Non-target screening of environmental pollutants in the context of risk assessment of European river basins: the NORMAN network perspective

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What's new in the analysis of complex environmental matrices?

Royal Society of Chemistry

Environmental Chemistry Group, Water Science Forum and the Separation Science Group Joint Meeting

London, 3 March 2017

Emerging pollutants in EU environmental policies

- Water Framework Directive **(2000/60/EC)**
  - **River basin specific pollutants**
  - EQS Directive **(2008/105/EC, 2013/39/EU)**
  - Groundwater Directive **(2006/118/EC)**
- Marine Strategy Framework Directive **(2008/56/EC)**
- Drinking Water Directive **(98/83/EC, 2015/1787)**
  - **Water Safety Plans**
- REACh **(EC Regulation 1907/2006)**
- *Water Reuse Policy Instrument (Directive, 2017??)*
- *Indoor environment (future Directive??)*
- *Soil, sludge*
- *Air – long-range transport*

More than 120 river basins in Europe: Are monitoring data comparable?

Danube River Basin District Management Plan

Ecological status and ecological potential

Chemical status

Where are we today?

- No good ecological status in most European river basins (WFD)
- Chemicals play a significant role for degradation
- Priority substances often do not explain effects ⇒ emerging pollutants?

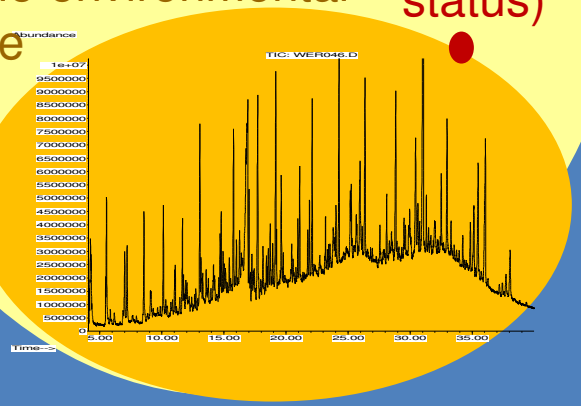
Where are we today?

Chemosphere

70 mio known chemicals
14 mio commercially available
> 100.000 in daily use

45 priority substances (chemical status)

> 5.000 compounds in a single environmental sample

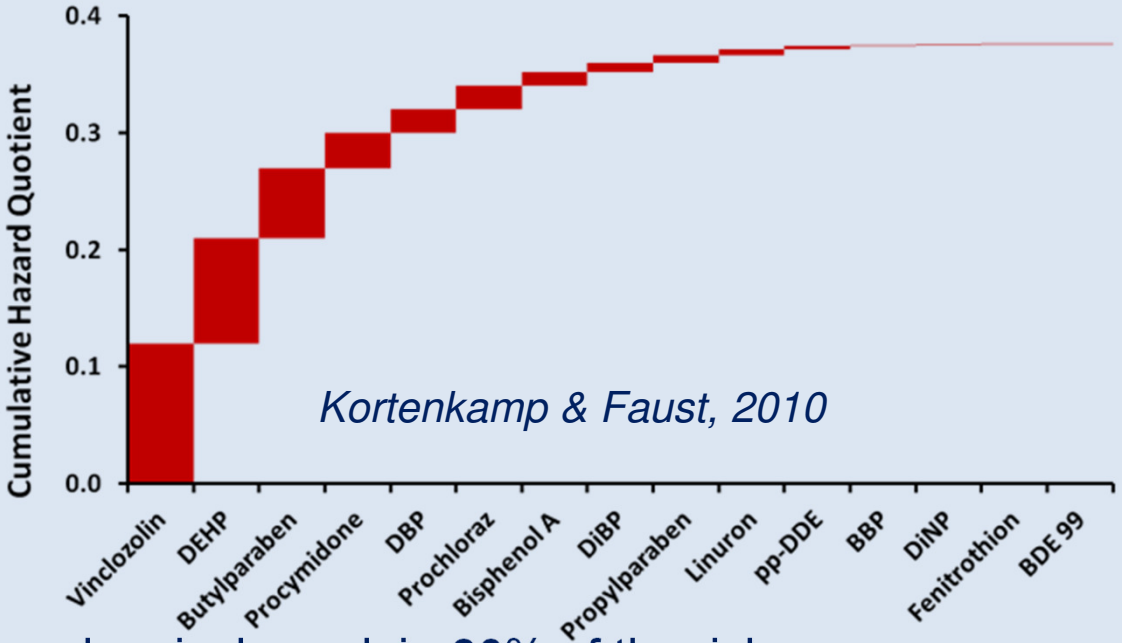


- Emerging pollutants (e.g. pharmaceuticals, personal care products, biocides…, transformation products)
  - in daily use
  - widespread
  - neither regulated nor monitored
  - often polar, ionic, multi-functional („difficult“)
  - often poorly retained in WWTPs
  - occur in mixtures
  - may exhibit great biological activity

solutions

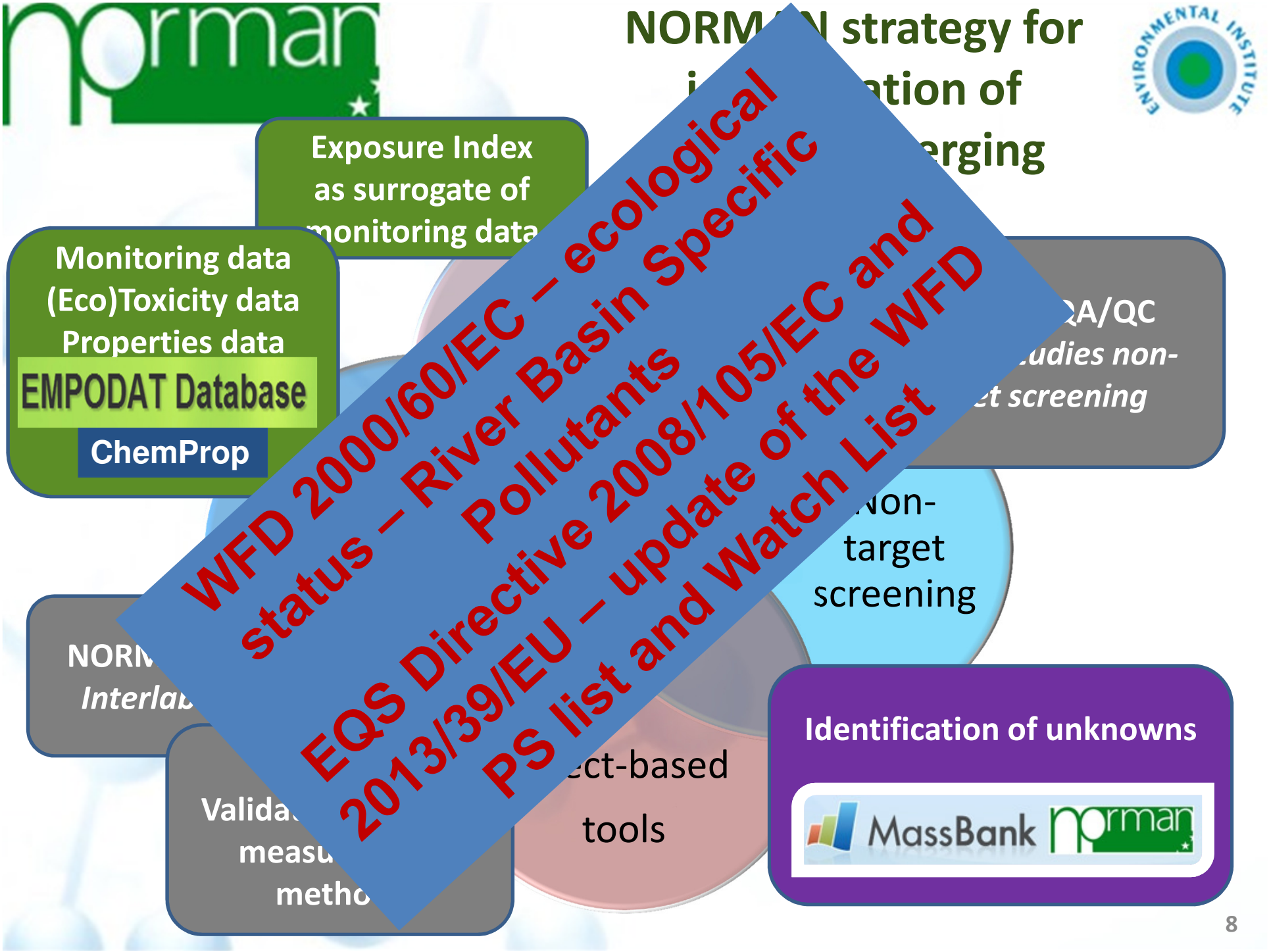
solutions

Many chemicals in the environment but only few determine the risk.
Example: mixture toxicity of 15 anti-androgenic compounds in typical environmental concentrations

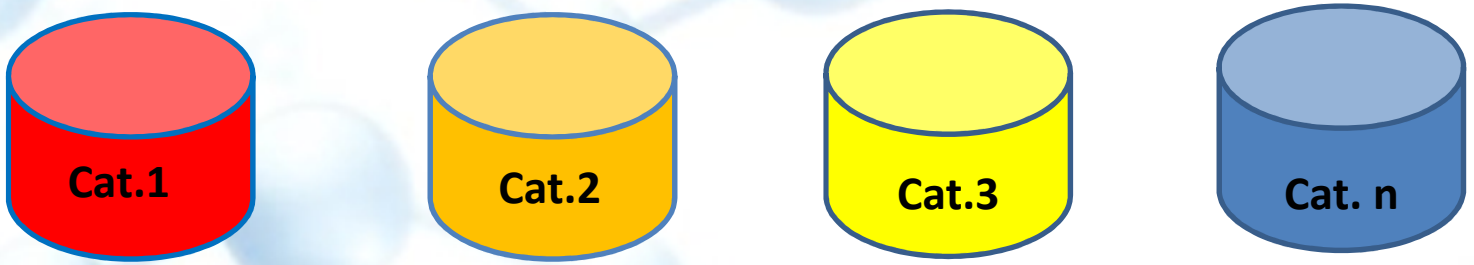


Five chemicals explain 80% of the risk
Efficient management needs to focus on these chemicals

NORMAN strategy for i…ation of …erging



Categorisation of substances by knowledge gaps & actions needed

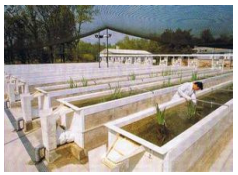


Action categories

1. Control / mitigation measures

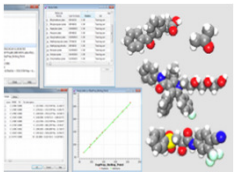
2. Screening campaigns

3. Rigorous hazard assessment

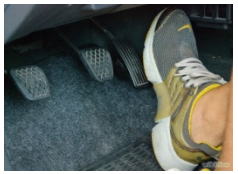


4. Improvement of analytical methods

5. Screening AND hazard assessment

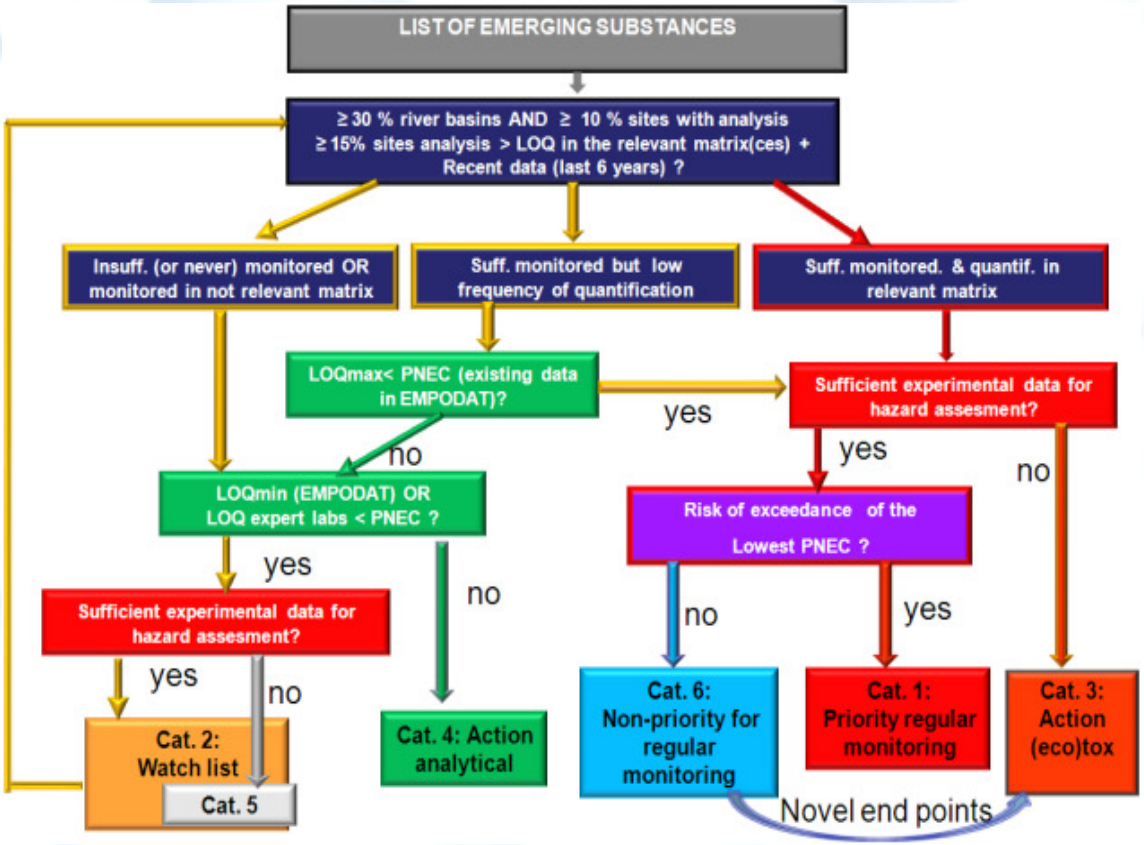


6. Reduced monitoring efforts



WG-1 : Priorisation of emerging substances

NORMAN Prioritisation scheme for emerging substances
(V. Dulio & P.C. von der Ohe, 2013, ISBN : 978-2-9545254-0-2)



- **Prioritisation by action categories (on the basis of identified knowledge gaps)**
- **Ranking within each category based on Occurrence + Hazard + Risk**

Risk indicators

Extent of Exceedance = MEC95 / Lowest PNEC

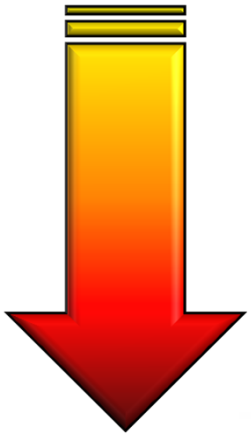
to address the intensity of impact

where:

- *MEC95 (95th percentile of the max conc. at each site)*
- *Lowest PNEC*
- *Equivalent to PEC/PNEC!*

Score for „Exceedance of environmental threshold“

MEC95/lowest PNEC <1 = 0
10≥ MEC95/lowest PNEC≥1 =0.1
100≥ MEC95/lowest PNEC>10 = 0.2
1000≥ MEC95/lowest PNEC>100 =0.5
MEC95/lowest PNEC>1000 = 1



Risk indicators

Frequency of Exceedance = n / N

to address the spatial exposure aspects

where:

- *n is the number of sites with MECsite > Lowest PNEC*
- *N is the total number of sites where the substance was measured*

Score: value between 0 and 1

- **Cat. 1, 3, 6:** calculated using RECENT DATA
- **Cat. 2, 4, 5:** calculated using ALL DATA (all YEARS)

Table 96: Results of the prioritisation of pollutants determined in the JDS3 surface water samples

