detection and identification of toxic and high-risk chemicals with LC/HMRS and machine learning

anneli kruve

Kruvelab.com

increased threat to Swedish drinking water

an offence more than every fifth day

anneli kruve

DAGENS NYHETER. Nyheter Sverige Världen Ekonomi Kultur Sport Klimatet Ledare DN Debatt Me

SVERIGE

Ökat hot mot svenskt dricksvatten – brott mer än var femte dag

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water analysis

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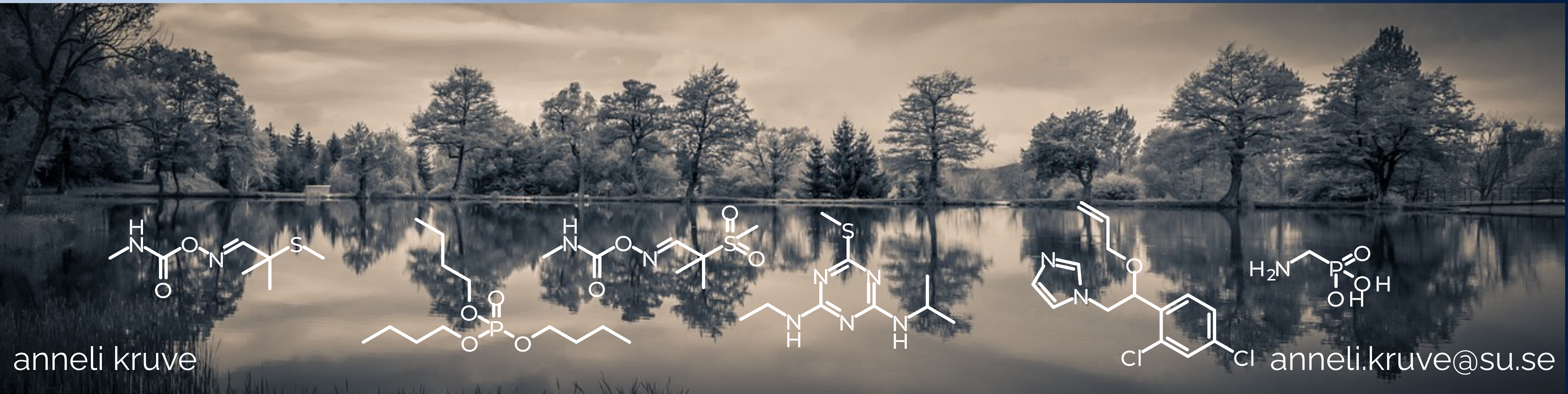
water analysis



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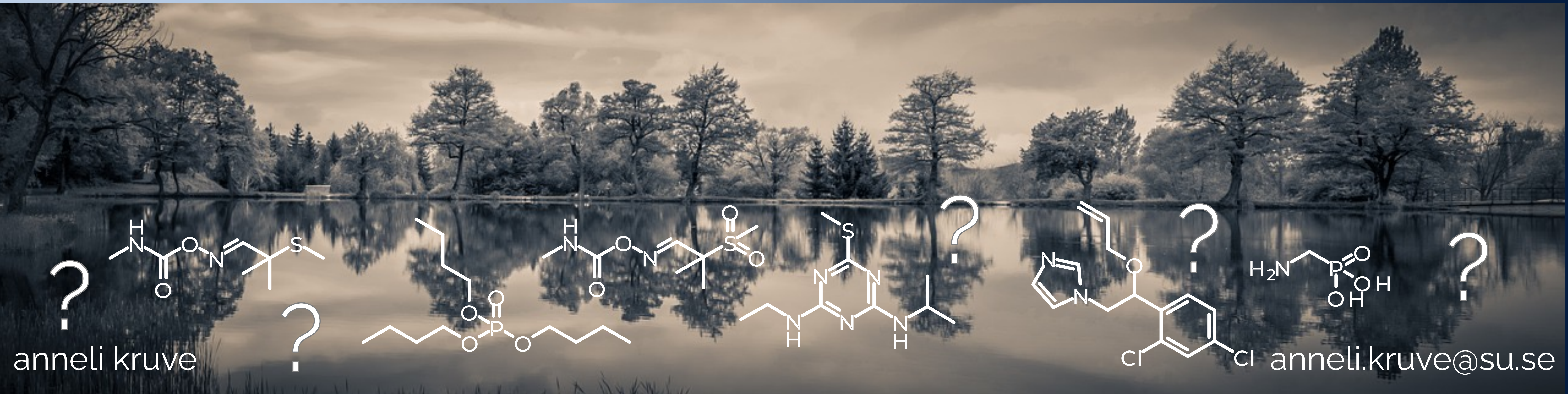
water analysis

