

Für Mensch & Umwelt

Umwelt 
Bundesamt

WaBoLu Wasserkurs 2023

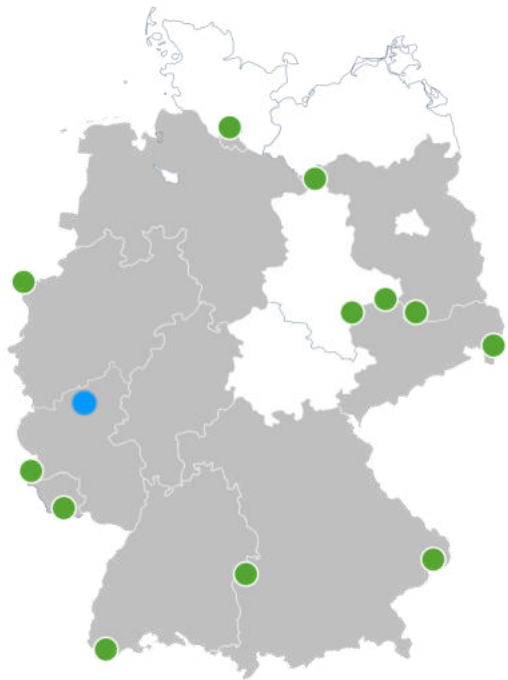
Non-Target Screening für die Gewässerbeobachtung der Zukunft

Anna Lena Kronsbein¹, Alexander Badry¹, Eric Rosenheinrich¹, Tobias Schulze¹, Kevin S. Jewell²,
Nicole Bandow¹, Jan Koschorreck¹

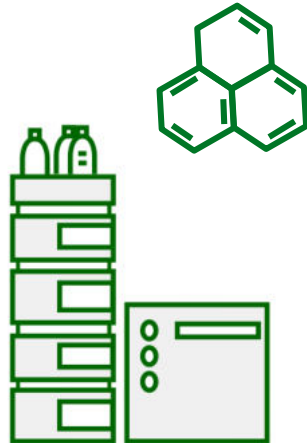
¹ Umweltbundesamt

² Bundesanstalt für Gewässerkunde

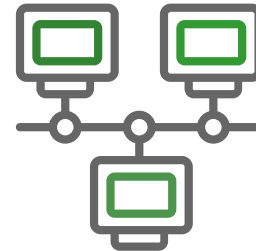
NTS-Daten für die Gewässerbeobachtung der Zukunft



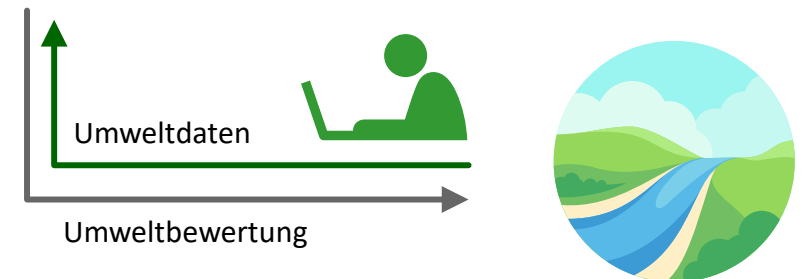
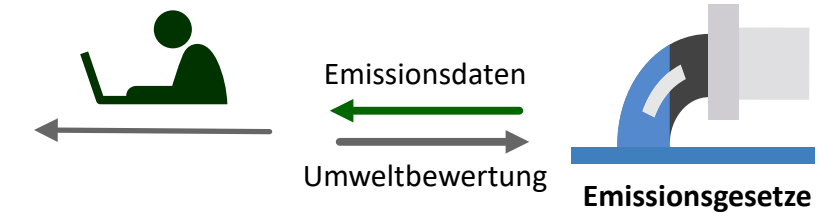
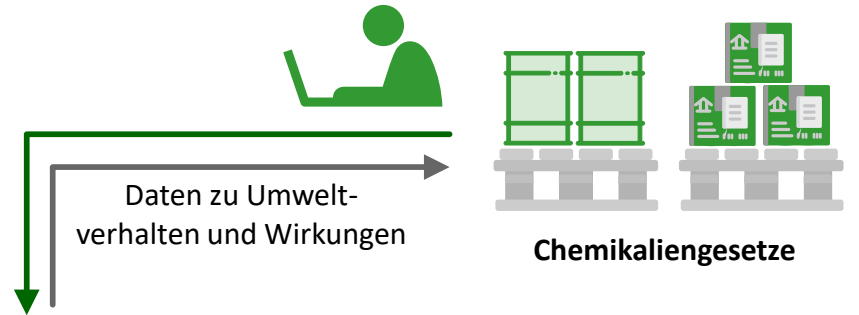
- UPB Jahrestrends
- BfG Tagestrends
- Bundesländerdaten