No.	Substance	CAS No.	No. of sites substance detected	C_{max}[1]	MEC_{95}[2]	Lowest PNEC/EQS	Key study	Type	EoE[3]	EoE score	FoE[4]	Final score
1	2,4-Dinitrophenol (DNP)	51-28-5	68	0.06	0.04	0.001	RIVM 2014	EQS chronic water[5]	40	0.2	1.00	1.20
2	PFOS (Perfluorooctansulfonate)	1763-23-1	63	0.026	0.02	0.00065	EU 2013	EQS chronic water[5]	31	0.2	0.93	1.13
3	Chloroxuron	1982-47-4	65	0.04	0.02	0.0024	James et al. 2009	PNEC acute	8.3	0.1	0.93	1.03
4	Desethylterbutylazine	30125-63-4	54	0.028	0.01	0.0024	RIVM 2014	EQS chronic water[5]	4.2	0.1	0.79	0.89
5	2-hydroxy atrazine	2163-68-0	53	0.06	0.02	0.002	Ecostat 2013	EQS chronic water[5]	10	0.1	0.76	0.86
6	Bromacil	314-40-9	31	0.19	0.14	0.01	INERIS 2013	EQS chronic water[5]	14	0.2	0.46	0.66
7	Dimefuron	34205-21-5	58	0.041	0.04	0.008	Oekotoxzentrum 2014	EQS chronic water[5]	5.0	0.1	0.56	0.66
8	Bisphenol A	80-05-7	30	1.94	1.03	0.1	Nendza 2003	EQS chronic water[5]	10	0.2	0.16	0.36
9	Benzo(g,h,i)perylene	191-24-2	65	0.029	0.003	0.002	CEC 2008	EQS chronic water[5]	1.5	0.1	0.26	0.36
10	Diazinon	333-41-5	21	0.009	0.01	0.001	Management Team PPDB 2009	PNEC acute	10	0.1	0.12	0.22
11	Indeno(1,2,3-c,d)pyrene	193-39-5	15	0.005		0.002	CEC 2008	EQS chronic water[5]			0.19	0.19
12	Linuron	330-55-2	32	1.42	1.12	0.26	Oekotoxzentrum 2014	EQS chronic water[5]	4.3	0.1	0.07	0.17
13	Amoxicillin	26787-78-0	33	0.28	0.08	0.078	van der Aa et al. 2011	PNEC chronic	1.0	0.1	0.03	0.13
14	Metazachlor	67129-08-2	30	0.03	0.02	0.019	INERIS 2014	EQS chronic water[5]	1.1	0.1	0.03	0.13
15	17beta-estradiol	50-28-2	8	0.029		0.0004	CEC 2011	EQS chronic water[5]			0.12	0.12
16	Benzo(a)pyrene	50-32-8	3	0.002		0.00017	EU 2013	EQS chronic water[5]			0.04	0.04
17	Diclofenac	15307-79-6	51	0.318	0.036	0.05	Oekotoxzentrum 2014	EQS chronic water[5]			0.04	0.04
18	Bentazon	25057-89-0	61	0.1	0.02	0.06	USEPA 2008	PNEC acute			0.01	0.01
19	Fipronil	120068-37-3	1	0.02		0.012	EU 2011	EQS chronic water[5]			0.01	0.01
20	Fluoranthene	206-44-0	58	0.02	0.006	0.0063	EU 2013	EQS chronic water[5]			0.01	0.01

1 C_{max} – Maximum concentration in µg/L reported in case the substance has been measured by several JDS3 laboratories

2 MEC_{95} – 95th percentile of the Maximum Environmental Concentration in µg/L; calculated only if the substance has been found above LOQ at minimum 20 sites

3 EoE – Extent of Exceedance

4 FoE – Frequency of Exceedance

5 Equal to Annual Average EQS (AA-EQS)

The Danube River Basin District Management Plan, Update 2015

List of NORMAN emerging substances

Delete / hide	Former emerging substance	Keep on NORMAN List	NEW: Add to NORMAN List
•38 compounds : • WFD PS (diuron, dichlorvos, dicofol, heptachlor, PCBs, BDE -47, BDE-153, BDE-154, HBCDD, PAHs, PFOS, etc.) • Microcystines • Well known Industrial Chem. (aniline, styrene, toluene, xylenes, etc.)	**• 72 compounds :** • Phathalates • Organotins • Nitro musks • Organo-lead • 8 well known PFASs • 23 herbicides / insecticides, banned in EU and /or not frequently detected / quantif • 10 surfactants (NPEOs, LAS)	**• 527 compounds** •8 Plasticisers •73 PPP • 20 PPP / biocides or biocides •209 Pharmas • 60 Pers care prod. •16 Flame retard. • 44 Ind. Chemicals • 8 PFASs, ….etc. **•61 DBP <u>(only</u>** drinking water)	**• 253 compounds :** • 67 flame ret. (used as alternatives to banned products) • 10 PPP most frequently detected, highest conc. or chronic EQS exceeded in recent studies • 118 PPP/biocides & biocides in use or under review • 50 PFASs in use • 8 Pharmas, etc.

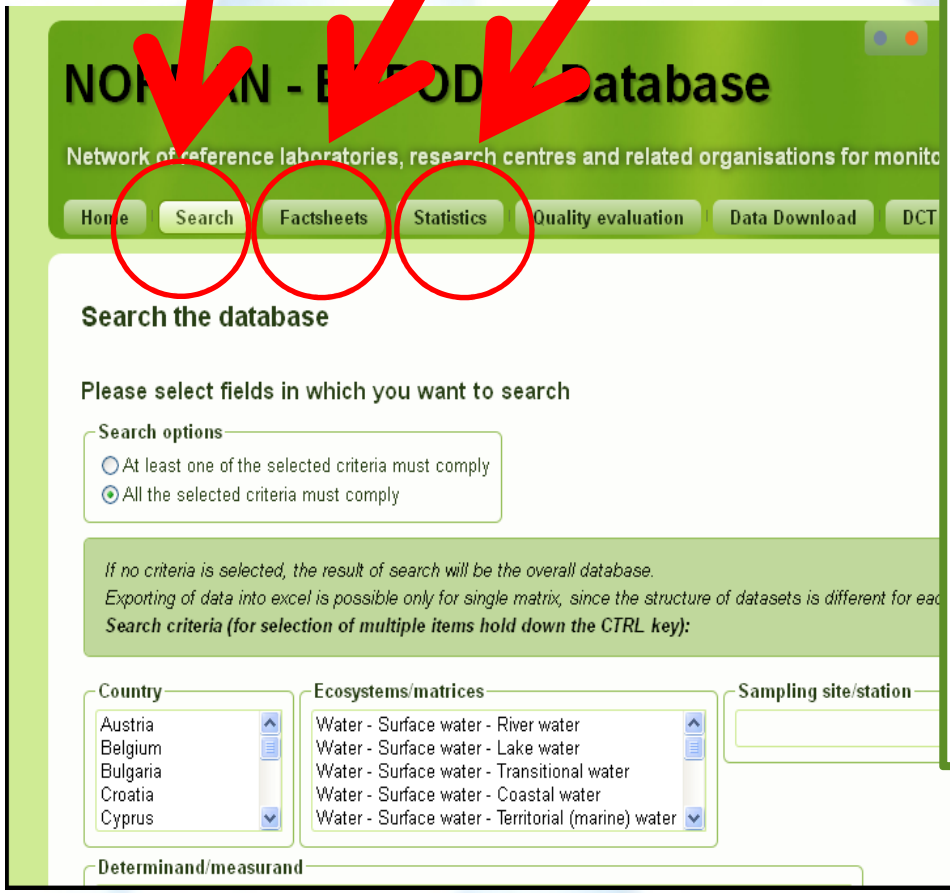
New NORMAN prioritisation list: 860 substances

NORMAN EMPODAT Database
Chemical Module

Search

Factsheets

Statistics



>10 million datasets
430 subst. from 25 countries

Matrices: water, sediment, biota

Harmonised data reporting format and metadata compatible for use by the Commission

Substance Prioritisation tool integrated in the database

ALL data accessible after registration

http://www.norman-network.net/empodat/

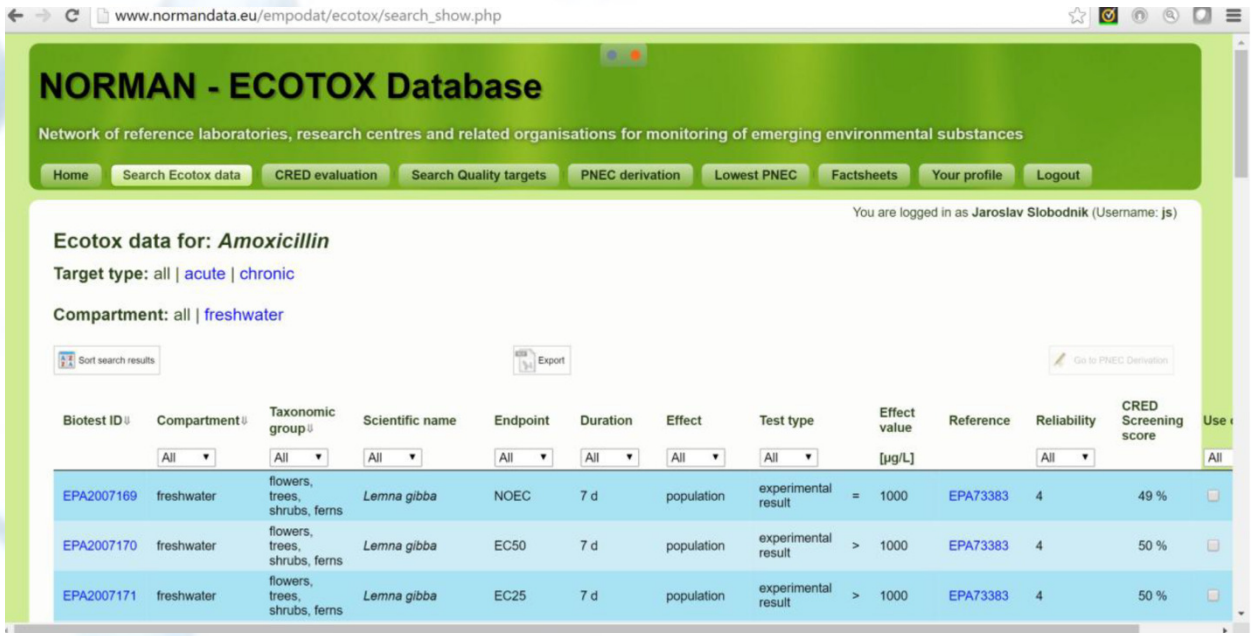
PNECs – how to trust them?

NORMAN ECOTOX database – unified effort of leading European ecotoxicologists

- Compilation of experimental ecotox data from databases
  ➔ about 500 substances
- QSAR prediction for substances with no experimental data
  ➔ all substances
- Collection of existing Quality Targets (PNECs)
  ➔ about 200 substances

Implementation of the CRED* system to ensure high quality and relevance of the data

➔ Derivation of the **Lowest PNEC** by NORMAN experts



NORMAN contribution to EC

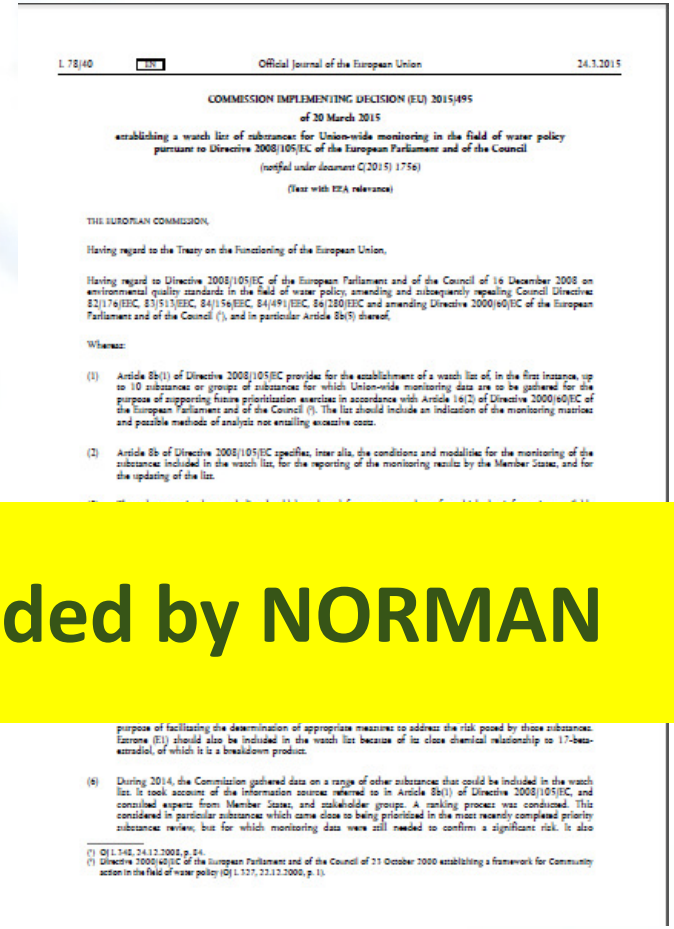
- Indicators of NORMAN prioritisation (e.g. frequency of PNEC exceedance + degree of PNEC exceedance…..) are included in the EC methodology

- EMPODAT used as an important data source to support EU Watch List and PS review

Commission implementing Decision (EU) 2015/495 of 20 March 2015

Substance name
17-Alpha-ethinylestradiol (EE2)
17-Beta-estradiol (E2), **Estrone (E1)**
Diclofenac
Methiocarb
2,6-ditert-butyl-4-methylphenol
2-Ethylhexyl 4-methoxycinnamate
Macrolide antibiotics: **Erythromycin** (CAS N° 114-07-8), **Clarithromycin** (CAS N° 81103-11-9), **Azithromycin** (CAS N° 83905-01-5).
Neonicotinoids: Imidacl… Thiaclo… Thiame… Clothia… Acetam…
Oxadiazon
Tri-allate

Watch list of substances for Union-wide monitoring in the field of water



L 78/40 EN Official Journal of the European Union 24.3.2015

COMMISSION IMPLEMENTING DECISION (EU) 2015/495
of 20 March 2015
establishing a watch list of substances for Union-wide monitoring in the field of water policy pursuant to Directive 2008/105/EC of the European Parliament and of the Council
(notified under document C(2015) 1756)
(Text with EEA relevance)

THE EUROPEAN COMMISSION,

Having regard to the Treaty on the Functioning of the European Union,

Having regard to Directive 2008/105/EC of the European Parliament and of the Council of 16 December 2008 on environmental quality standards in the field of water policy, amending and subsequently repealing Council Directives 82/176/EEC, 83/513/EEC, 84/156/EEC, 84/491/EEC, 86/280/EEC and amending Directive 2000/60/EC of the European Parliament and of the Council (1), and in particular Article 8b(3) thereof,

Whereas:

(1) Article 8b(1) of Directive 2008/105/EC provides for the establishment of a watch list of, in the first instance, up to 10 substances or groups of substances for which Union-wide monitoring data are to be gathered for the purpose of supporting future prioritisation exercises in accordance with Article 16(2) of Directive 2000/60/EC of the European Parliament and of the Council (2). The list should include an indication of the monitoring matrices and possible methods of analysis not entailing excessive costs.

(2) Article 8b of Directive 2008/105/EC specifies, inter alia, the conditions and modalities for the monitoring of the substances included in the watch list, for the reporting of the monitoring results by the Member States, and for the updating of the list.

purpose of facilitating the determination of appropriate measures to address the risk posed by those substances. Estrone (E1) should also be included in the watch list because of its close chemical relationship to 17-beta-estradiol, of which it is a breakdown product.

(6) During 2014, the Commission gathered data on a range of other substances that could be included in the watch list. It took account of the information sources referred to in Article 8b(1) of Directive 2008/105/EC, and consulted experts from Member States, and stakeholder groups. A ranking process was conducted. This considered in particular substances which came close to being prioritised in the most recently completed priority substances review, but for which monitoring data were still needed to confirm a significant risk. It also

(1) OJ L 348, 24.12.2008, p. 84.
(2) Directive 2000/60/EC of the European Parliament and of the Council of 23 October 2000 establishing a framework for Community action in the field of water policy (OJ L 327, 22.12.2000, p. 1).