thousands of chemicals
detected
in the environment



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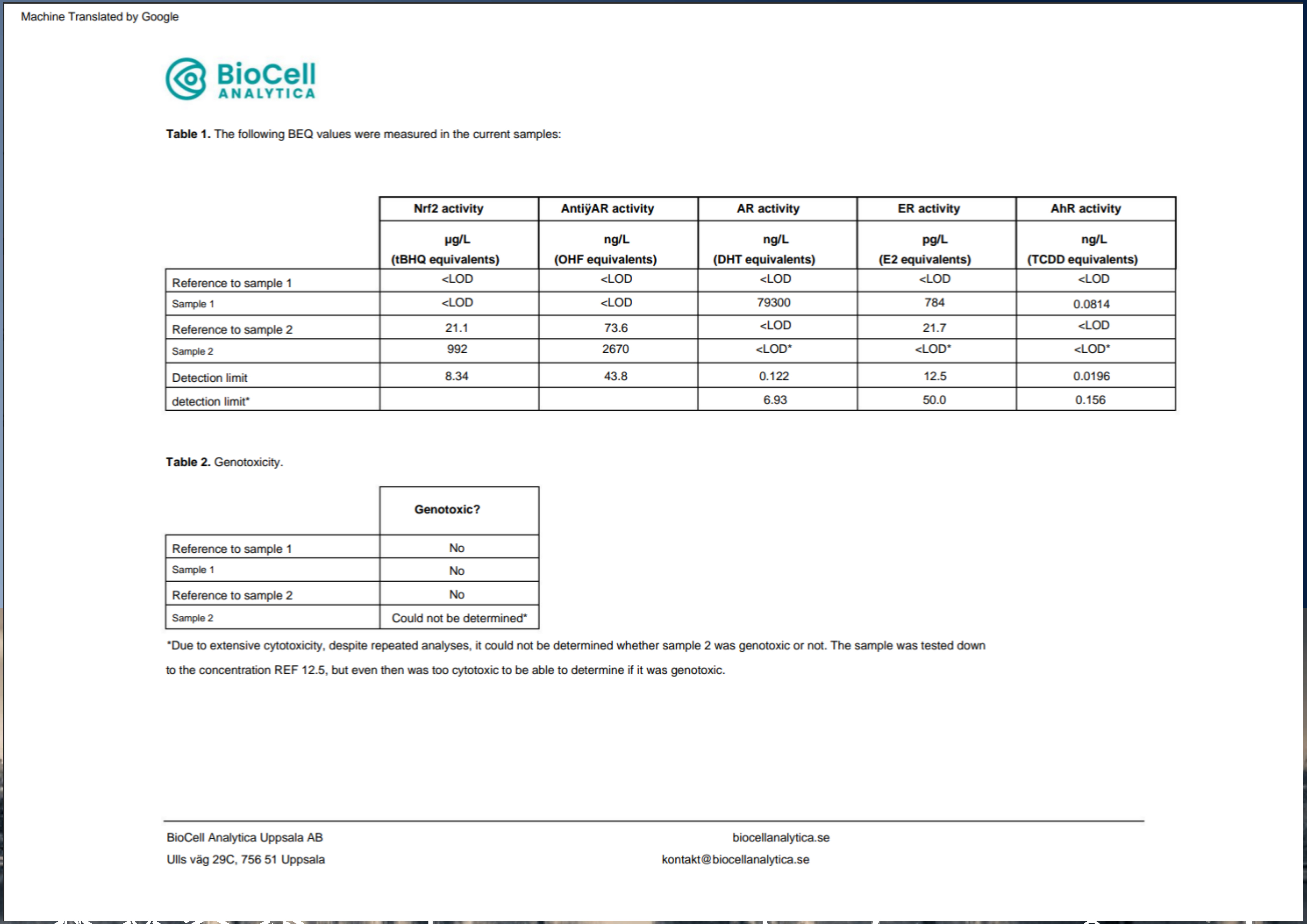

