEX



INTEGRATED EXPODURE



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THANK YOU

Dr. Vicent Yusà (yusa_vic@gva.es) // Vicent.Yusa@uv.es)