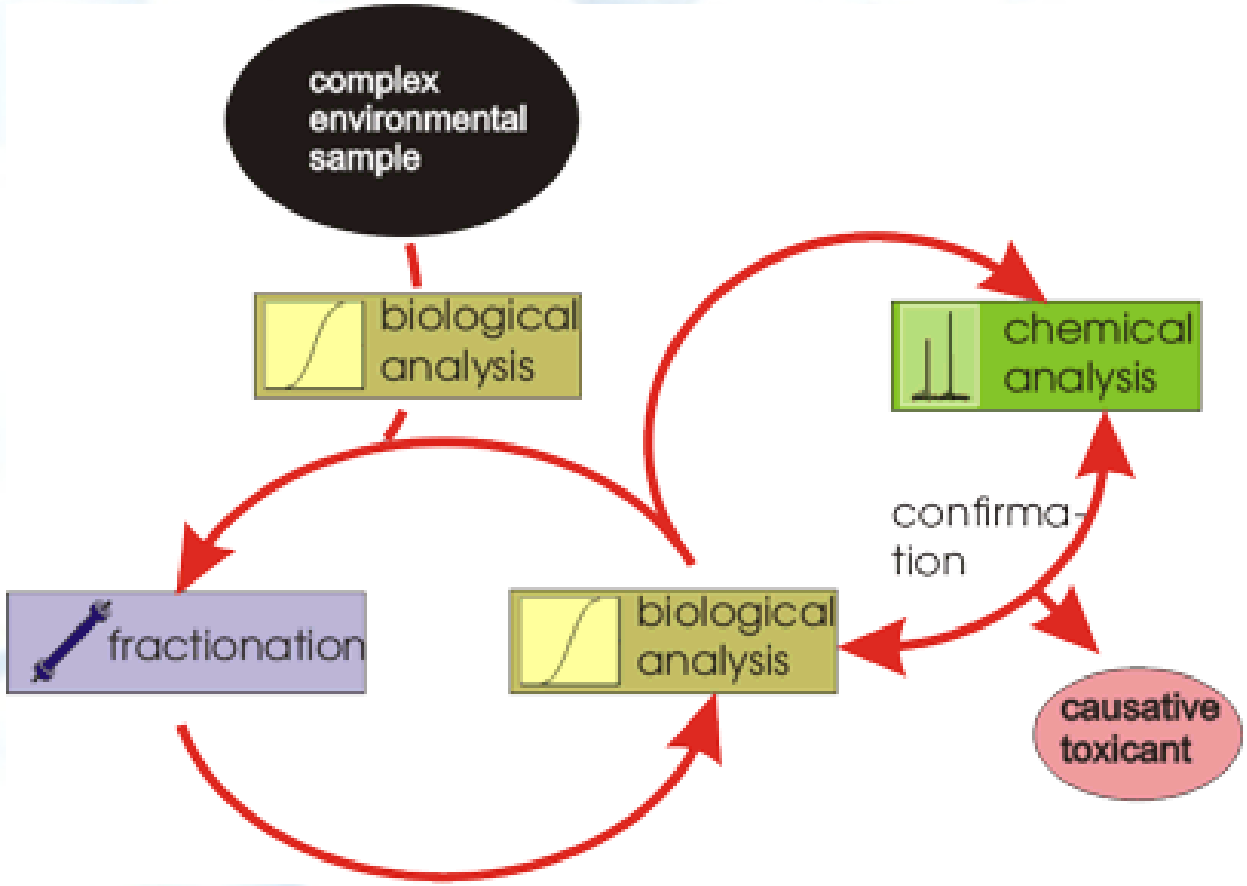
➔ 8 substances recommended by NORMAN

What needs to be improved

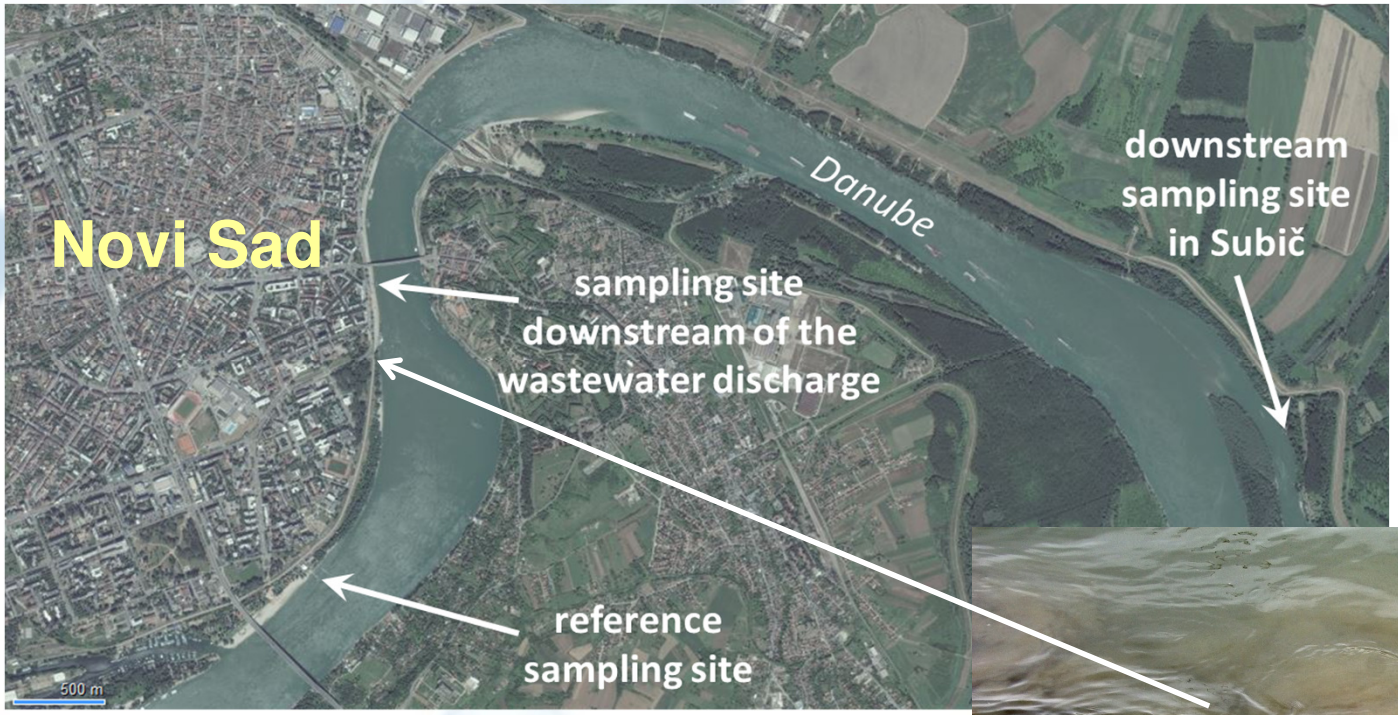
- Attention is focused on already well-**known contaminants**
  - **Metabolites** and **transformation products**?
  - Risk of overlooking new emerging pollutants
- Individual pollutants are assessed as if they would occur in isolation
  - Mixture effects are not taken into account in prioritisation methodologies

Effect directed analysis (EDA)

Effect-directed identification of site- and basin-specific key toxicants for the establishment of cause-effect relationships and improved risk assessment



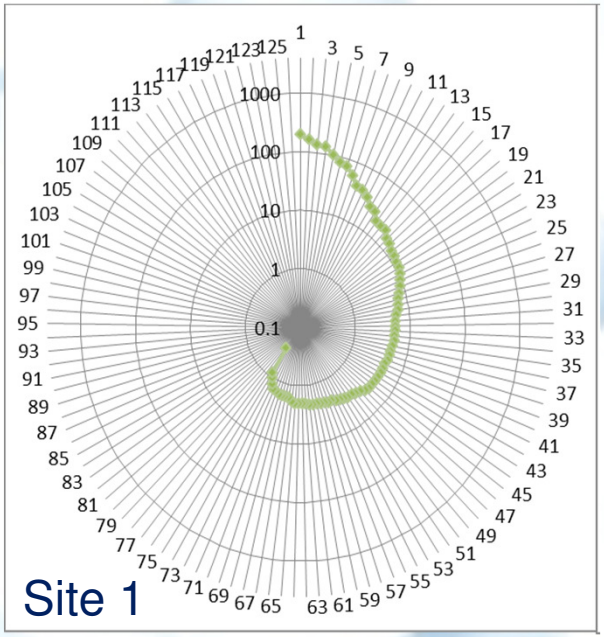
Novi Sad - Sampling sites at the River Danube



http://www.bing.com/maps/?FORM=HDRSC4

Analytical screening

Upstream of Novi Sad wastewater effluent

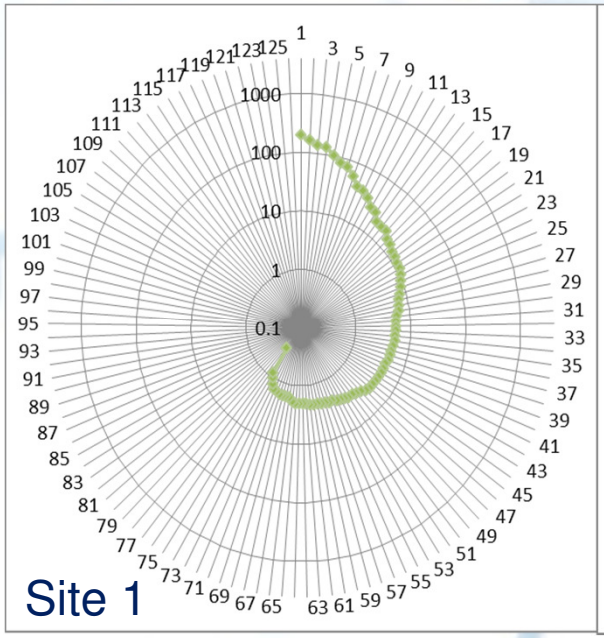


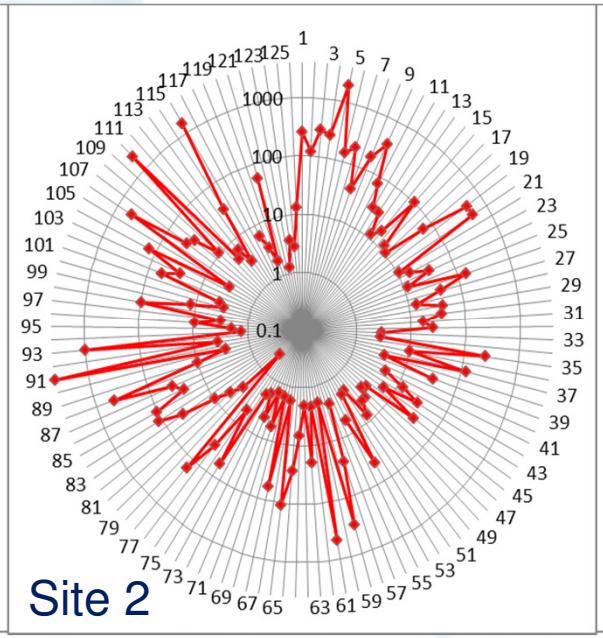
No.	Compound	C (ng/L)
1	Enalapril	194,2
2	2-Benzothiazolesulfonic acid	164,2
3	Sucralose	133,9
4	1H-Benzotriazole	133,7
5	Metformin	98,8
6	4- and 5-Methyl-1H-benzotriazole	81,3
7	Carbamazepine	74,1
8	p-Toluenesulfonamide	58,2
9	N-Acetyl-4-aminoantipyrine	41,4
10	Hexa(methoxymethyl)melamine	39,4
11	N-Formyl-4-aminoantipyrine	34,7
12	Metolachlor ESA	26,8
13	Triphenylphosphine oxide	24,9

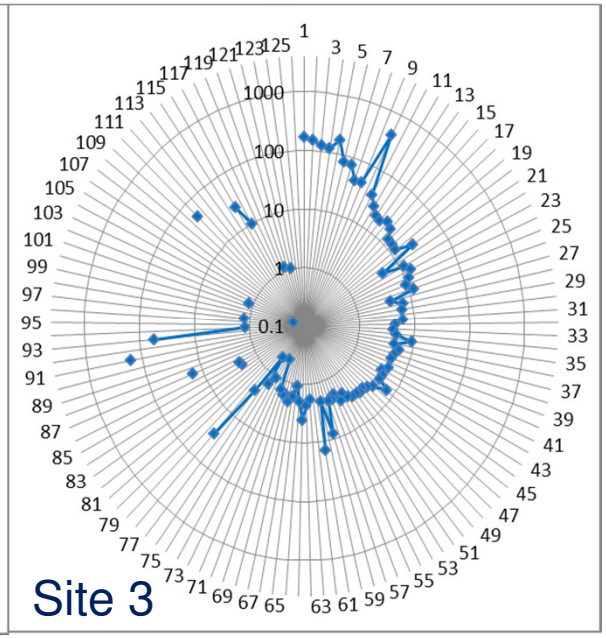
Pharmaceuticals and transformation products

Industrial chemical | Food/stimulant | Pesticides/biocides

Analytical screening



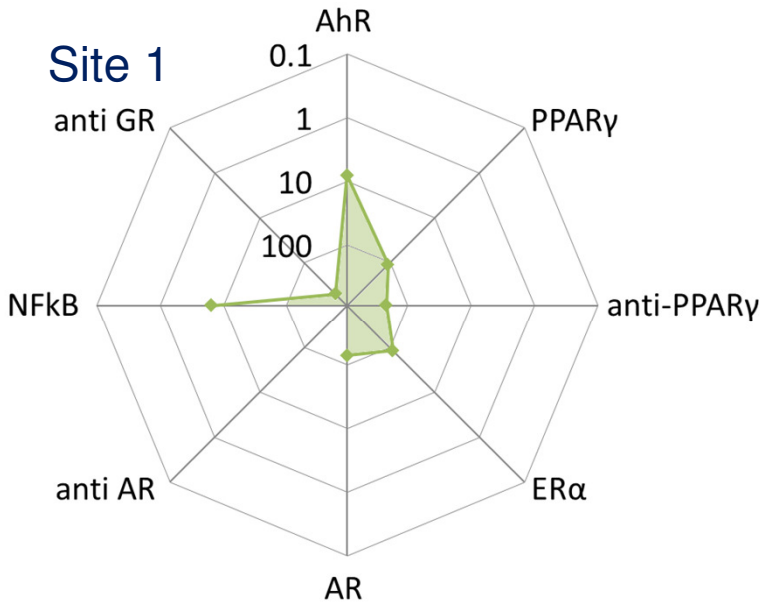


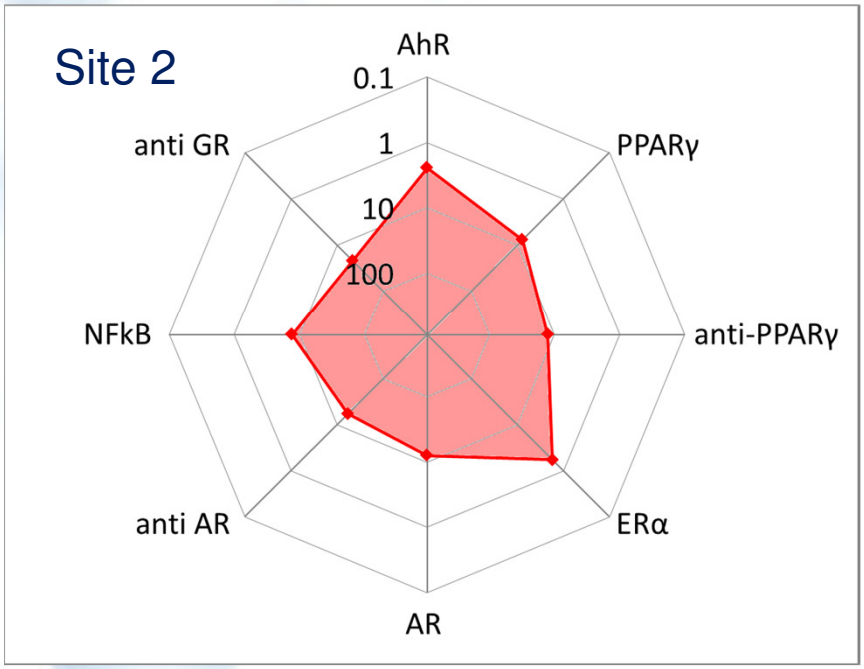


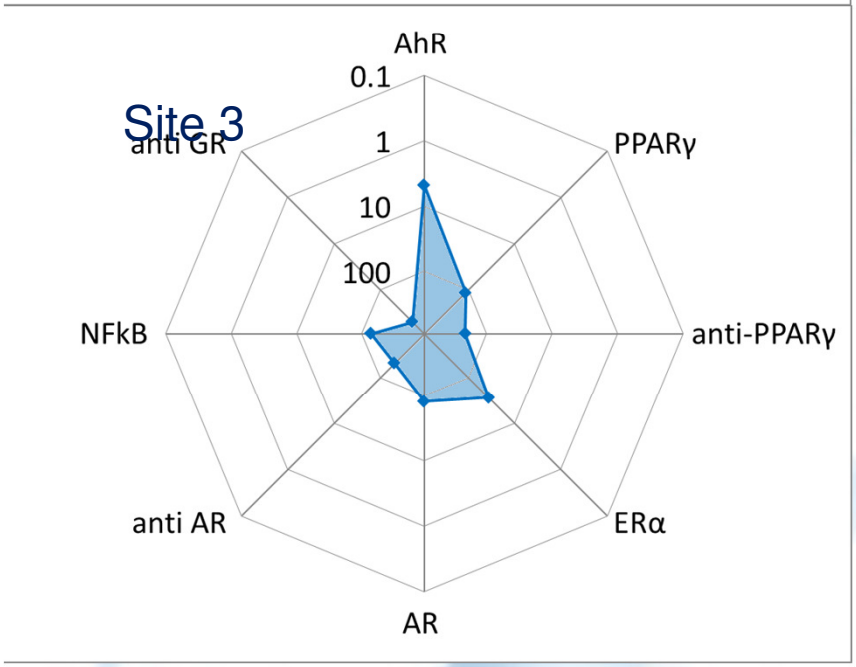
- Chemical screening: 125 target compounds detected
- Strong influence on contamination pattern downstream of Novi Sad wastewater input
- Almost re-establishment of original pattern 7 km downstream
- Some significantly enhanced compounds remain