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BioCell ANALYTICA

Table 1. The following BEQ values were measured in the current samples:

	Nrf2 activity	AntiÿAR activity	AR activity	ER activity	AhR activity
	μg/L (tBHQ equivalents)	ng/L (OHF equivalents)	ng/L (DHT equivalents)	pg/L (E2 equivalents)	ng/L (TCDD equivalents)
Reference to sample 1	<LOD	<LOD	<LOD	<LOD	<LOD
Sample 1	<LOD	<LOD	79300	784	0.0814
Reference to sample 2	21.1	73.6	<LOD	21.7	<LOD
Sample 2	992	2670	<LOD*	<LOD*	<LOD*
Detection limit	8.34	43.8	0.122	12.5	0.0196
detection limit*			6.93	50.0	0.156

Table 2. Genotoxicity.

	Genotoxic?
Reference to sample 1	No
Sample 1	No
Reference to sample 2	No
Sample 2	Could not be determined*

*Due to extensive cytotoxicity, despite repeated analyses, it could not be determined whether sample 2 was genotoxic or not. The sample was tested down to the concentration REF 12.5, but even then was too cytotoxic to be able to determine if it was genotoxic.

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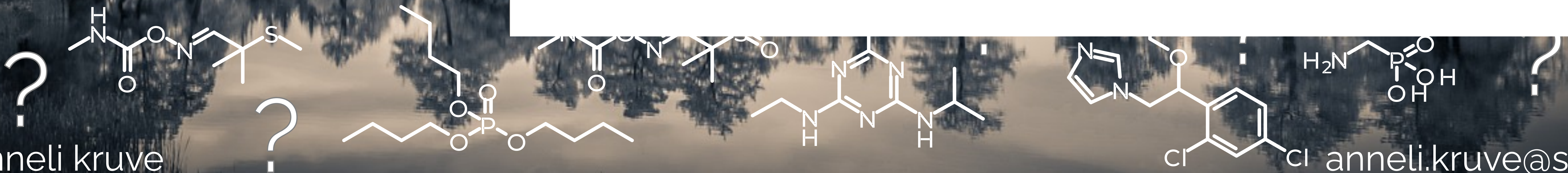

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how to …

PRIORITIZE

risk

IDENTIFY

structure

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how to …

PRIORITIZE

risk

IDENTIFY

structure

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prioritization

Szabo et al. Anal Chem 2024

sample → online prioritization → HRMS → offline prioritization → chemical risk

online prioritization:
- top N
- inclusion list
- isotopic abundance
- adduct formation
- MS^2 analysis

offline prioritization:
- recursive inc/exc
- relative abundance
- differential analysis
- metabolic logic
- modelling

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prioritization of chemicals

TOXICITY

ecotoxicity and endocrine disruptors

CONCENTRATION

exposure to potentially toxic chemicals

RISK

$$\mathrm{PriorityScore} = \frac{c_{\mathrm{predicted}}}{\mathrm{AC}_{50}^{\mathrm{5th\ percentile}}}$$