Stoffe und Mischungen

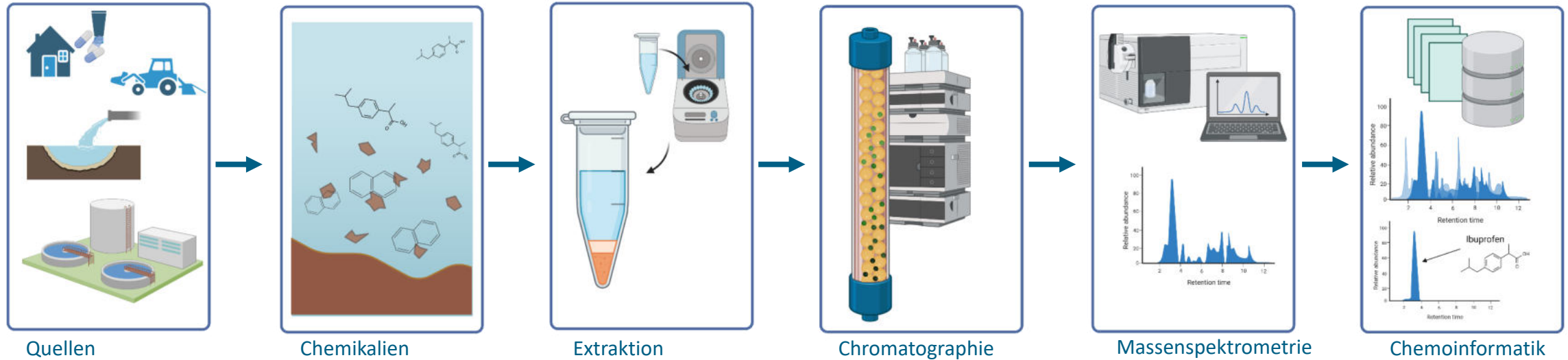


Gewässerbeobachtung der Zukunft

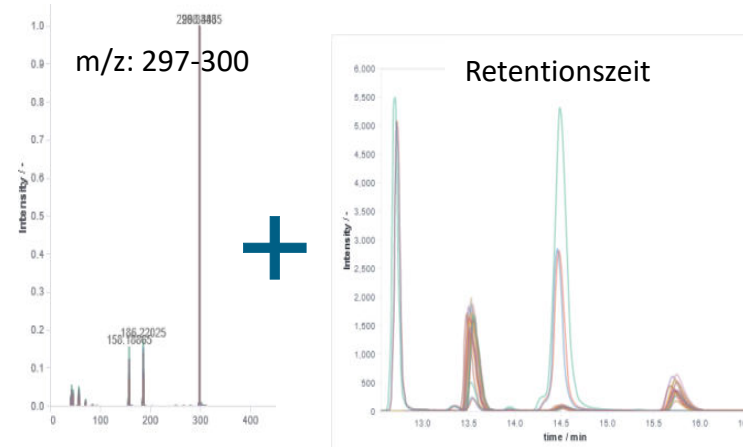


NTS: Von der Umweltprobe zur Substanzliste

Created with BioRender.com

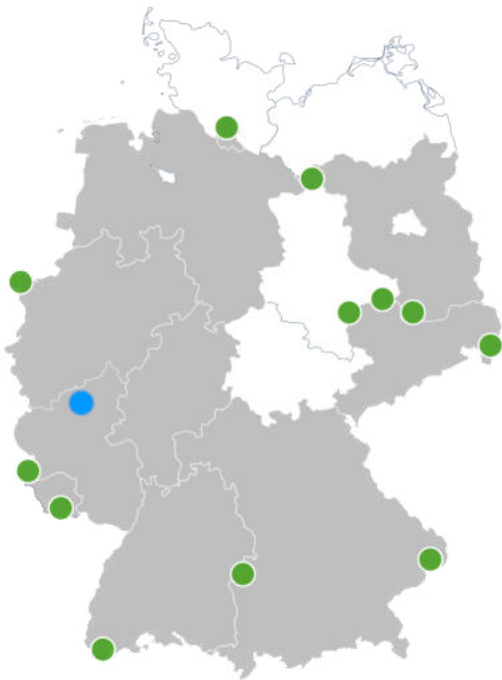


➔ Substanzlisten

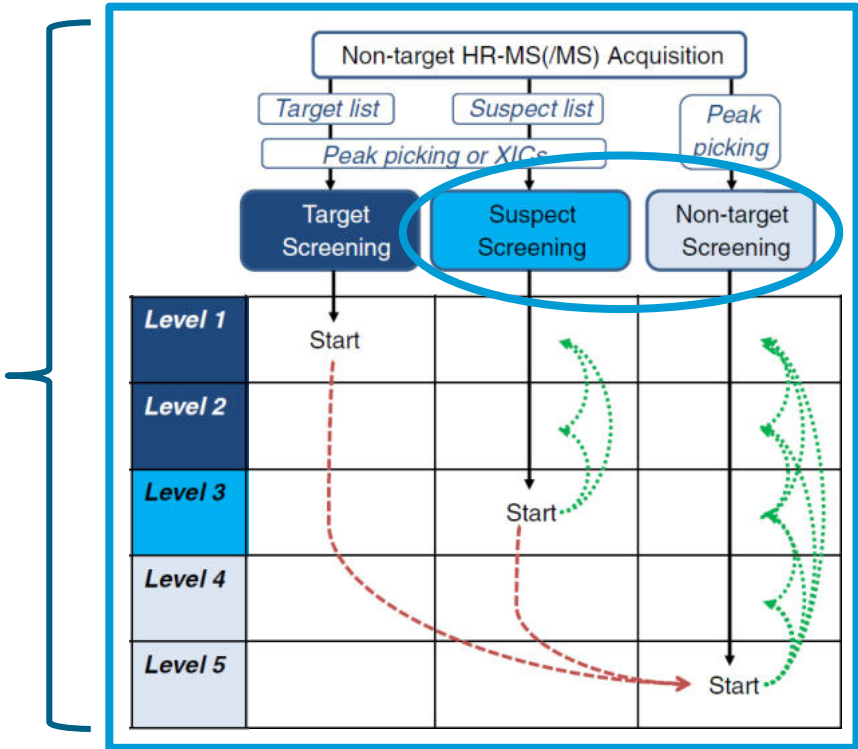
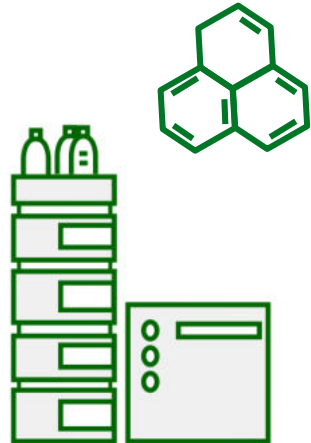


Compoun...	Polarity	m/z	Min m/z	Max m/z	RT (min)
5'-Deoxy-5'...	pos	298.0962	298.0929	298.0984	4.89
Octyl-decyl...	pos	298.346	298.3431	298.3486	13.06
thyl-hexyl...	pos	298.3458	298.3422	298.3481	13.82
6PP...	pos	299.1739	299.1706	299.1769	14.07
...	pos	298.2726	298.2699	298.2751	9.7
6-Methylthio...	neg	297.065	297.0613	297.0713	5.13
Perfluorobut...	neg	298.9419	298.9407	298.9437	10.33