Bioanalytical screening

Relative enrichment factors (REFs)







- agonistic effects → EC_{10}
- antagonistic effects → $SR_{0.2}$
- adaptive stress response → $EC_{IR1.5}$

When monitoring data are missing? NORMAN Exposure Index

Important gaps as regards monitoring data for a lot of pharmaceuticals and biocides

- For industrials chemicals, exposure index based on 3 components:

1. Quantity used (*ATscore*)
2. Release During Use (*UIscore*)
3. Wide Dispersive Use (*RoUscore*)

$$Exposure\ index = \frac{ATscore + UIscore + RoUscore}{3}$$

CREATION OF AN EXPOSURE INDEX FOR PHARMACEUTICALS

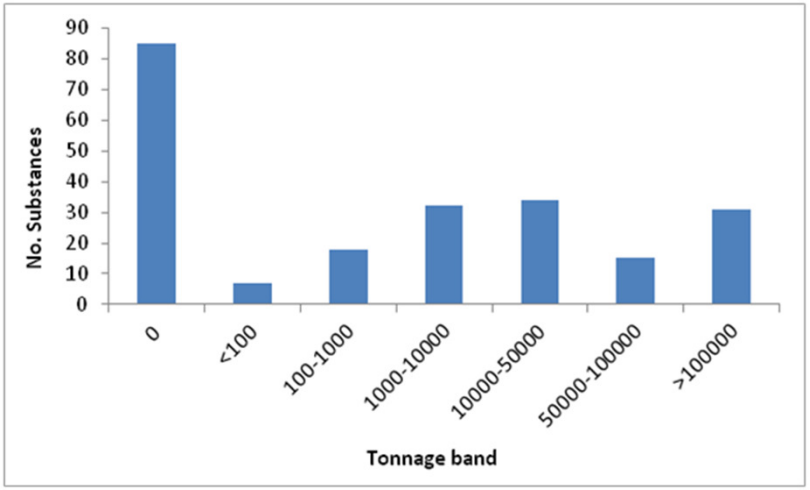
Consumption score (QU factor)

The annual consumption of a given substance in Europe, by kilograms per year

Data from countries and year

Countries	Year
Germany	2009
Sweden	2011
Netherlands	2014
Switzerland	2009
Italy	2014
France	2004

- Distribution of the pharmaceuticals by consumption band



EI - Summary of the results of 3 different scenarios

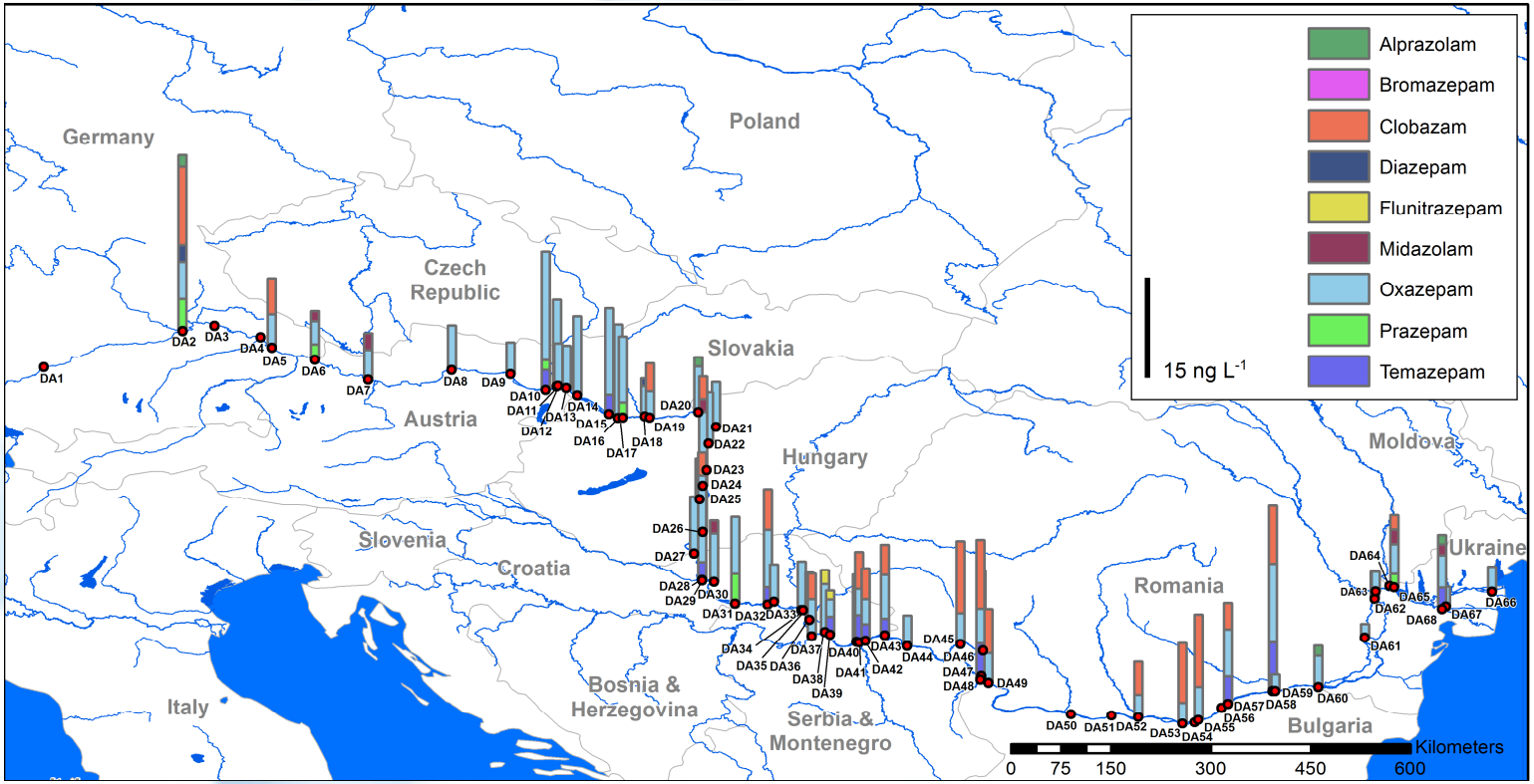
Rank	Scenario 1		Scenario 2		Scenario 3	
	Substance name	Exposure Index	Substance name	Exposure Index	Substance name	Exposure Index
1	Furosemide	8,58	Furosemide	8,58	Furosemide	10,00
2	Hydrochlorothiazide	8,08	Hydrochlorothiazide	8,08	Hydrochlorothiazide	9,38
3	Amoxicilline	7,78	Amoxicilline	7,78	Morphine	9,38
4	Oxytetracycline	7,78	Morphine	7,78	Amoxicilline	9,00
5	Tetracycline	7,78	Ciprofloxacine	7,62	Cefazoline	8,88
6	Ciprofloxacine	7,62	Lidocaine	7,62	Cefalexin	8,38
7	Lidocaine	7,62	Prednisolone	7,56	Prednisolone	8,38
8	Ampicilline	7,48	Metformine	7,40	Trimethoprime	8,38
9	Metformine	7,40	Cefazoline	7,38	Gentamicin	8,25
10	Cefalexin	7,38	Ofloxacin	7,24	Iohexol	8,25
11	Cefazoline	7,38	Gentamicin	7,18	Iopromide	8,25
12	Ofloxacin	7,24	Trimethoprime	7,16	Omeprazole	8,00
13	Gentamicin	7,18	Dexamethasone	7,12	Phenobarbital	7,88
14	Trimethoprime	7,16	Cefalexin	6,98	Primidone	7,88
15	Dexamethasone	7,12	Iohexol	6,88	Ciprofloxacine	7,75
16	Naproxene	6,98	Iopromide	6,88	Clotrimazole	7,75
17	Iohexol	6,88	Diclofenac	6,72	Flucloxacillin	7,75
18	Iopromide	6,88	Omeprazole	6,68	Penicillin V	7,75
19	Codeine	6,86	Codeine	6,66	Iomeprol	7,63
20	Cloxacillin	6,78	Clotrimazole	6,66	Ofloxacin	7,63

Benzodiazepines in the Danube River Basin

JDS3 - most relevant oxazepam and clobazam

Occurence frequencies: 85 and 31%

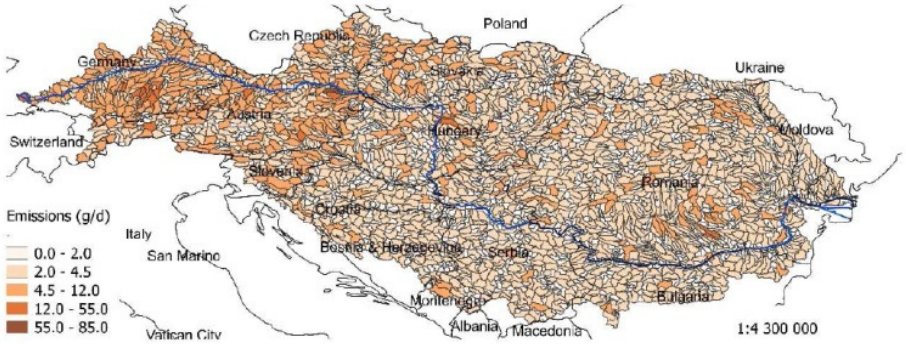
Cmax: oxazepam 15 ng/l (AT), clobazam 11 ng/l (DE)



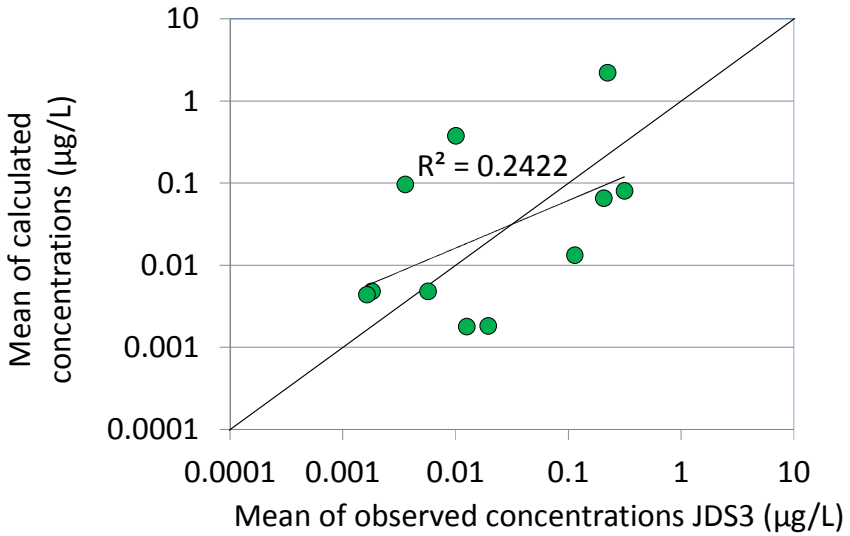
Fick et al., Screening of benzodiazepines in thirty European rivers, Chemosphere, accepted for publication; http://dx.doi.org/10.1016/j.chemosphere.2017.02.126

Modelling - First results for the Danube Basin

- Emission estimates and simulation of PFOS and PFOA
  Environmental Pollution 207 (2015) 97-106
  Chemosphere 144 (2016) 803-810

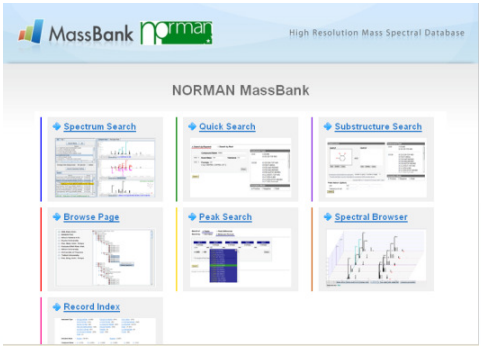


- 11 Industrial chemicals:
  - modelled emissions
  - modelled concentrations
- Comparison to measured concentrations at JDS3 stations
- Extension for additional > 8000 substances and Europe-wide

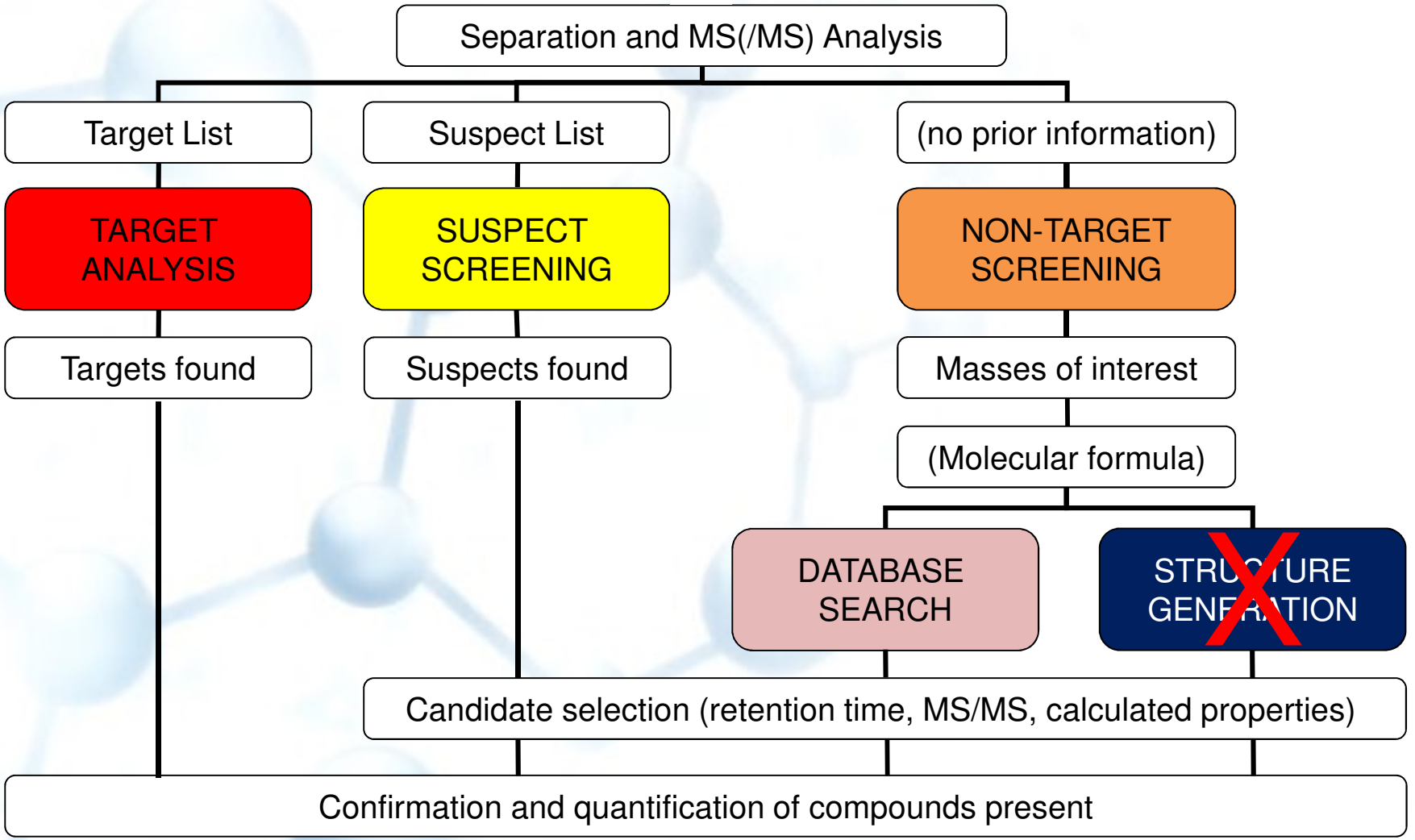


NORMAN Collaborative Trial 2013 - 2014 Non-target screening of organic substances in river water samples

Emma L. Schymanski, Heinz P. Singer, Jaroslav Slobodnik, Ildiko M. Ipolyi, Peter Oswald, Martin Krauss, Tobias Schulze, et al., Non-target screening with high-resolution mass spectrometry: critical review using a collaborative trial on water analysis, Anal. Bioanal. Chem., DOI 10.1007/s00216-015-8681-7, 2015.