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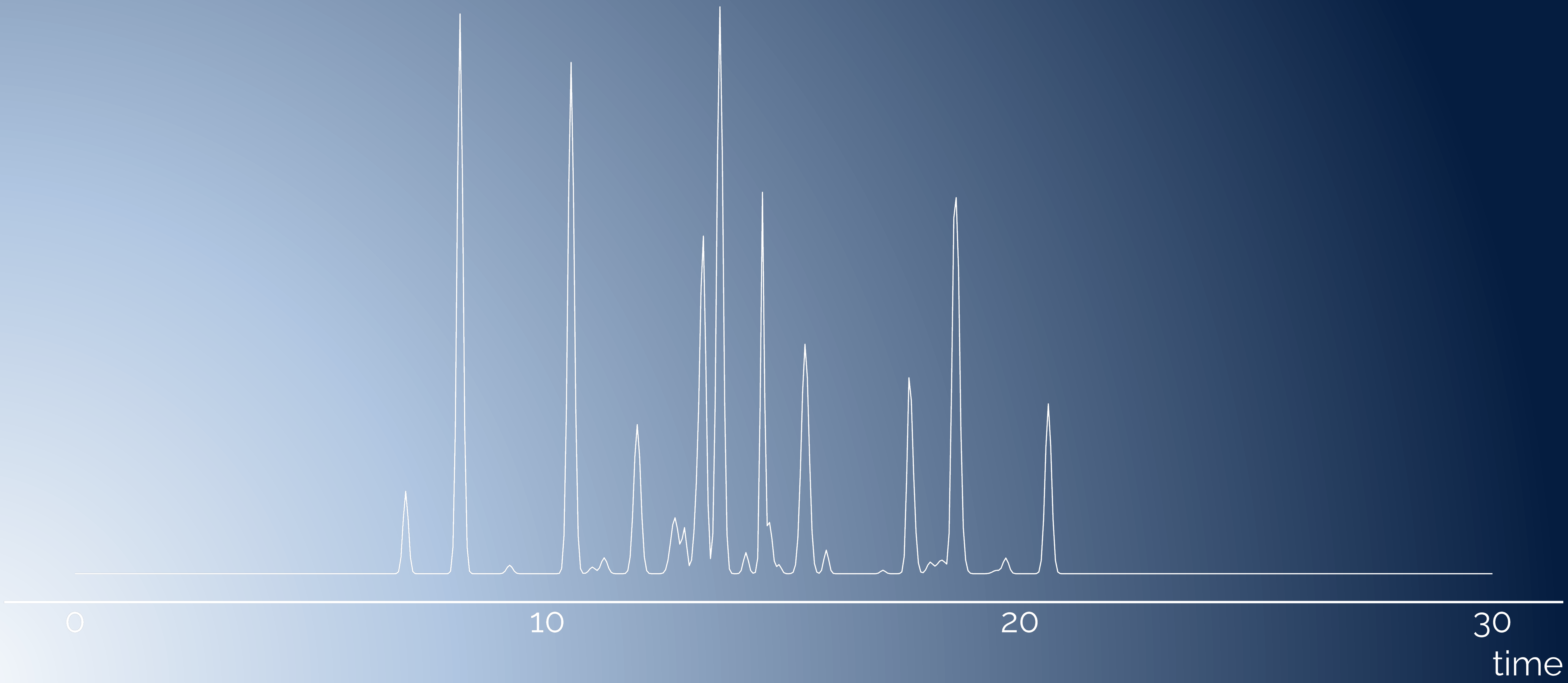
toxicity