NTS-Daten für die Gewässerbeobachtung der Zukunft



- UPB Jahrestrends
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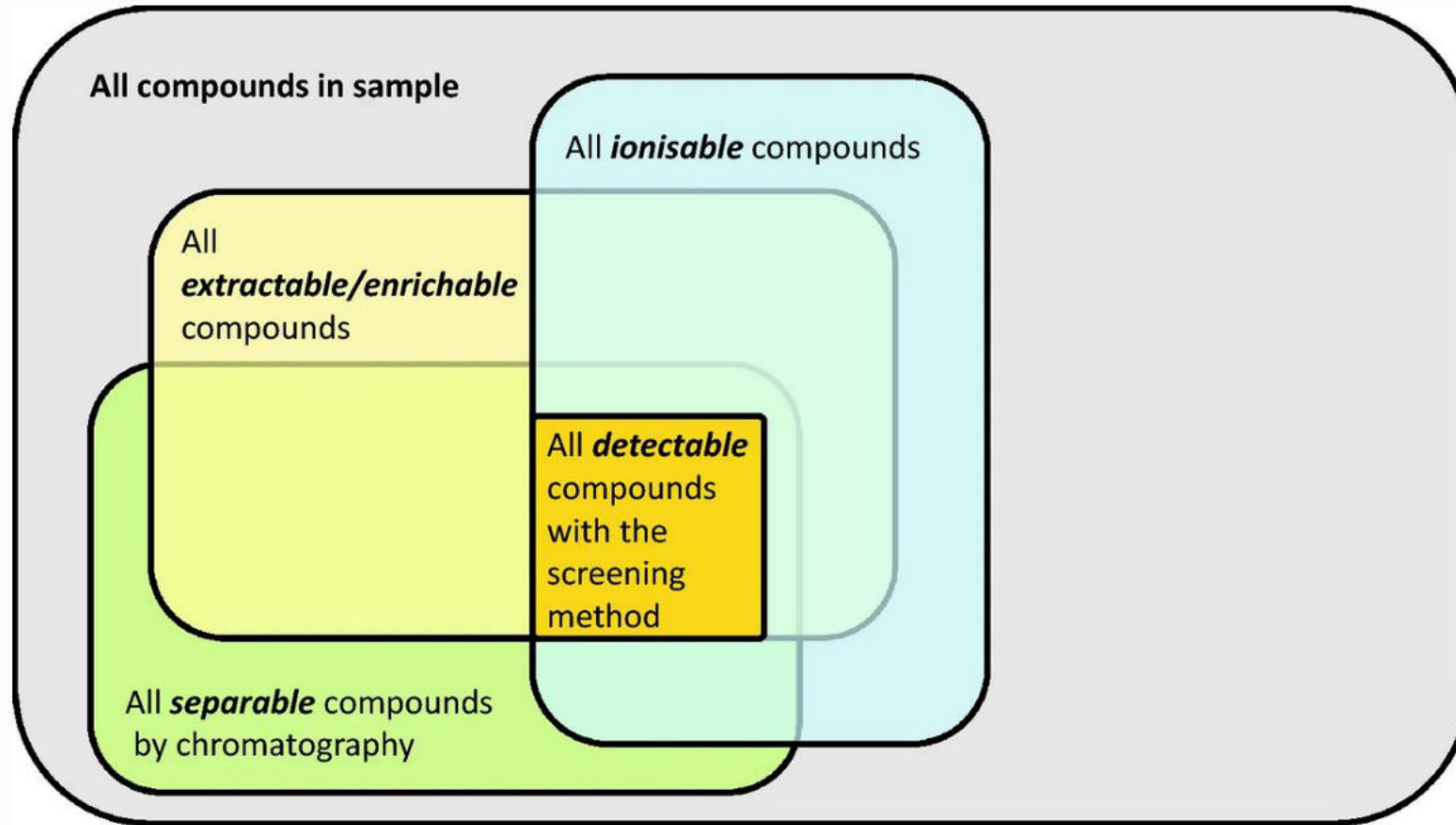


Non-Target HRMS (Schymanski et al. (2015)/ NORMAN network)

1. Target screening:
 - Referenzstandard
 - Quantifizierung möglich
2. Suspect screening:
 - Hinweise zu möglichen Substanzkandidaten
 - Kein Referenzstandard
 - Nur Aussage über detects möglich
3. Non-Target screening (NTS):
 - Keine Hinweise zu möglichen Substanzkandidaten
 - Kein Referenzstandard

NTS im Gewässermonitoring – Eine Methode für alle Stoffe?

From: [NORMAN guidance on suspect and non-target screening in environmental monitoring](#)



[Hollender et al. 2023](#)

Matrix

- Schwebstoffe – adsorbierende Substanzen
- Wasser – polare Substanzen

Flüssigchromatographie

- LC-HRMS (QToF)
- nicht volatile, polare-semi polare Substanzen

Analytik

- Optimiert für Arzneimittel ([Nürnberg et al 2015](#))
- $\log K_{ow}$: -4.16 bis 7.75