Status quo of identification approaches for LC/GC-MS



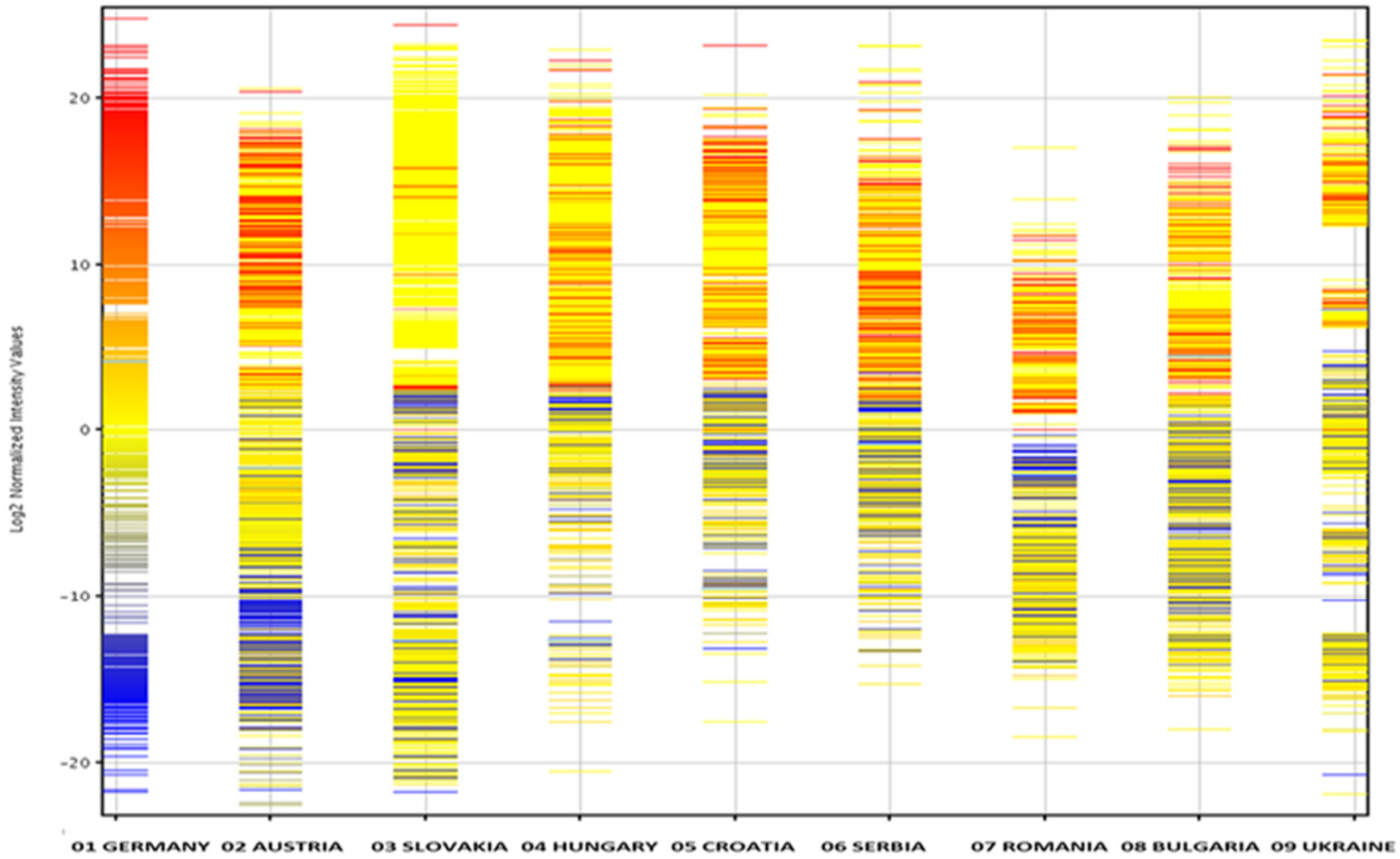
NORMAN CT – non target screening

Received datasets

	Name of organization / institute	Name of laboratory	Total DCTs	LC-MS	GC-MS
1	IAREN- Water Institute of the Northern Region	Laboratory of Chromatography			
2	NIVA	NIVA Oslo	1	1	
3	SUEZ Environment	CIRSEE	1	1	1
4	T. G. Masaryk Water Research Institue	Reference Laboratory for Environment Components and Waste			
5	University of Antwerp	Toxicological Centre	1	1	
6	Technische Universitaet Muenchen	Chair of Urban Systems Engineering	1	1	
7	University Jaume I (UJI)	Research Institute for Pesticides and Water	1		1
14	Ministry of the Environment of Canada	Laboratory Services Branch			
15	BRGM	Laboratory Division			
16	IRSTEA	LAMA	1	1	
17	Environmental Institute (EI), SK	Analytical Laboratory	1		1
18	Helmholtz-Centre for Environmental Research - UFZ	Effect-directed analysis	1	1	
19	University of Padua / Department of Chemistry	Group of Analytical Chemistry	1	1	
20	University of Bordeaux	team LPTC, laboratory EPOC (UMR 5805 CNRS)			
21	Masaryk University / Faculty of Science	RECETOX			
22	Bundesanstalt für Gewässerkunde	Gewässerchemie	1	1	
23	Zweckverband Landeswasserversorgung	Betriebs- und Forschungslaboratorium	1	1	
24	Croatian Waters	Central Water Management Laboratory	1	1	
25	University of Tuebingen	Environmental Chemistry	1	1	
26	University of Umea	Department of Chemistry	1		1
			18	**16**	**7**

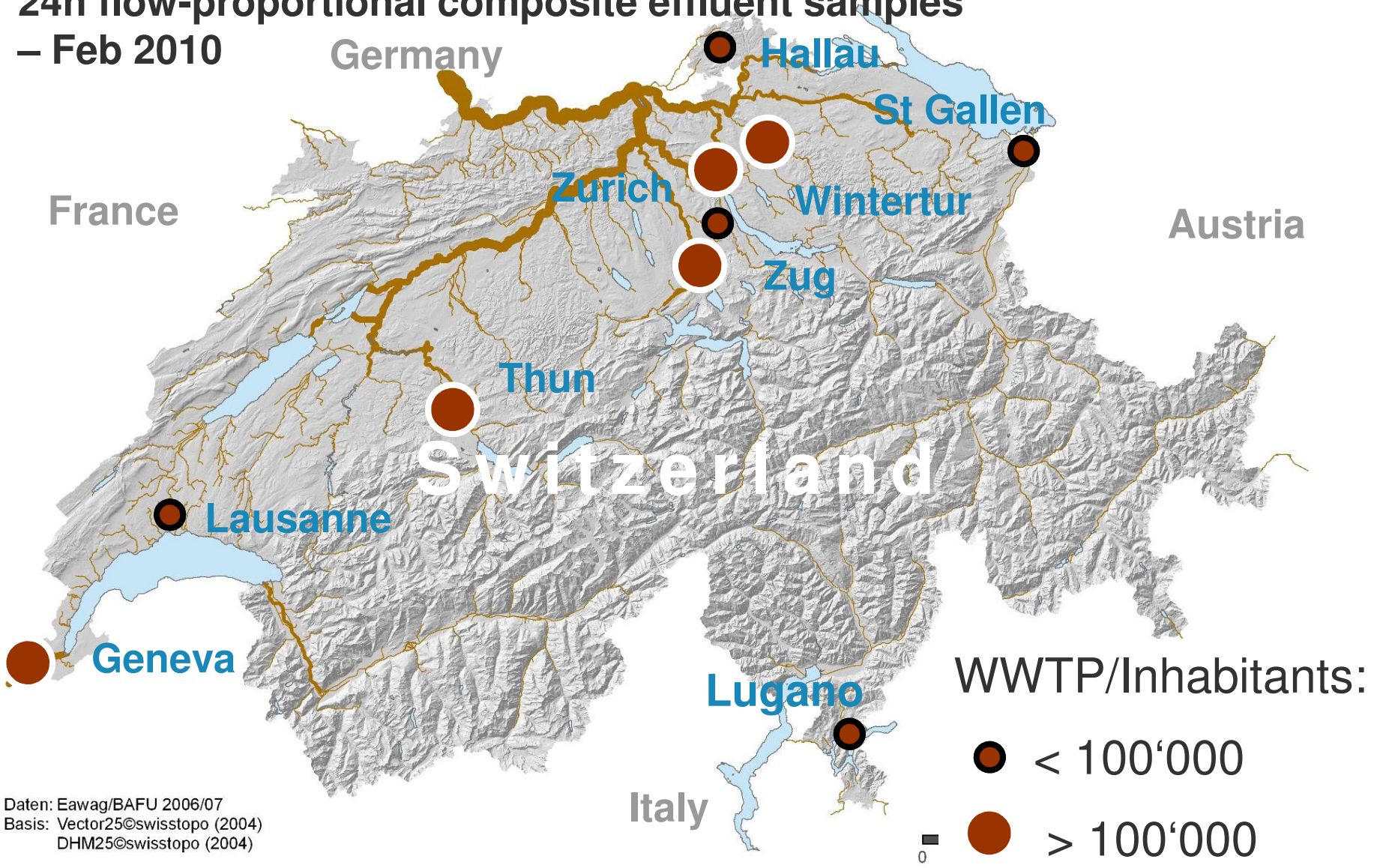
LC-HR-MS: T&S 1072; NT&Unknown 21380
GC-MS: T&S 348; NT&Unknown 116
Less then 5% overlap

Distribution of 7767 different mass spectral processed features through the Danube river and its tributaries; Danube countries are shown on x-axis and normalised signal intensity values are represented on y-axis; each single feature/compound is represented by a horizontal bar at a fixed position on the chart (position given by a unique combination of retention time, accurate mass spectrum, name, molecular formula, etc.) and the intensity of signal increase is indicated by blue (low) to red (high) colour

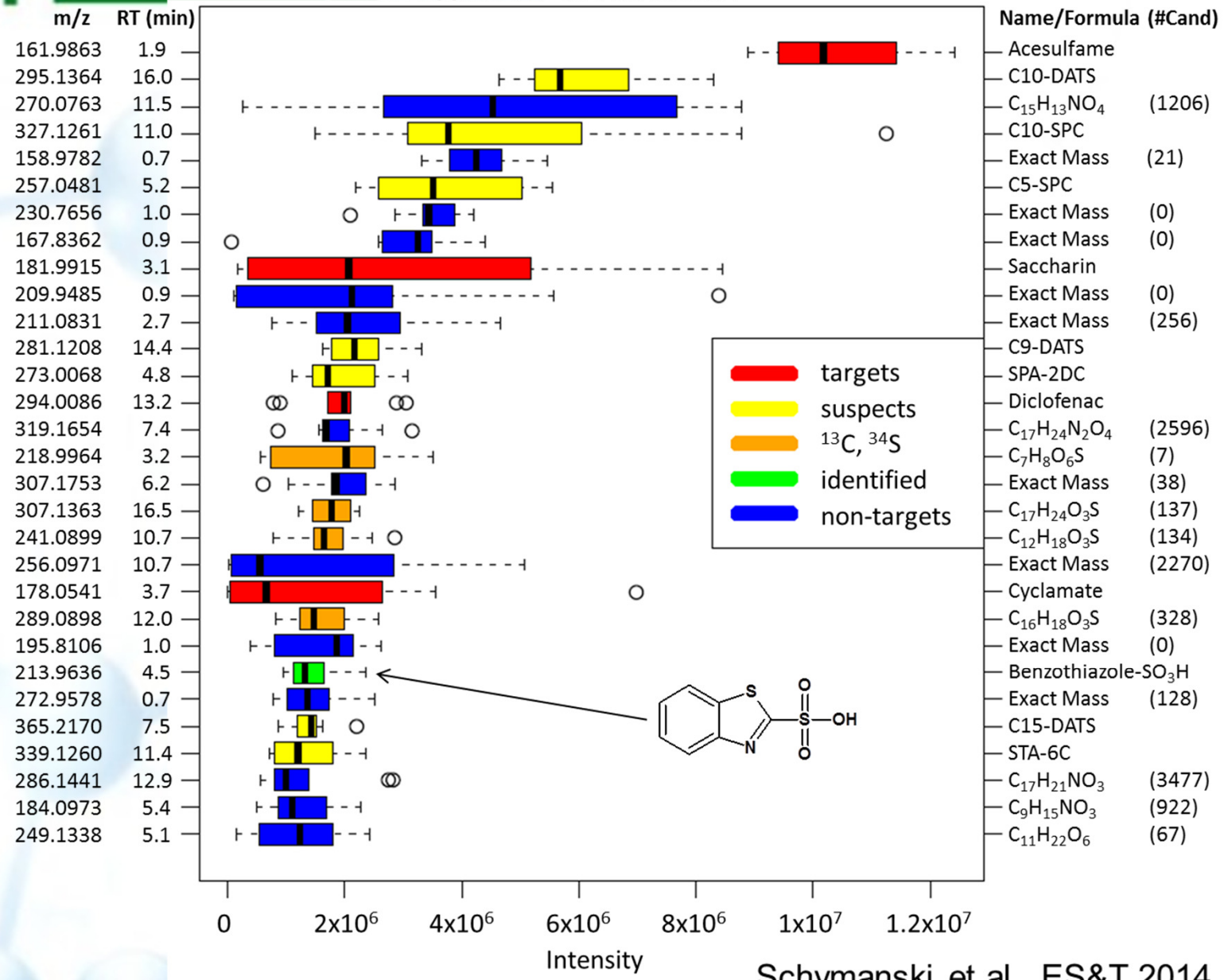


Example Switzerland: Sampling of 10 typical wastewater treatment plants

24h flow-proportional composite effluent samples – Feb 2010



Characterisation of wastewater effluent (10 plants)- ranking by presence and intensity – negative ionisation



Schymanski et al., ES&T 2014, 48:1811-1818

Template for storing LC-HRMS data

Template

- Organization Details
- RTI and RI calibration
- Sample meta-data
- Analysis
- Methods LC-MS(MS)
- Methods GC-MS(MS)

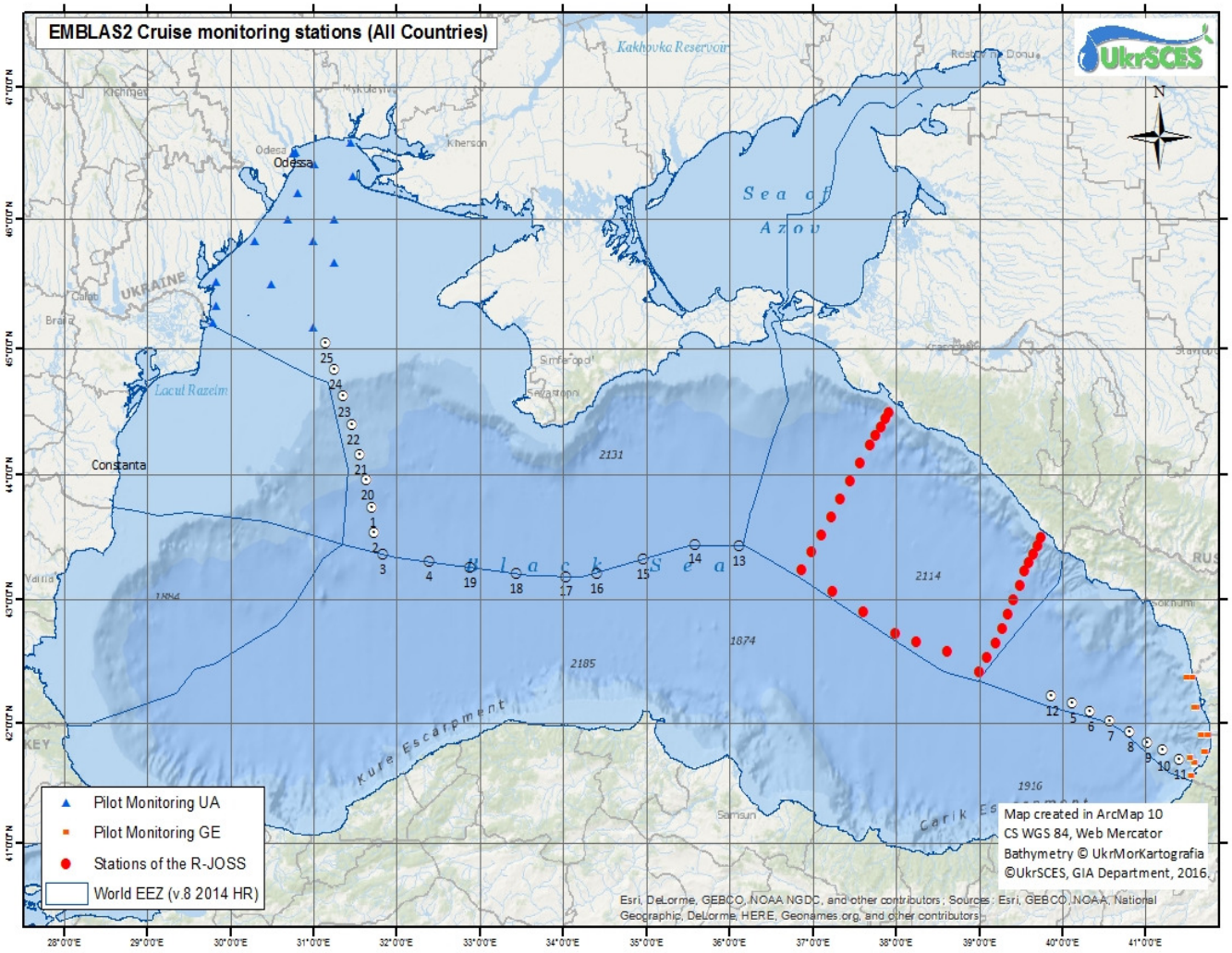
Sample identification (link to the raw data file name)	Retention time in the 1st column [min]	Retention time in the 2nd column [sec]	Mass of ion [m/z] (peak or component)	Intensity of the ion	Intensity of the ion in the blank	Ion type		MS/MS available	Category	Proposed identification (name of the substance or n.i. for not identified)	Molecular formula	Exact. Mass
							Other					