of detected chemicals

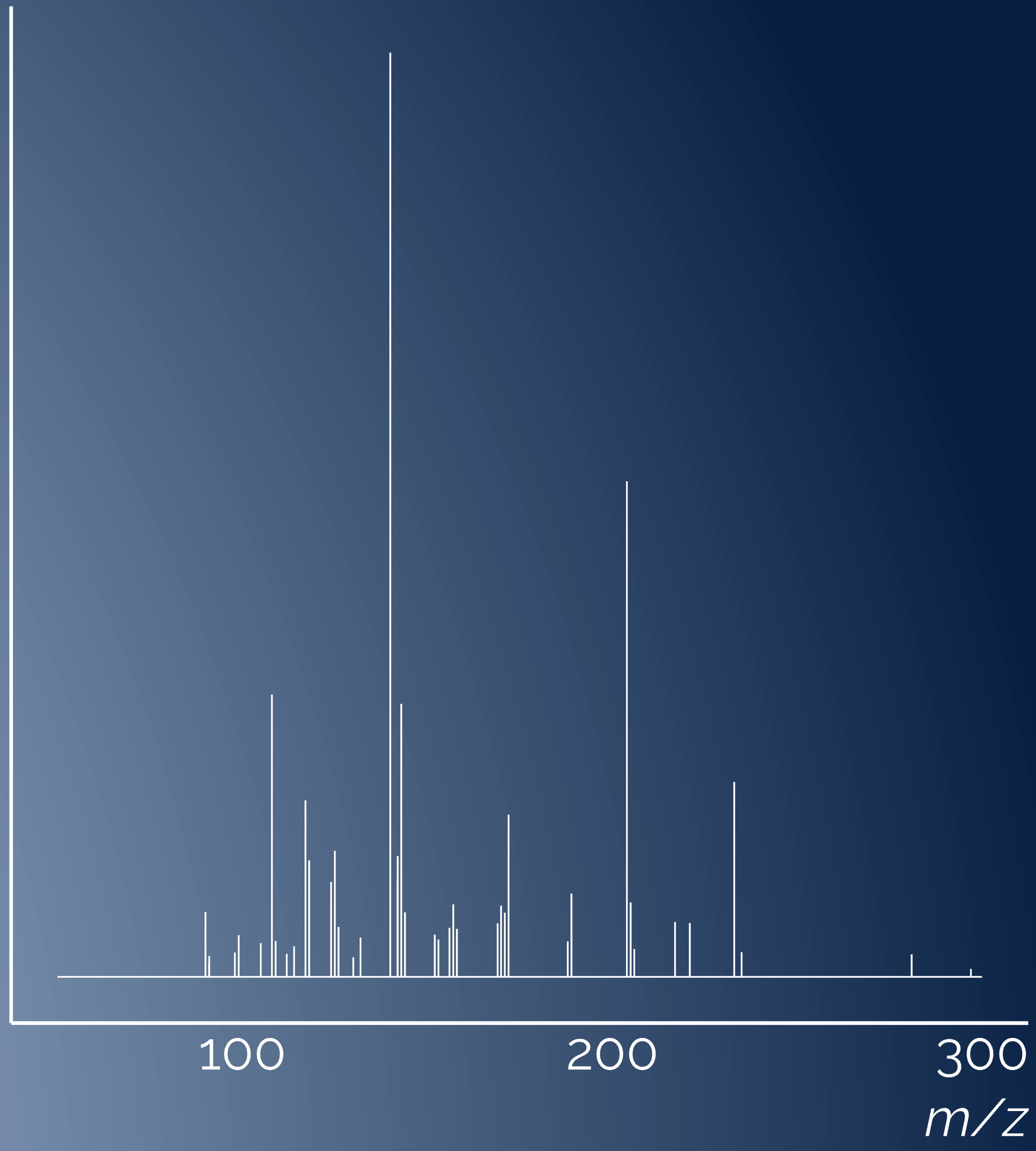
nontarget screening

with LC/HRMS



toxicity assessment

from spectra
to structure
to toxicity



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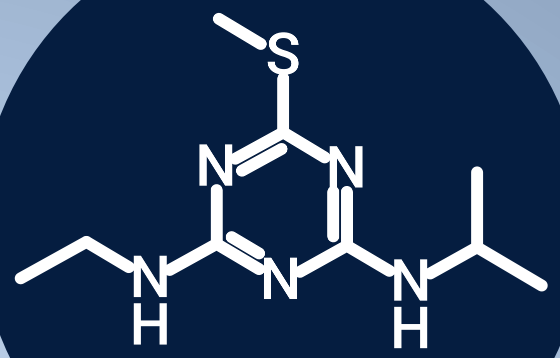
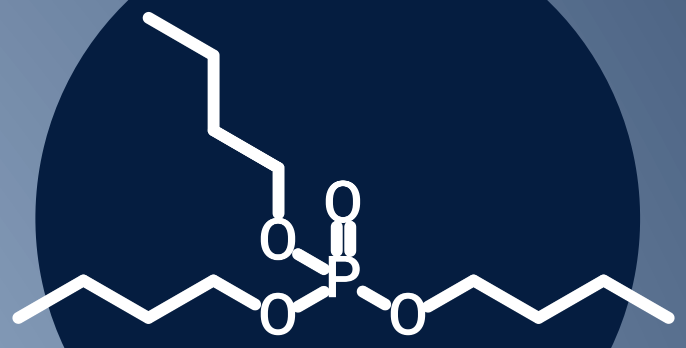
toxicity assessment



LC_{50} = 9.3 mg/L

known structure
known toxicity



LC_{50} = ? mg/L

known structure
unknown toxicity

?