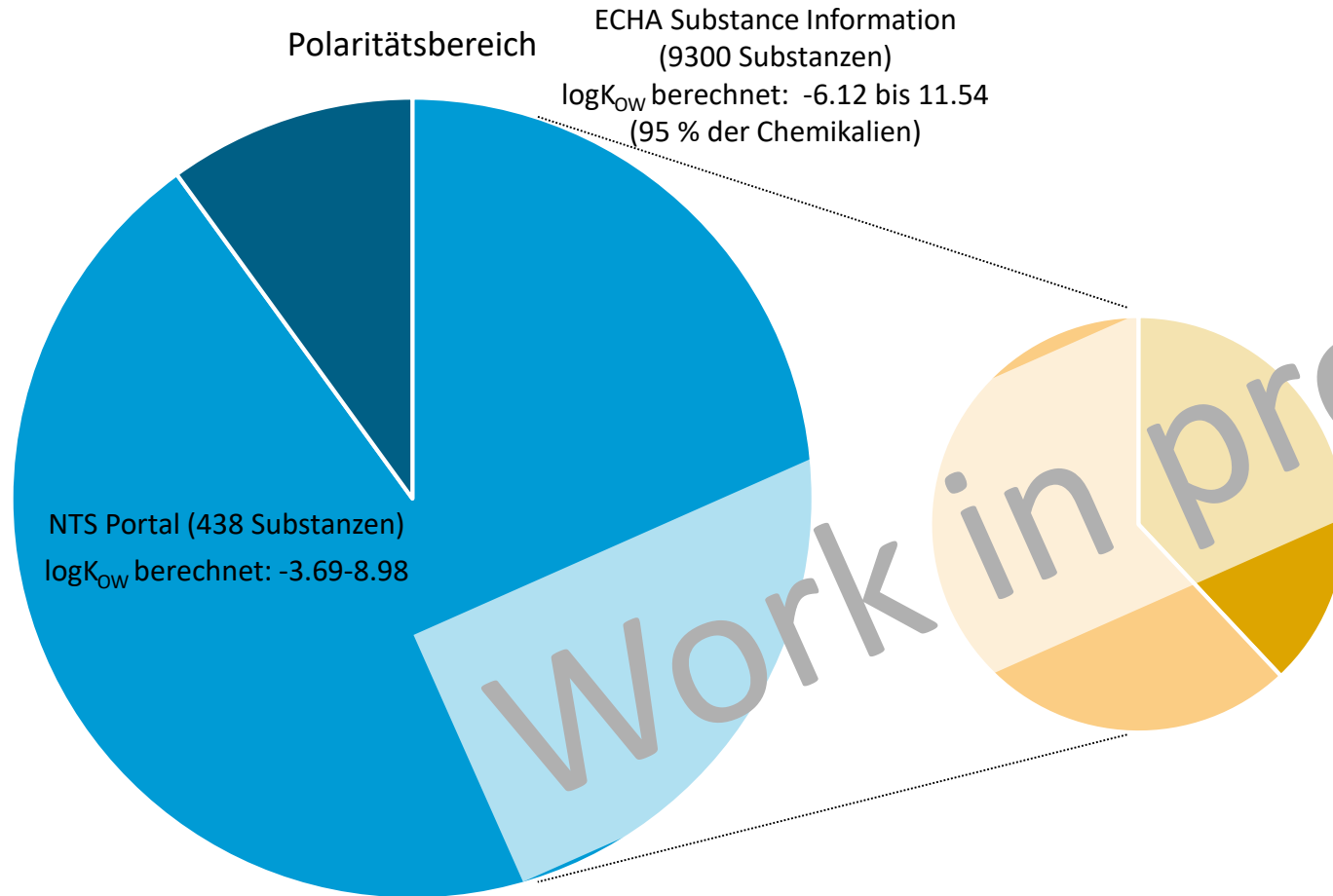
Datenevaluierung

- [Jewell et al. 2019](#)

Screening “Nachweisgrenze”

- Unterer Nanogramm - Bereich

NTS im Gewässermonitoring – Eine Methode für alle Stoffe?

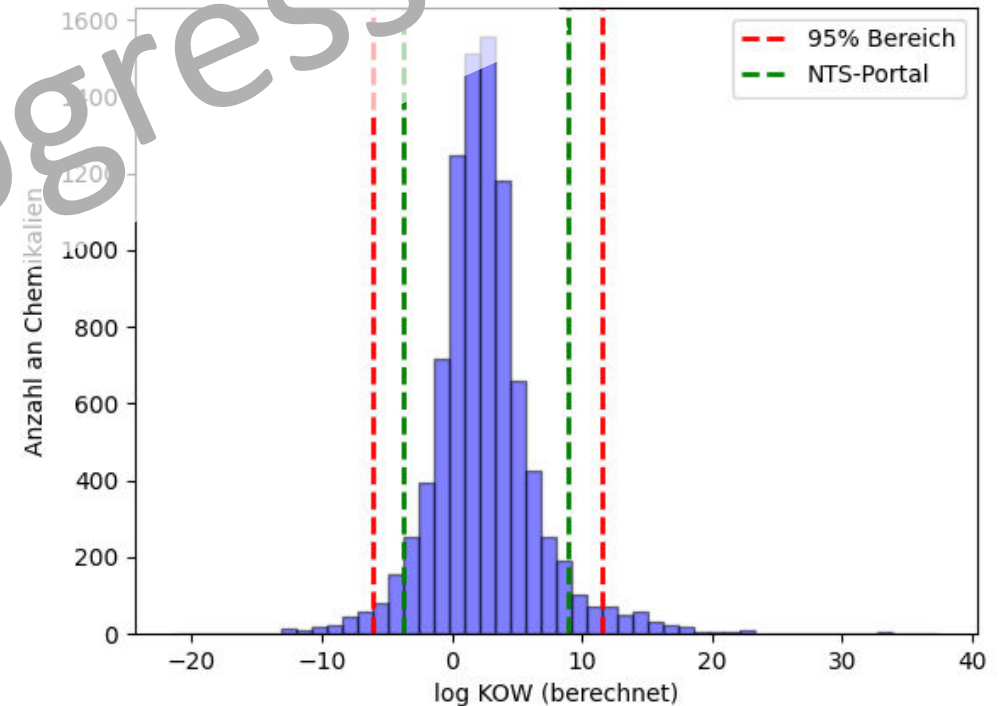


■ durch NTS Portal abgedeckt

■ nicht durch NTS Portal abgedeckt

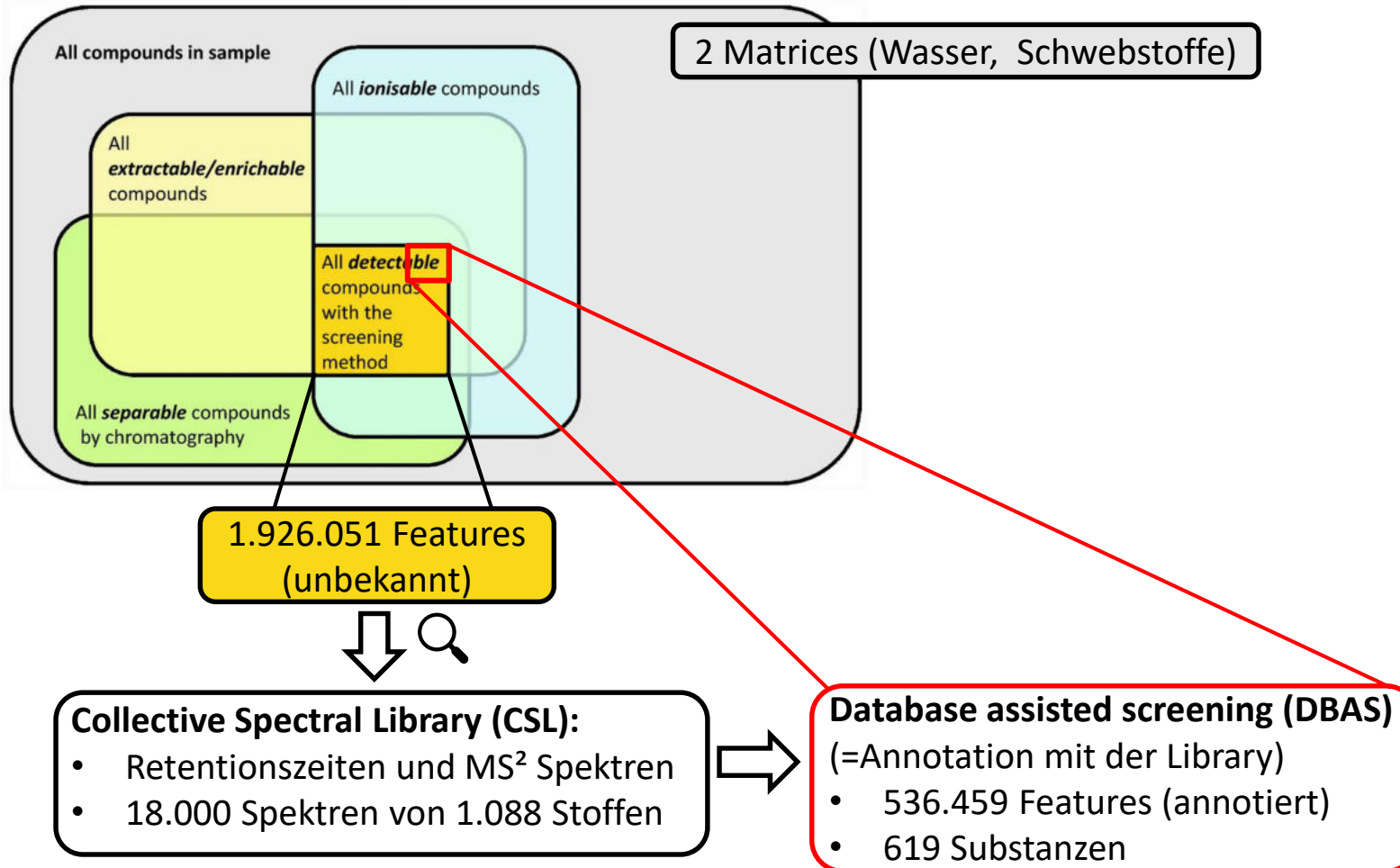
■ sedimentrelevant
 ($\log Kow > 3$)