Identifier: SMILES	CAS No.	Estimated concentration [ug/l]	Level of confirmation of identification	Component information	Optional		
				Fragment masses of detected compounds	Retention Time Index LC-MS (Letzel's index; other index)	Retention Time Index LC-MS (UoA approach)	Retention Time Index GC-MS (Kovat's index)

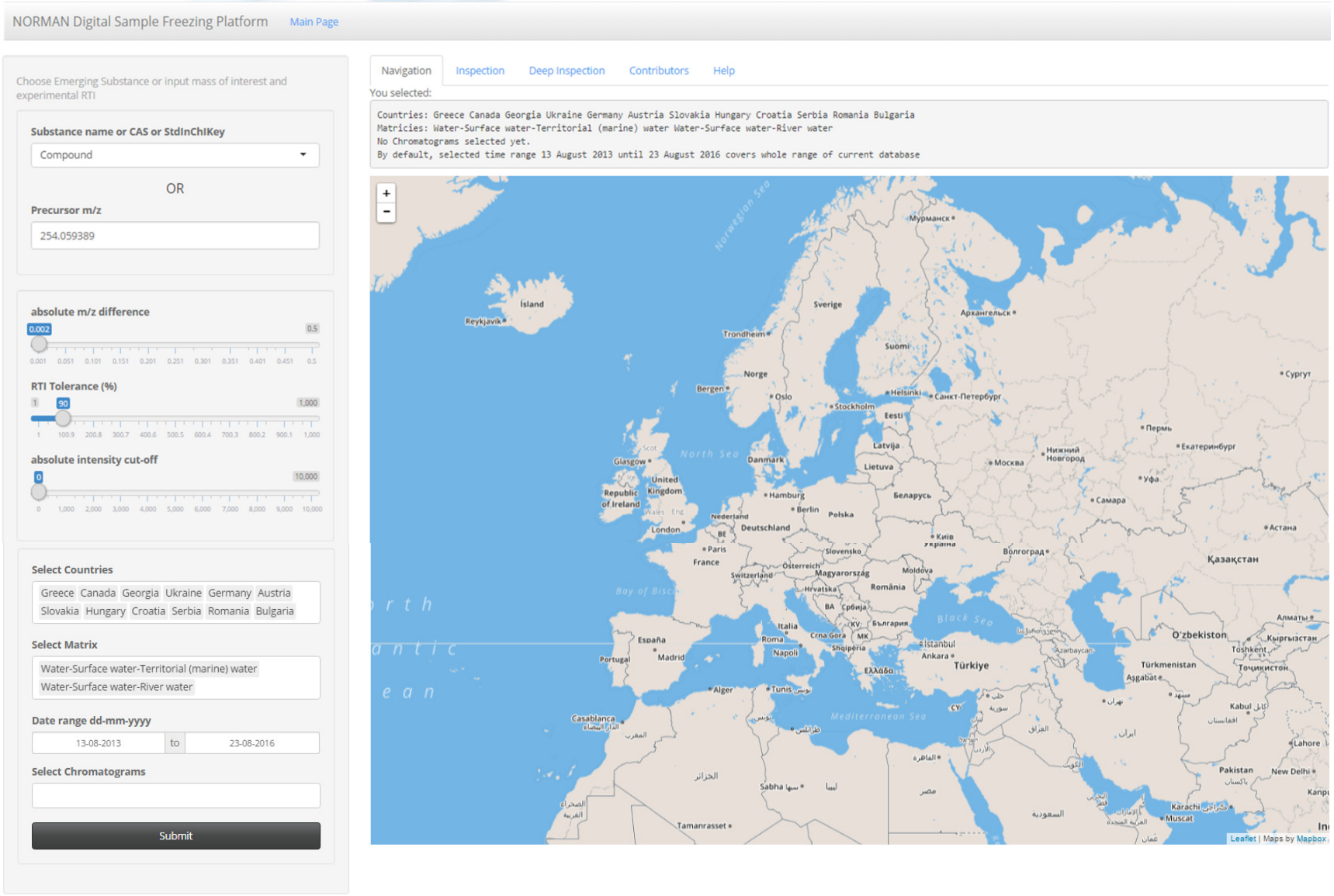
Date of sampling (DD/MM/YYYYY)	Date of analysis (DD/MM/YYYYY)	Serial No. in Method LC-MS(MS) or GC-MS(MS) worksheet	LC-MS - files attached				GC-MS - files attached
			Raw chromatogram: Positive/negative mode; different CE; MS-MS... (ShortName ORGANISATION_PROJECT_SampleID_Instrument_Ionisation_IonisationMode_Fragmentation-eV_DDMMYYYY.ext)				Raw chromatogram; EI/PCI/NCI; MS/MS... (mzXML) (ShortName ORGANISATION_PROJECT_SampleID_Instrument_IonisationMode_Fragmentation_DDMMYYYY.ext)
			No. of peaks	Intensity cut-off value	Data analysis report (mzML/mzXML)	Data aquisition	

www.emblasproject.org

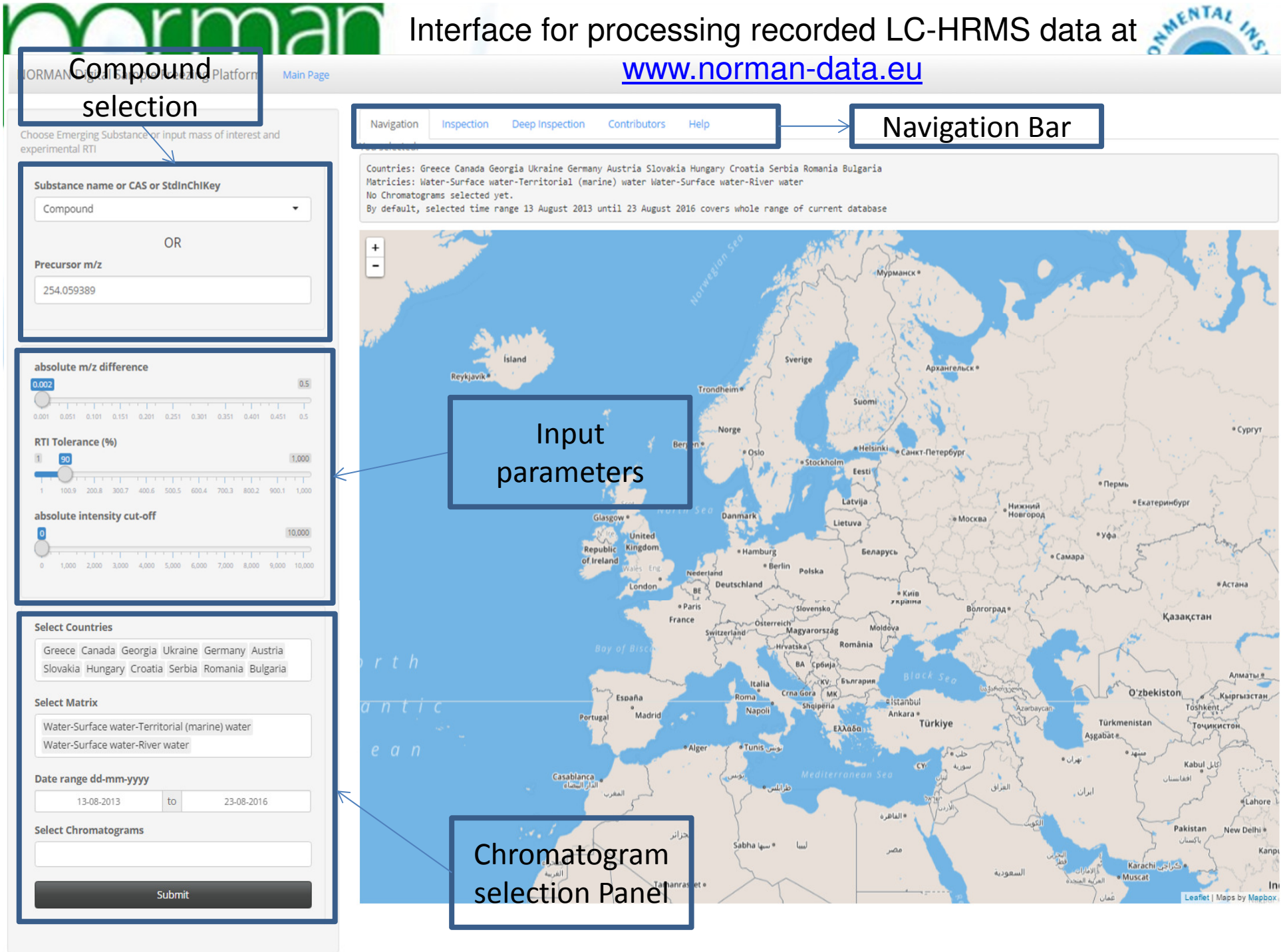
National pilot monitoring studies (NPMS)/Joint Open Sea Survey (JOSS) – May-June 2016 – August/September 2017



Interface for processing recorded LC-HRMS data at www.norman-data.eu



Interface for processing recorded LC-HRMS data at www.norman-data.eu



NORMAN-SusDat at http://www.norman-network.net/datatable/

NORMAN-SusDat: NORMAN Suspect List Exchange Merged Data Table

Reset search results

Show 100 entries

Name	Monoisotopic_mass	Formula	CAS_No	ChemSpiderID	Pred_RTI_Positive_ESI	Pred_RTI_Negative_ESI	MS_Ready_SMILES
à,à'-Dihydroxy-m-diisopropylbenzene	194.130679816	C12H18O2	1999-85-5	67383	469.722663392872	403.859229291849	c1c(cccc1C(C)(C)O)C(C)(C)O
à-resorcylic acid	154.0266	C7H6O4	99-10-5	7146	254.997123336099	100.133600509058	OC(=O)c1cc(O)cc(O)c1
α,α'-propylenedinitrilodi-o-cresol	282.1368	C17H18N2O2	94-91-7	4512376	547.17350520936	356.211474862575	CC(CN/C=C\1/C=CC=CC1=O)N/C=C\1/C=CC=CC1=O
α,α,α,β,β,β-hexafluoro-m-xylene	214.0217	C8H4F6	402-31-3	21111807	641.346948252083	563.215944057811	FC(F)(F)c1cccc(c1)C(F)(F)F

Choose Emerging Substance or input mass of interest and experimental RTI

Substance name or CAS or StdInChIKey

Benzotriazole

Choose Ionization

Positive

Adduct

M+H

Predicted RTI Positive

-94 266 981

-94 14 122 230 338 446 554 662 770 878 981

Reset

OR

Cases in which substance is not in SusDat

Choose Emerging Substance or input mass of interest and experimental RTI

Precursor m/z

254.05938905

Choose Ionization

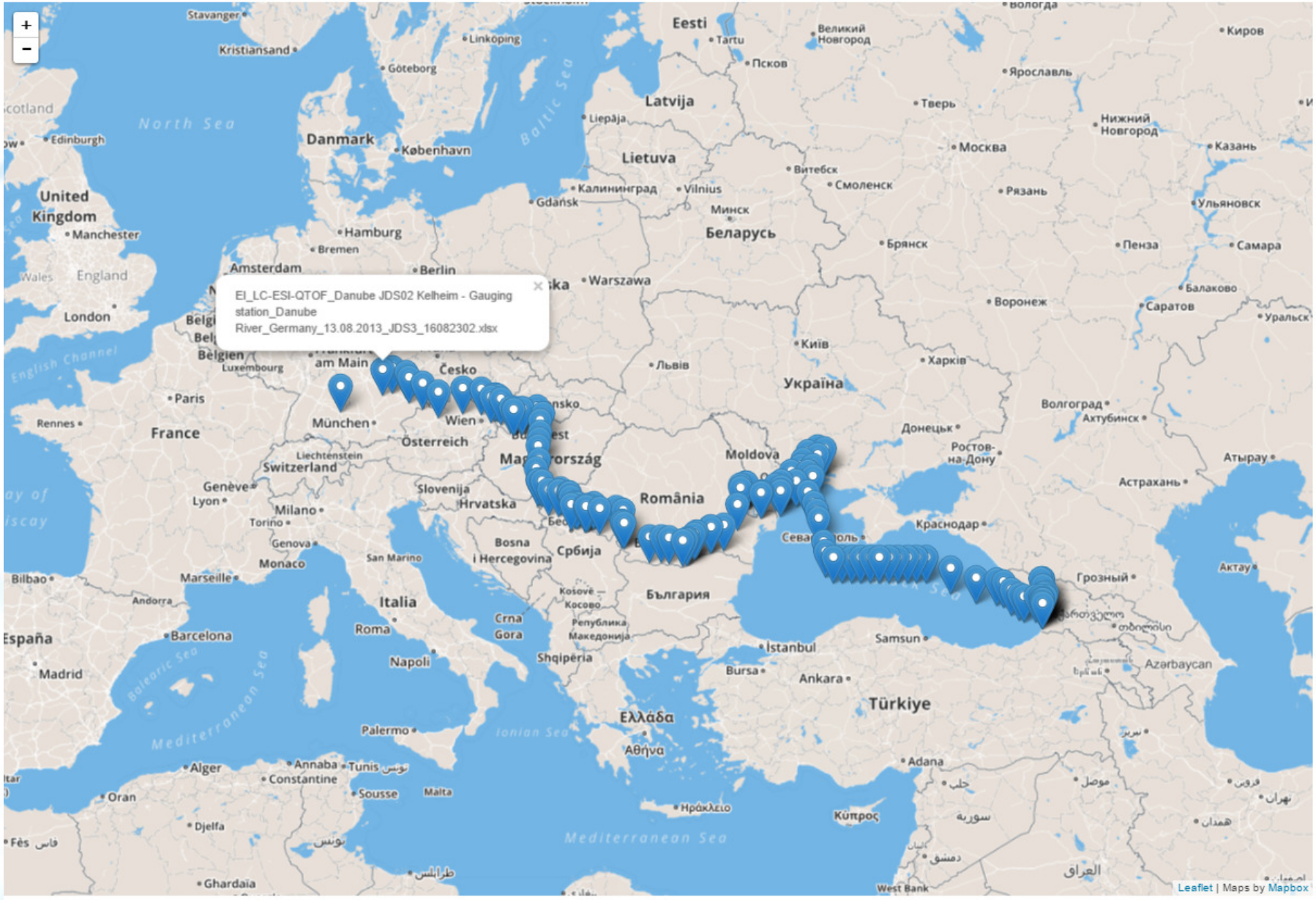
Positive

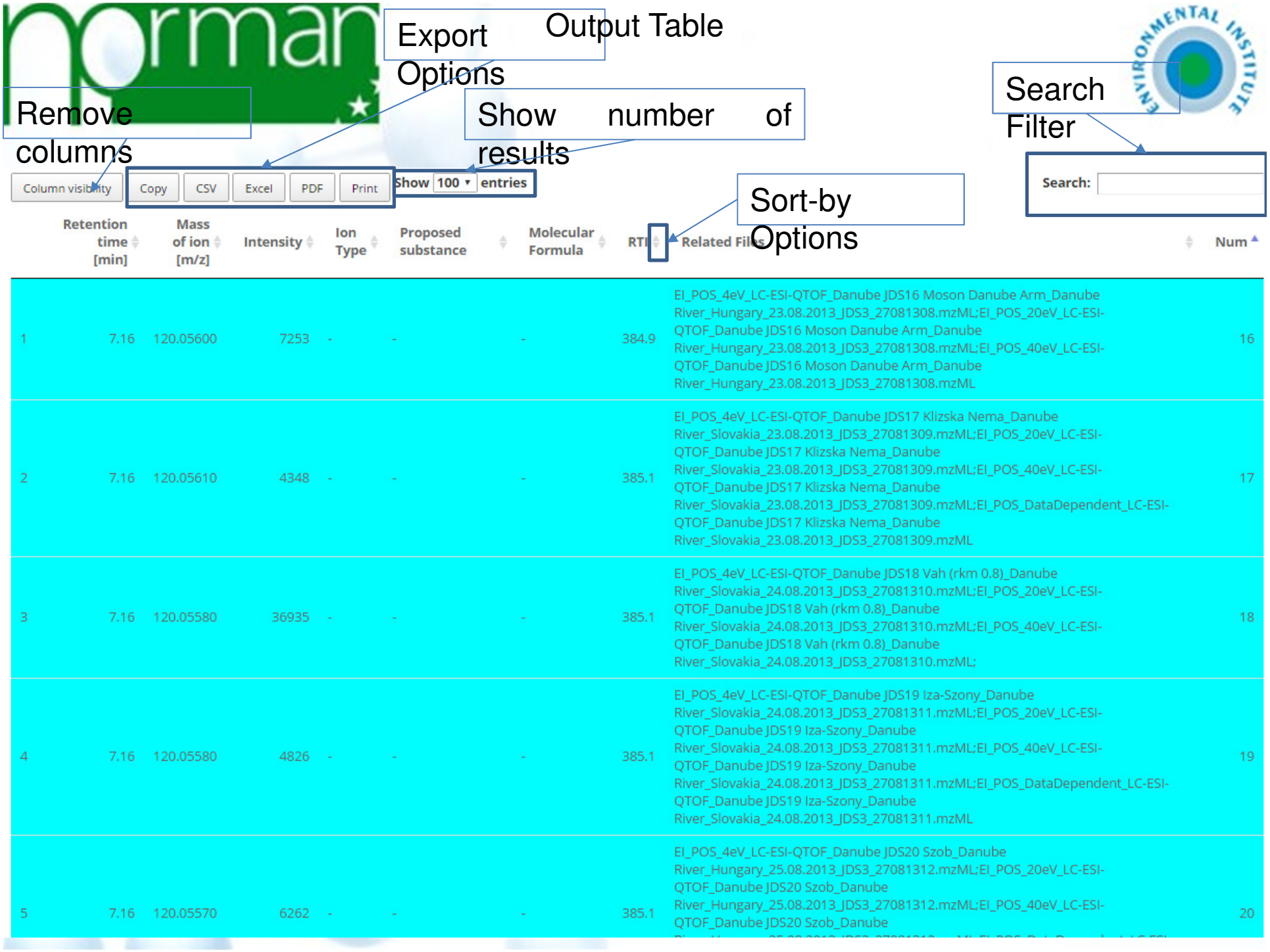
Experimental RTI

500

Reset

Selection of River water samples from JDS3 (68) and EMBLAS seawater samples (55)