LC_{50} = ? mg/L

unknown structure
unknown toxicity

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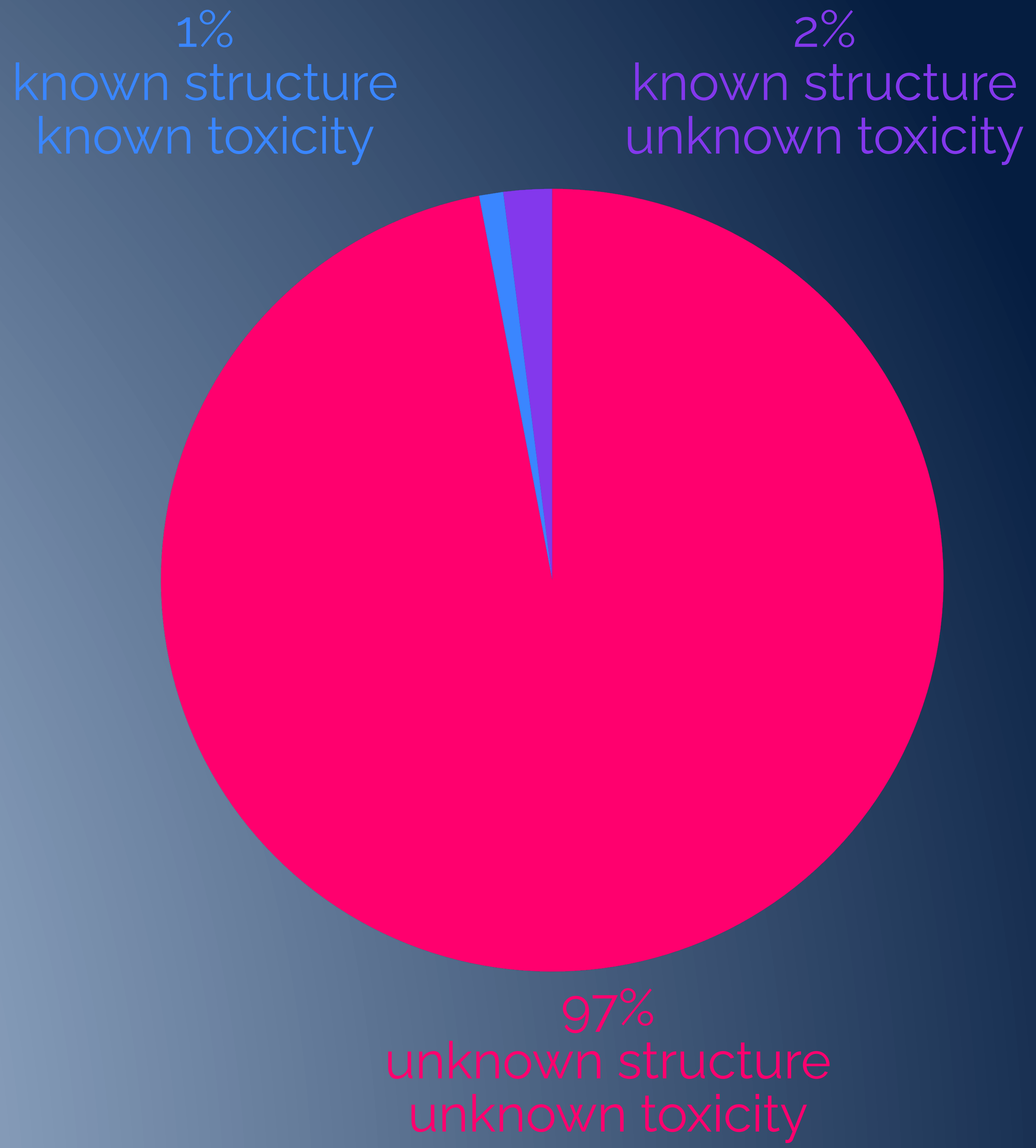
toxicity assessment

vast majority of detected chemicals remain unknown



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predicting toxicity

of detected chemicals

endpoints

ECOTOXICITY

fathead minnow, bluegill, and rainbow trout

ENDOCRINE DISRUPTION

AhR, AR, ER, MMP, P53, …

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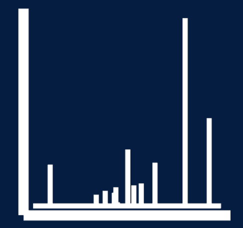
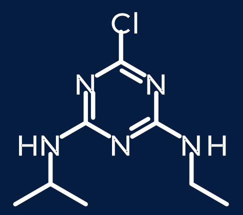
workflow