■ wasserrelevant
 ($\log Kow < 3$)



Sicherheitslevel von NTS-Nachweisen im behördlichen Monitoring

<https://doi.org/10.1186/s12302-023-00779-4> – Hollender et al. 2023
 From: NORMAN guidance on suspect and non-target screening in environmental monitoring



Identifikationslevel für HRMS Daten (Alygizakis et al. 2023 /NORMAN)

Requirements	Identification Points (IP) earned
Precursor ion (Accuracy < 2 mDa / 5 ppm, R>15000)	mandatory
Retention time ± 0.2 min (only applicable in target)	0.40 ✓
Predicted Retention time index (only applicable in suspect where retention time match is not available, validated approach with provided uncertainty)	0.15
Isotopic fit (at least one isotope: abundance and accuracy of M+1, M+2,...)	0.20 ✓
Most intense experimental fragment ion	0.20 ✓
All other experimental fragment ions Number of experimental fragments normalized to the total number of fragments in the library	0.20 ✓
The "All other experimental fragment ions" score is penalized if the number of other experimental fragments present in the database is 2 or less	-0.10
<i>In silico</i> predicted fragment ions in case experimental fragments are not available Number of experimental fragments normalized to the total number of fragments in the library max number of fragments in library=10 most intense	0.20
Only DIA	-0.10

Identification Levels	IP Score
Level 1	>0.75-1.00
Level 2	>0.60-0.75
Level 3	0.50-0.60
Level 4	0.20-<0.50
Level 5	0.00-0.20

<https://doi.org/10.1016/j.trac.2023.116944>

Priorisierungskonzepte

Räumliche- Kategorisierung

- Nachweise an Standorten

- Bewertungsmaßstab:
- Detektionsrate



Ubiquität (Exposition)

Trend-Kategorisierung

- Lineare Trends
- Nicht-lineare Trends

- Bewertungsmaßstab:
- Signifikanz
 - Effektstärke



Trends (Exposition)

Hazard Priorisierung

- Registrierungs dossiers
- QSAR: JANUS/OECD

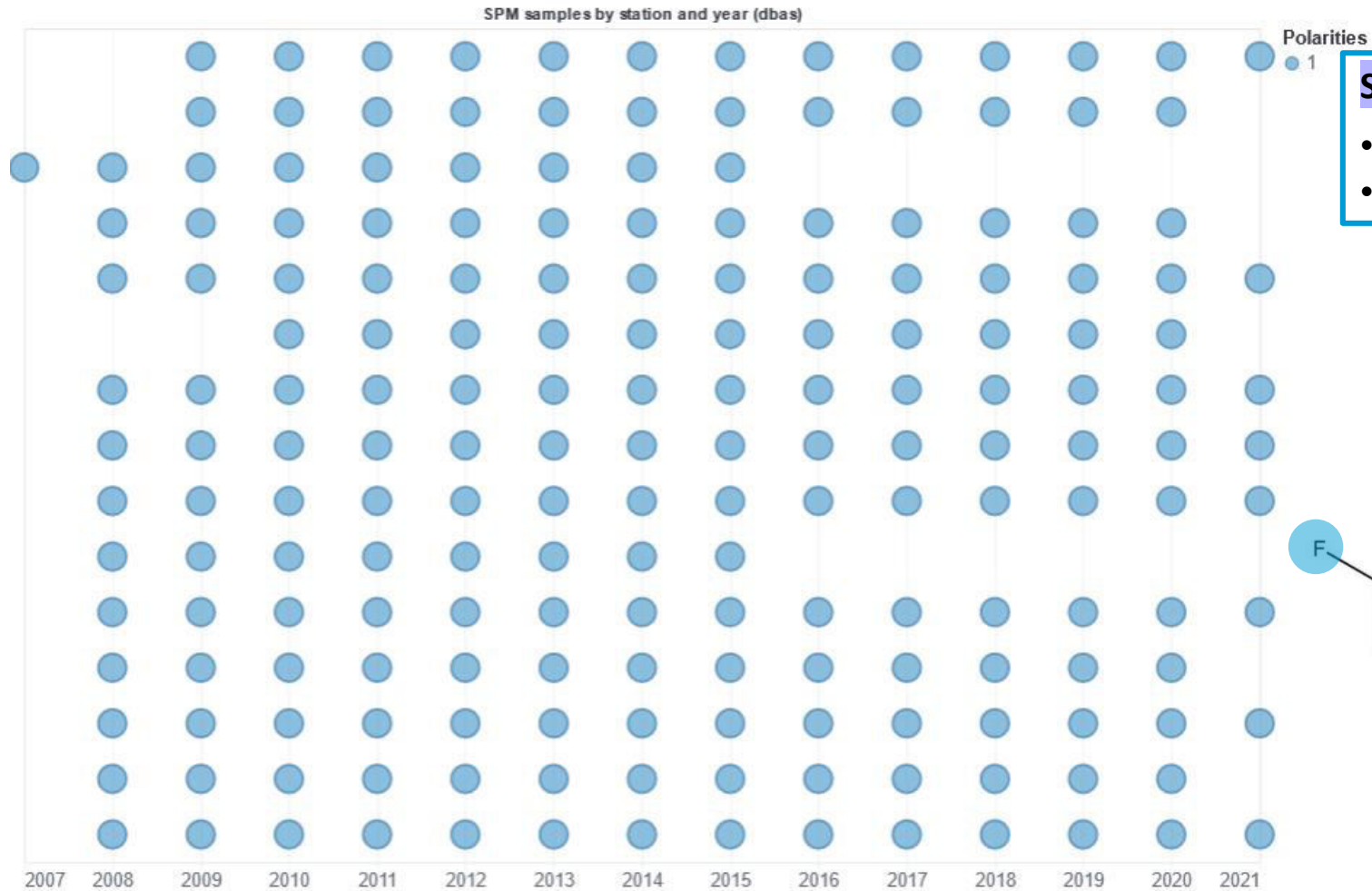
Bewertungsmaßstab:

- PBT/PMT
- CMR (Trinkwasser)



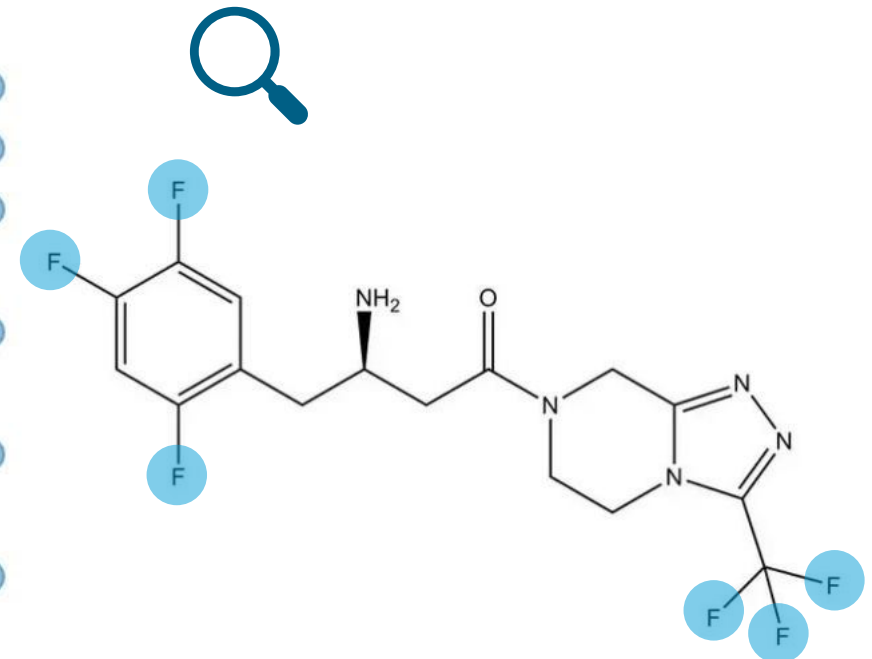
Toxizität

Identifizierung neuauftretender Problemstoffe und Mischungen

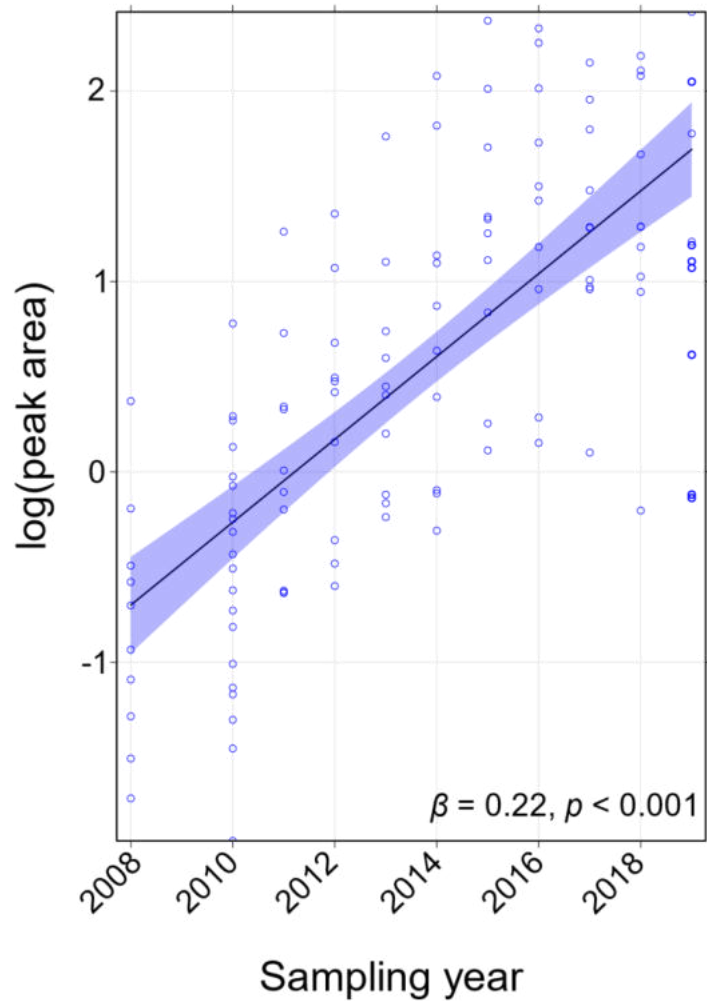
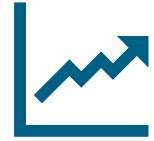


Sitagliptin:

- An 15/15 Messtationen detektiert
- Insgesamt 293 Detektionen



Identifizierung neuauftretender Problemstoffe und Mischungen - TRENDS



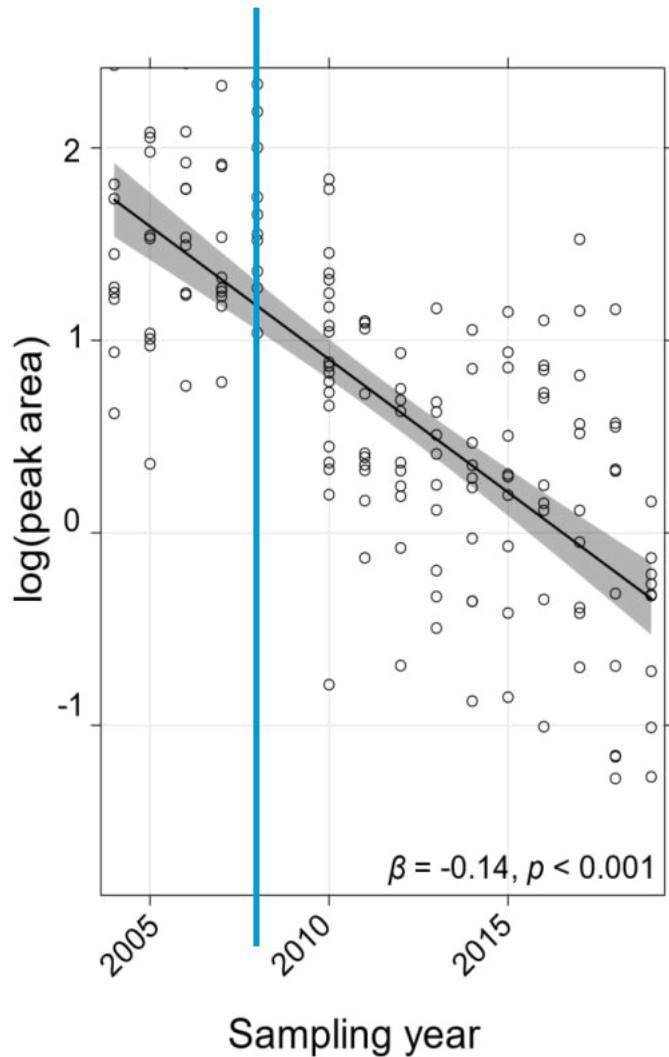
Sitagliptin:

- Typ 2 Diabetespräparat
- FDA-Zulassung in 2006, EU 2007
- Biotransformation: hauptsächlich unveränderte Exkretion via Urin
- Gefahrenbasierte Parameter: P, not B, not T (EMA, 2018)
- JANUS: vP, C

EMA assessment report für Janumet

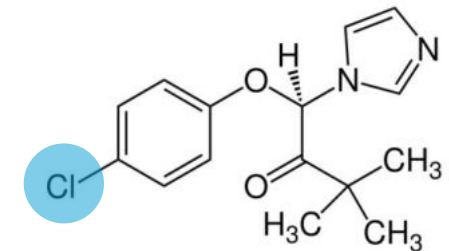
- *"The comparison of the predicted concentration in surface water with the predicted no effect concentration did not result in risk quotients above 1 (for algae, fish and water flea) or 0.1 (for microorganisms)."*
(EMEA/517697/2009, 29 July 2009)

Identifizierung neuauftretender Problemstoffe und Mischungen - TRENDS



Climbazole:

- Antimykotisch und fungistatisch wirksames Biozid
- Wurde zum Beispiel in Antischuppen-Shampoos verwendet
- Beschränkung in 2008
- Sehr giftig für Wasserorganismen
- JANUS: vP, C, R



Properties of concern



Under assessment as Endocrine Disrupting

6-PPD Chinon

Exposure to 6-PPD Quinone at Environmental Concentrations Causes Abnormal Locomotion and Neurodegeneration in *Caenorhabditis elegans*

Xin Hua, Xiao Feng, Geyu Jiang, Ji Chen, and Dayong Wang

ACS Publications

Abstract: 6-PPD quinone (6-PPDQ) can be transformed from 6-PPD...

INTRODUCTION
Rubber tire wear (RTW) is a major source of urban contamination that has received increased attention in recent years. The increasing production of RTW-generated particles results in severe contamination, such as metals, persistent organic pollutants (POPs), and phytochemicals (PPNs), causing serious environmental pollution after entering the environment.^{1–4} Among the PPNs, N-(1,3-dimethylbutyl)N'-phenyl-p-phenylenediamine (6-PPD) is most widely considered as an antioxidant in the rubber tire. Usually, 6-PPD gradually oxidizes to 6-PPD quinone (6-PPDQ) through the changes in the aliphatic structures. Recently, 6-PPDQ was detected to be detected in road wear particle leachates, indicating the release of 6-PPDQ.^{5–10} In addition, 6-PPDQ was reported to be detected in various environmental media, such as water, sediment, indoor particles, and dust, ranging from ng to μg.^{11–15}

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Research Article

A ubiquitous tire rubber-derived chemical induces acute mortality in coho salmon

Zhenye Tian,¹ Hong Zhao,¹ Katherine E. Peter,¹ Melissa Gonzalez,¹ Jill Meyer,¹ Christopher Sloan III,¹ Sumner Pratt,¹ Emma Blomquist,¹ Rachel Hartigan,¹ Alan E. Cortner,¹ Matthew Clark-Rose,¹ Diana Walker-Carlson,¹ David Hargrett,¹ Amy Jones,¹ James Fan-Havens,¹ Susan Hargrett,¹ Richard Lusk,¹ Alissa Silvestro,¹ Barbara Foster,¹ Matthew L. Schmidt,¹ Jay M. Davis,¹ Michael C. Dodd,¹ Amber Thompson,¹ Jennifer K. Maltby,¹ Edward F. Rabinovich,¹ and Robert W. Howcroft,¹ *Environ. Toxicol. Chem.* 2022, 41, 4642–4652

ABSTRACT: 6-PPD quinone (6-PPDQ) can be transformed from 6-PPD in the environment. Nevertheless, the potential neurotoxicity of 6-PPDQ after long exposure and the underlying mechanism are largely unknown. In *Caenorhabditis elegans*, we have observed that 0.1–10 μg/L of 6-PPDQ caused several-fold increased locomotion behavior, bioleaching, the neurodegeneration of the motor neurons was observed in 10 μg/L of 6-PPDQ-exposed nematodes, observed neurodegeneration was associated with the activation of the C. elegans DRE-1-mediated signaling cascade. In this signaling cascade, expression of *egl-49*, *egl-52*, *egl-53*, *egl-54*, and *egl-55* were increased by 10 μg/L of 6-PPDQ. Moreover, average gene expression values required for the control of *egl-49*, expression of *egl-1* and *egl-2* were decreased by 0.1–10 μg/L of 6-PPDQ and expression of *egl-7* and *egl-10* were increased by 10 μg/L of 6-PPDQ.