	Retention time [min]	Mass of ion [m/z]	Intensity	Ion Type	Proposed substance	Molecular Formula	RTI	Related Files	Num
1	7.16	120.05600	7253	-	-	-	384.9	EI_POS_4eV_LC-ESI-QTOF_Danube JDS16 Moson Danube Arm_Danube River_Hungary_23.08.2013_JDS3_27081308.mzML;EI_POS_20eV_LC-ESI-QTOF_Danube JDS16 Moson Danube Arm_Danube River_Hungary_23.08.2013_JDS3_27081308.mzML;EI_POS_40eV_LC-ESI-QTOF_Danube JDS16 Moson Danube Arm_Danube River_Hungary_23.08.2013_JDS3_27081308.mzML	16
2	7.16	120.05610	4348	-	-	-	385.1	EI_POS_4eV_LC-ESI-QTOF_Danube JDS17 Klizska Nema_Danube River_Slovakia_23.08.2013_JDS3_27081309.mzML;EI_POS_20eV_LC-ESI-QTOF_Danube JDS17 Klizska Nema_Danube River_Slovakia_23.08.2013_JDS3_27081309.mzML;EI_POS_40eV_LC-ESI-QTOF_Danube JDS17 Klizska Nema_Danube River_Slovakia_23.08.2013_JDS3_27081309.mzML;EI_POS_DataDependent_LC-ESI-QTOF_Danube JDS17 Klizska Nema_Danube River_Slovakia_23.08.2013_JDS3_27081309.mzML	17
3	7.16	120.05580	36935	-	-	-	385.1	EI_POS_4eV_LC-ESI-QTOF_Danube JDS18 Vah (rkm 0.8)_Danube River_Slovakia_24.08.2013_JDS3_27081310.mzML;EI_POS_20eV_LC-ESI-QTOF_Danube JDS18 Vah (rkm 0.8)_Danube River_Slovakia_24.08.2013_JDS3_27081310.mzML;EI_POS_40eV_LC-ESI-QTOF_Danube JDS18 Vah (rkm 0.8)_Danube River_Slovakia_24.08.2013_JDS3_27081310.mzML;	18
4	7.16	120.05580	4826	-	-	-	385.1	EI_POS_4eV_LC-ESI-QTOF_Danube JDS19 Iza-Szony_Danube River_Slovakia_24.08.2013_JDS3_27081311.mzML;EI_POS_20eV_LC-ESI-QTOF_Danube JDS19 Iza-Szony_Danube River_Slovakia_24.08.2013_JDS3_27081311.mzML;EI_POS_40eV_LC-ESI-QTOF_Danube JDS19 Iza-Szony_Danube River_Slovakia_24.08.2013_JDS3_27081311.mzML;EI_POS_DataDependent_LC-ESI-QTOF_Danube JDS19 Iza-Szony_Danube River_Slovakia_24.08.2013_JDS3_27081311.mzML	19
5	7.16	120.05570	6262	-	-	-	385.1	EI_POS_4eV_LC-ESI-QTOF_Danube JDS20 Szob_Danube River_Hungary_25.08.2013_JDS3_27081312.mzML;EI_POS_20eV_LC-ESI-QTOF_Danube JDS20 Szob_Danube River_Hungary_25.08.2013_JDS3_27081312.mzML;EI_POS_40eV_LC-ESI-QTOF_Danube JDS20 Szob_Danube	20

Navigation | Inspection | Deep Inspection | Contributors | Help

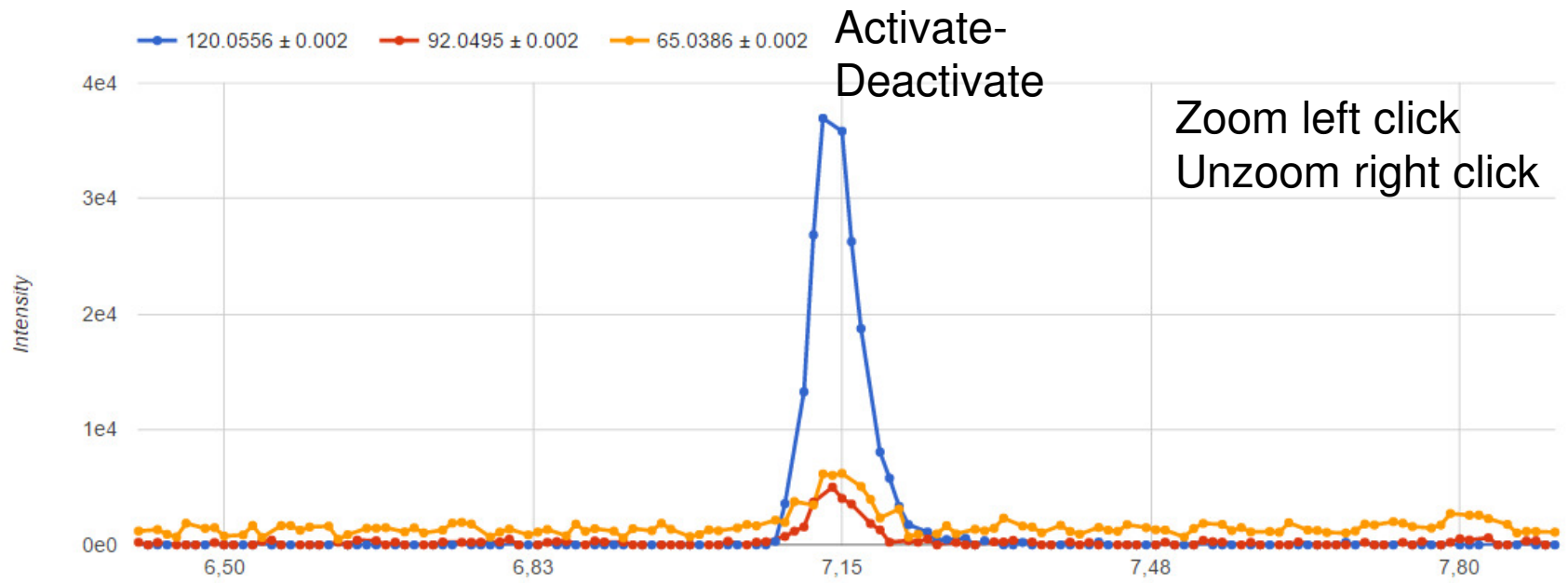
Select Chromatogram

EI_LC-ESI-QTOF_Danube JDS18 Vah (rkm 0.8)_Danube River_Slovakia_24.08.2013_JDS3_27081310.xlsx

	mz	accuracy_mDa	chromatogram	show
1	120.0556	0.0020	EI_POS_4eV_LC-ESI-QTOF_Danube JDS18 Vah (rkm 0.8)_Danube River_Slovakia_24.08.2013_JDS3_27081310.mzML	☑
2	92.0495	0.0020	EI_POS_20eV_LC-ESI-QTOF_Danube JDS18 Vah (rkm 0.8)_Danube River_Slovakia_24.08.2013_JDS3_27081310.mzML	☑
3	65.0386	0.0020	EI_POS_40eV_LC-ESI-QTOF_Danube JDS18 Vah (rkm 0.8)_Danube River_Slovakia_24.08.2013_JDS3_27081310.mzML	☑

More extracted ion chromatograms in different collision energy layers

Submit (Press the button everytime changes are done to the table above)



Successful identification of Benzotriazole (mz=120.0556 and qualifiers 92.0495, 65.0386) in sample JDS18

Navigation | Inspection | Deep Inspection | Contributors | Help

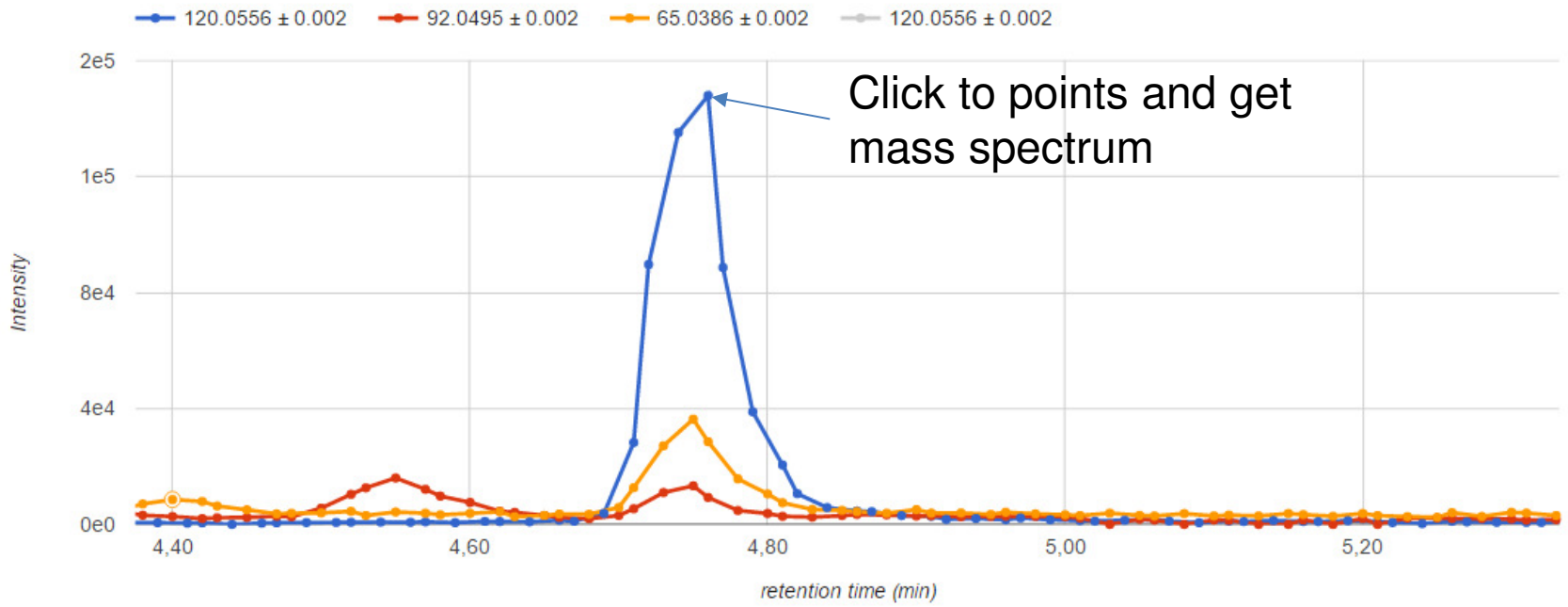
Select Chromatogram

UoA_LC-ESI-QTOF_Seawater UA07_Black Sea_Ukraine_19.05.2016_EMBLAS II_17856.xlsx

	mz	accuracy_mDa	chromatogram	show
1	120.0556	0.0020	UoA_POS_4eV_LC-ESI-QTOF_Seawater UA07_Black Sea_Ukraine_19.05.2016_EMBLAS II_17856.mzML	☑
2	92.0495	0.0020	UoA_POS_25eV_LC-ESI-QTOF_Seawater UA07_Black Sea_Ukraine_19.05.2016_EMBLAS II_17856.mzML	☑
3	65.0386	0.0020	UoA_POS_25eV_LC-ESI-QTOF_Seawater UA07_Black Sea_Ukraine_19.05.2016_EMBLAS II_17856.mzML	☑
4	120.0556	0.0020	UoA_POS_DataDependent_LC-ESI-QTOF_Seawater UA07_Black Sea_Ukraine_19.05.2016_EMBLAS II_18152.mzML	☑

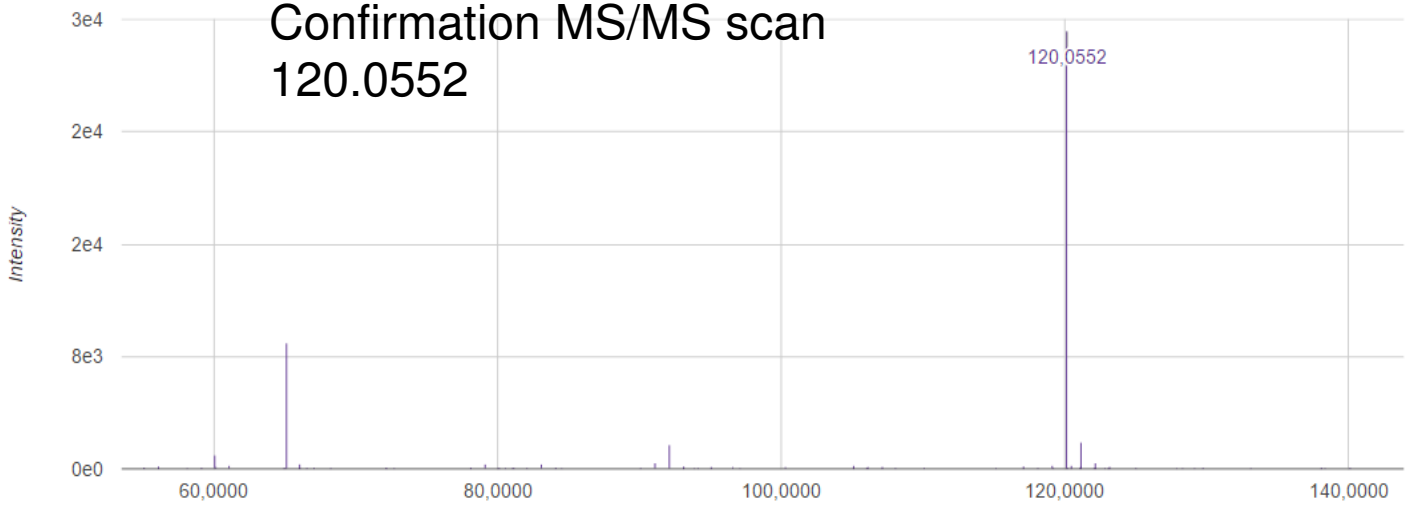
Data dependent chromatogram available!!!

Submit (Press the button everytime changes are done to the table above)

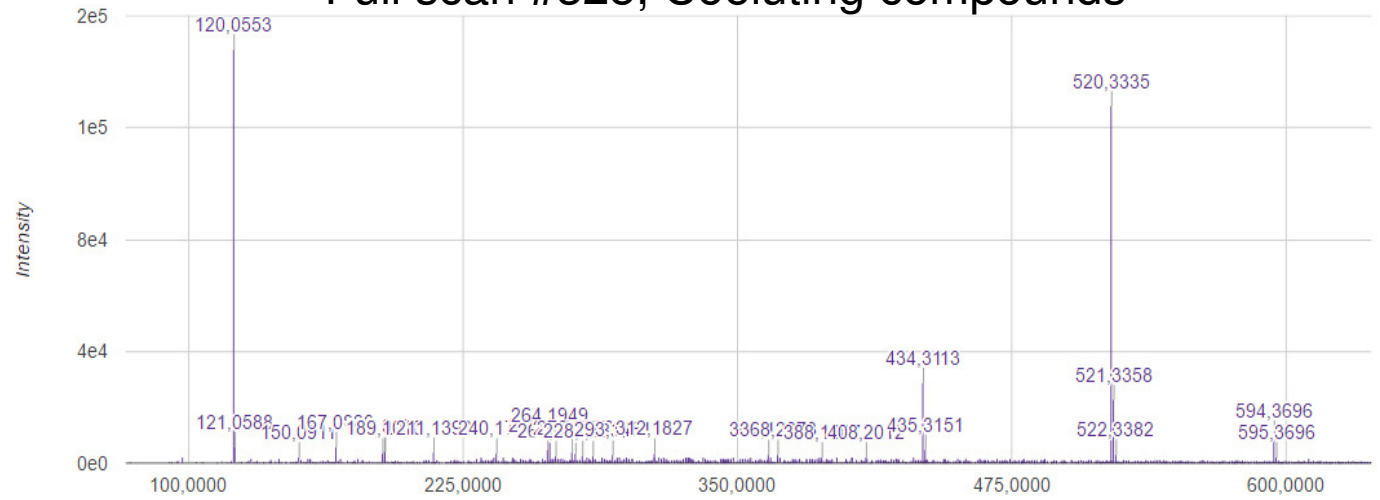


Successful identification of Benzotriazole (mz=120.0556 and qualifiers 92.0495, 65.0386) in sample UA07

Confirmation MS/MS scan
120.0552



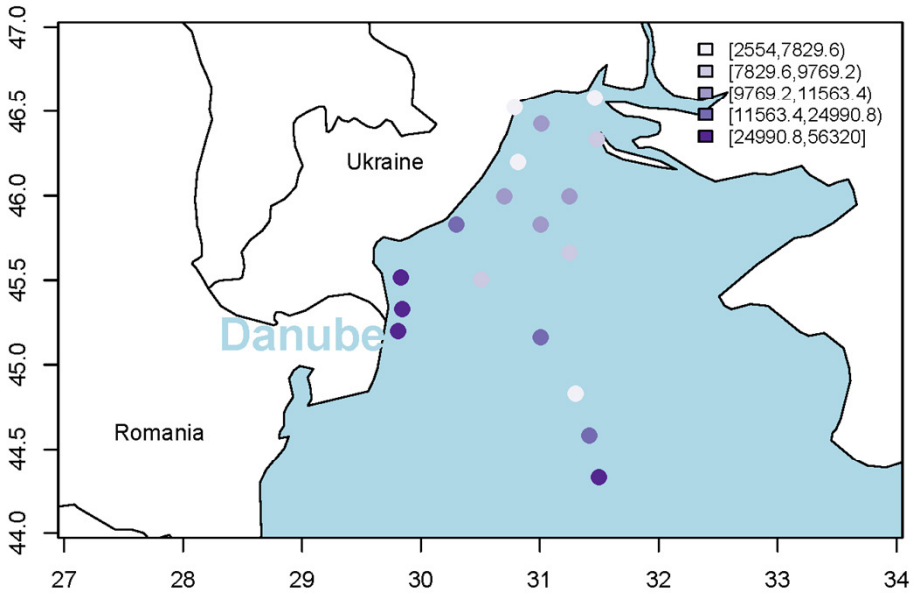
Full scan #325, Coeluting compounds

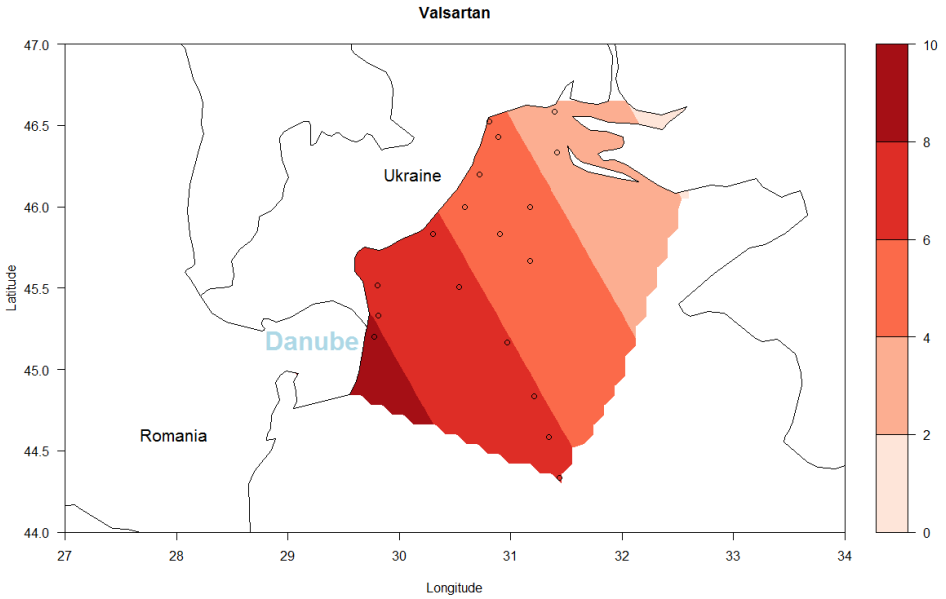


- ✓ Benzotriazole detected in 56 samples (27/68 JDS3 and 29/55 Black Sea)
- ✓ Metformin detected in 48 samples (29/68 JDS3 and 19/55 Black Sea)

EMBLAS – UA shelf of the Black Sea 15512 PI and 4928 NI components; statistical analysis - 633 substances certainly transported from the Danube to the Black Sea; 31 substances found retrospectively in both JDS3 (2013) and NPMS UA (2016) datasets

Valsartan





Name	[M+H]+	Molecular Formula	error [mDa]	error [ppm]	ΔRT	mSigma	Q1	Q2	Q3	Identification Level
Metolachlor	284.1412	C15H22ClNO2	-0.30	-1.06	0.17	25.4	252.1151	176.1427	212.0826	1
Metolachlor-ESA	330.137	C15H23N1O5S1	0.10	0.30	0.08	103*	298.1106	-	-	1
4-Acetamidoantipyrin	246.1237	C13H15N3O2	1.00	4.06	0.07	26.5	228.1127	204.1124	159.0905	1
4-Formylaminoantipyrine	232.1081	C12H13N3O2	0.50	2.15	0.06	17.2	214.0975	83.0604	187.0865	1
Benzotriazole (BTR)	120.0556	C6H5N3	-0.80	-6.66	0.01	35	92.0494	65.0384	-	1
Cotinine	177.1022	C10H12N2O1	-0.50	-2.82	0.13	38.5	80.0495	98.0600	70.06513	1
cotinine-Hydroxy	193.0972	C10H12N2O2	-0.40	-2.07	0.16	8.8	80.0495	134.0600	-	1
Metformin	130.1087	C4H11N5	-0.50	-3.84	0.13	6.1	71.0604	60.0556	85.0517	1
Carbamazepine	237.1022	C15H12N2O1	0.70	2.95	0.16	11.8	194.0964	192.0806	-	1
Dimethenamide	276.082	C12H18ClNO2S	0.00	0.00	0.13	19.3	244.0557	-	-	1
Atenolol acid (Metoprolol acid)	268.1543	C14H21N1O4	0.40	1.49	0.17	93.7*	191.0702	116.1070	-	1
Metoprolol	268.1907	C15H25N1O3	-2.40	-8.95	0.05	17.4	74.0607	116.1070	-	1

Result of retrospective analysis

Information about compound and site detected

NAME	SMILES	Area	Intensity	Sample
Sulfamethoxazole	1cc(no1)NS(=O)(=O)c2ccc(cc2	18465	2836	Danube Site 52

Mass accuracy

Exp. m/z	Theor. m/z	Error (ppm)/mDa
254.0609	254.0594	5.9/1.5

Retention time index

Exp Rt	Pred. Rt	Error Rt
4.68	4.25	0.43

Isotopic fit match

Isotopic Fit
95.6

Qualifier Ions (mass accuracy and isotopic fit)

Q1 (ppm/isotopic fit)	Q2 (ppm/isotopic fit)	Q3 (ppm/isotopic fit)	Score
-2.21/85	-1.02/79	N.D.	86

Combined score indicating quantitative result of identification confidence

TrAMS Trace Analysis and Mass Spectrometry Group

enviPath

HighChem Identification made simple

Database of 676 antibiotics

EnviPath SW for generation of transformation products

Database of >20,000 TPs

SEARCH in digitally stored samples if present

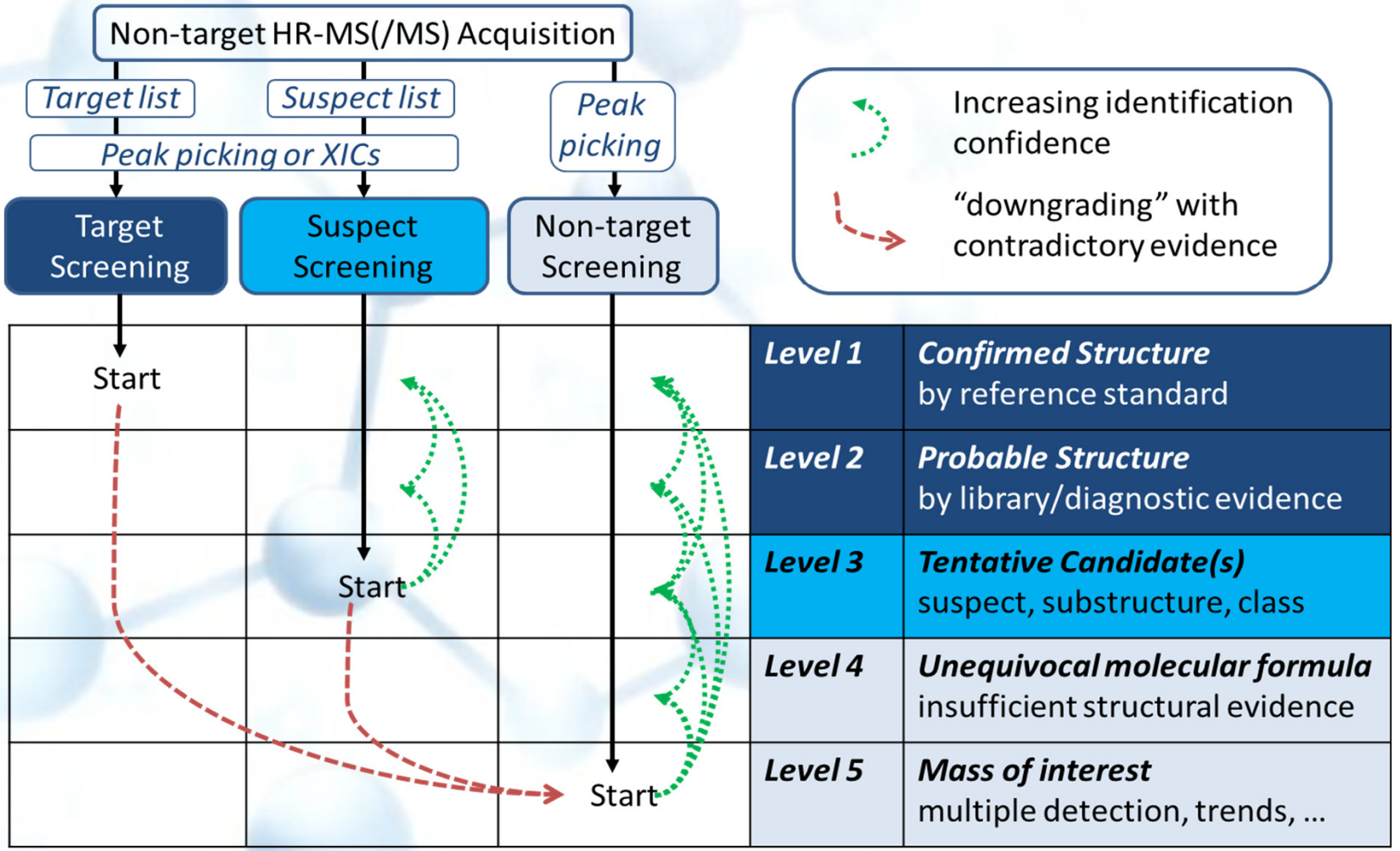
Derive PNECs

Prioritise

H2020-MSCA-ITN-2015/675530, ANtibioticS and mobile resistance elements in WastEwater Reuse applications: risks and innovative solutions

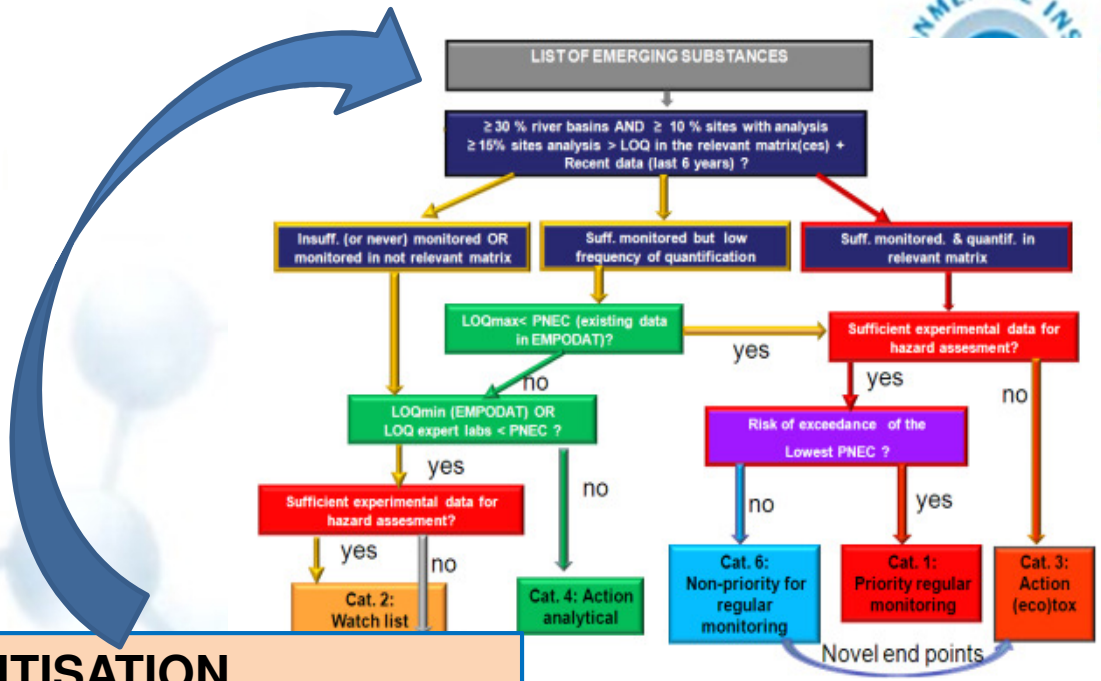
ANSWER

Non-target screening – categorisation and prioritisation



Schymanski et al., 2015. Non-target screening with high resolution mass spectrometry: Critical review using a collaborative trail on water analysis. Analytical & Bioanalytical Chemistry.

Non-target screening
prioritisation



GO TO NORMAN TARGET PRIORITISATION

NT CATEGORY 1 - FoA* + EoE + FoE (0-3)

NT CATEGORY 2A - FoA* + EoE + FoE (semiquantitative info available)
NT CATEGORY 2B – FoA* (0-1; semiquantitative info NOT available)

NT CATEGORY 3 – FoA*

NT CATEGORY 4 – FoA*

* Cut-off: 4 countries, 100 sites detected

10 Recommendations for the review of the WFD

(STOTEN 576 (2017) 720–737)

Recommendation: A key objective for the prioritization of substances and for monitoring programs should be to **minimize the risk of overlooking chemicals** that might pose significant threats to ecosystems and environmental health. This may be achieved **by prioritization of individual chemicals on a broader basis** compared to currently used schemes.

Outlook. Enhanced efforts will be required to **integrate suspect and non-target screening approaches into the prioritization process** and to establish open access databases and **multivariate analysis** of environmentally detected **chemical patterns** and **toxicity profiles**.

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Towards the review of the European Union Water Framework management of chemical contamination in European surface water resources

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Conclusions

It is not possible to develop the necessary knowledge and methodologies solely at the national scale

- **Usage data – exposure index**
- **Bioassays for mixture effects – trigger values**
- Continuous **improvement of analytical expertise and data quality**
- **European database** for storage of large dataset – raw full scan MS chromatograms for retrospective analysis
- Common views of the scientific community on research needs and **priorities for future legislation – DATABASE WORKSHOP, Berlin, 8-9 June 2017**

http://www.norman-network.net/
http://www.solutions-project.eu/