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Research Article

Toxicological effects of 6PPD and 6PPD quinone in rainbow trout

Shubham Verheyne, Adam G. Cox, Prabhendra Sivasubramanian, A. Clark

ABSTRACT: 6-PPD quinone (6-PPDQ) can be transformed from 6-PPD in the environment. Nevertheless, the potential neurotoxicity of 6-PPDQ after long exposure and the underlying mechanism are largely unknown. In *Caenorhabditis elegans*, we have observed that 0.1–10 μg/L of 6-PPDQ caused several-fold increased locomotion behavior, bioleaching, the neurodegeneration of the motor neurons was observed in 10 μg/L of 6-PPDQ-exposed nematodes, observed neurodegeneration was associated with the activation of the C. elegans DRE-1-mediated signaling cascade. In this signaling cascade, expression of *egl-49*, *egl-52*, *egl-53*, *egl-54*, and *egl-55* were increased by 10 μg/L of 6-PPDQ. Moreover, average gene expression values required for the control of *egl-49*, expression of *egl-1* and *egl-2* were decreased by 0.1–10 μg/L of 6-PPDQ and expression of *egl-7* and *egl-10* were increased by 10 μg/L of 6-PPDQ.

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Research Article

Acute Toxicity of the Tire Rubber-Derived Acute Toxicity of the Tire Rubber-Derived Acute Toxicity of the Tire Rubber-Derived

Markus Brinkmann, David Montgomery, Summer Schlinger, J. A. Clark

ABSTRACT: 6-PPD quinone (6-PPDQ) can be transformed from 6-PPD in the environment. Nevertheless, the potential neurotoxicity of 6-PPDQ after long exposure and the underlying mechanism are largely unknown. In *Caenorhabditis elegans*, we have observed that 0.1–10 μg/L of 6-PPDQ caused several-fold increased locomotion behavior, bioleaching, the neurodegeneration of the motor neurons was observed in 10 μg/L of 6-PPDQ-exposed nematodes, observed neurodegeneration was associated with the activation of the C. elegans DRE-1-mediated signaling cascade. In this signaling cascade, expression of *egl-49*, *egl-52*, *egl-53*, *egl-54*, and *egl-55* were increased by 10 μg/L of 6-PPDQ. Moreover, average gene expression values required for the control of *egl-49*, expression of *egl-1* and *egl-2* were decreased by 0.1–10 μg/L of 6-PPDQ and expression of *egl-7* and *egl-10* were increased by 10 μg/L of 6-PPDQ.

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Research Article

Transformation Products of Tire Rubber Antioxidant 6PPD in Heterogeneous Gas-Phase Ozonation: Identification and Environmental Occurrence

Haoqi Nina Zhao, Ximin Hu, Zhenyu Tian, Melissa Gonzalez, Craig A. Rideout, Katherine T. Peter, Michael C. Dodd,¹ and Edward F. Rabinovich,¹ *Environ. Toxicol. Chem.* 2022, 41, 4653–4662

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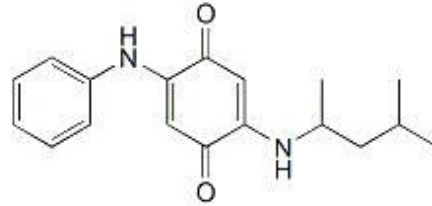
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Und jetzt konkret?

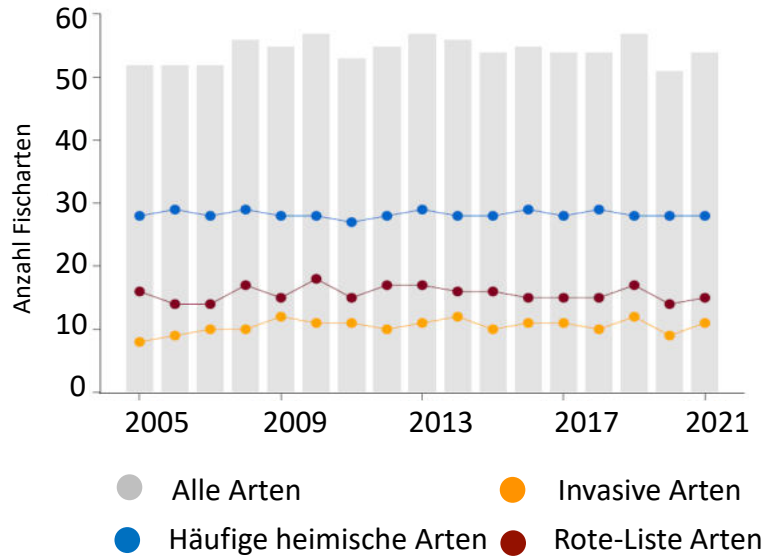
Verändert 6-PPD Chinon aus dem Reifenabrieb die Fischdiversität?



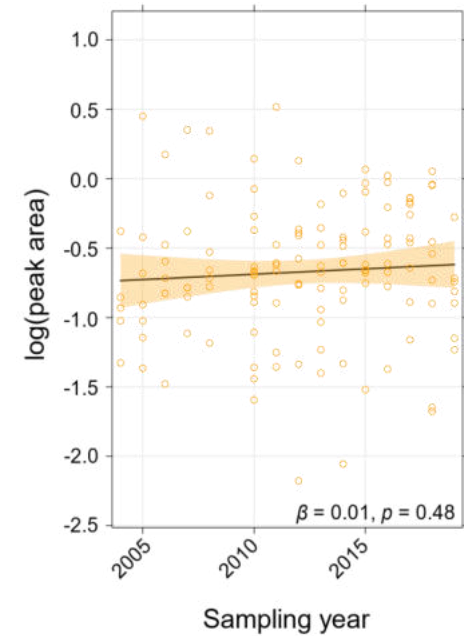
Hazard classification & labelling



Artenzahl Fische



Messintensität 6-PPD Chinon



Take home messages

- Harmonisierung und Standardisierung wird benötigt
- Verfügbarkeit behördliche Daten notwendig
- (Retrospektive) Trendanalyse liefert:
 - Hinweise auf das Verhalten von Stoffen (und Mischungen) in der Vergangenheit
 - Erlaubt die Modellierung von Verläufen
 - Effektivitätskontrolle von RMM
- Kooperationen essentiell für übergreifende räumlich-/zeitliche Bewertungen inkl. Frühwarnsystemen



**Non-Target Screening im
Gewässerschutz**
Vom Messen zum Bewerten



Vielen Dank für Ihre Aufmerksamkeit!

Anna Lena Kronsbein

AnnaLena.Kronsbein@uba.de

NTS Workshop in Berlin

14.-15.12.2023