MS^2 spectra



structure as SMILES

molecular descriptors

toxicity prediction

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workflow

MS² spectra

molecular descriptors

toxicity prediction

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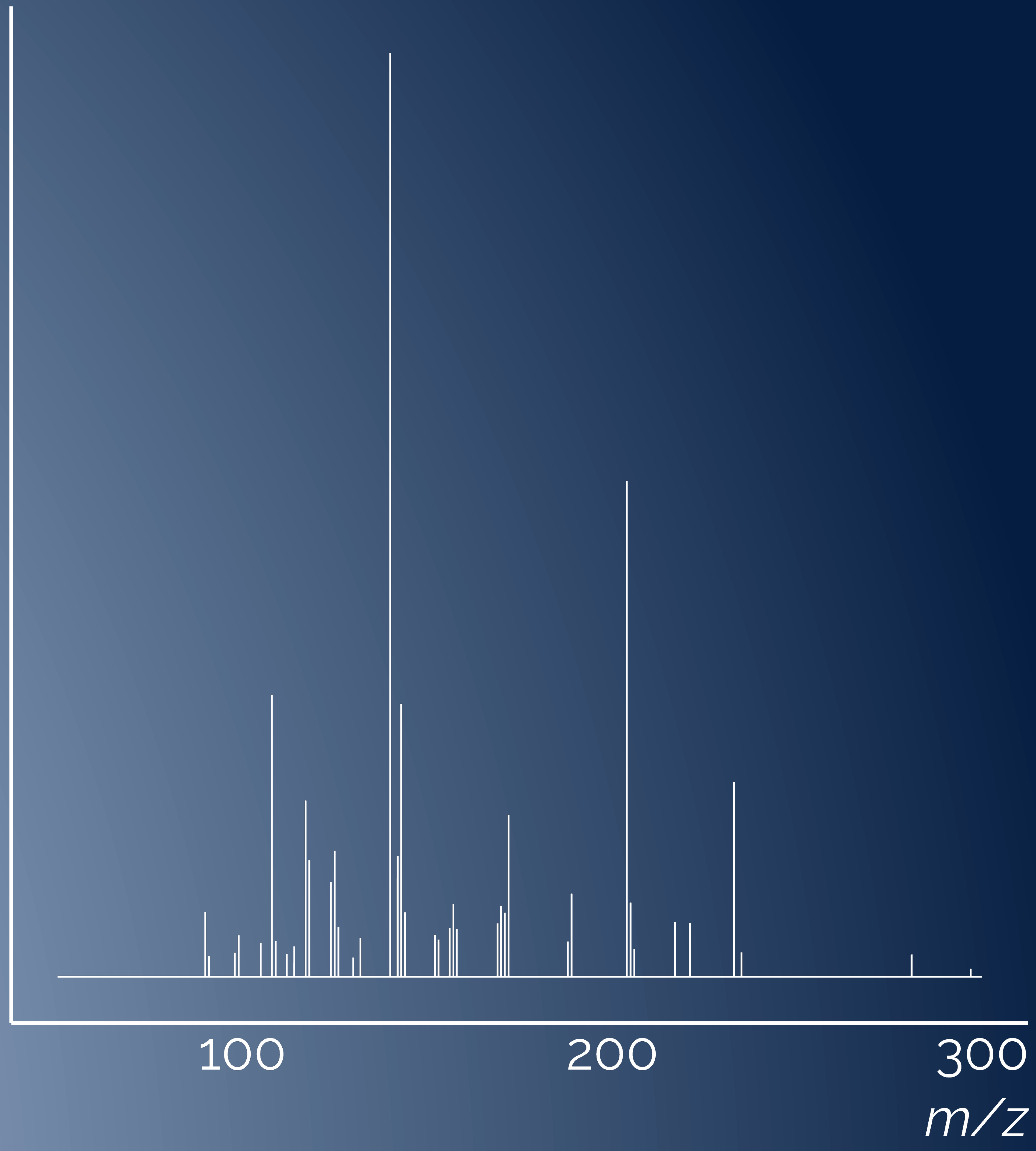
information available

in mass spectra



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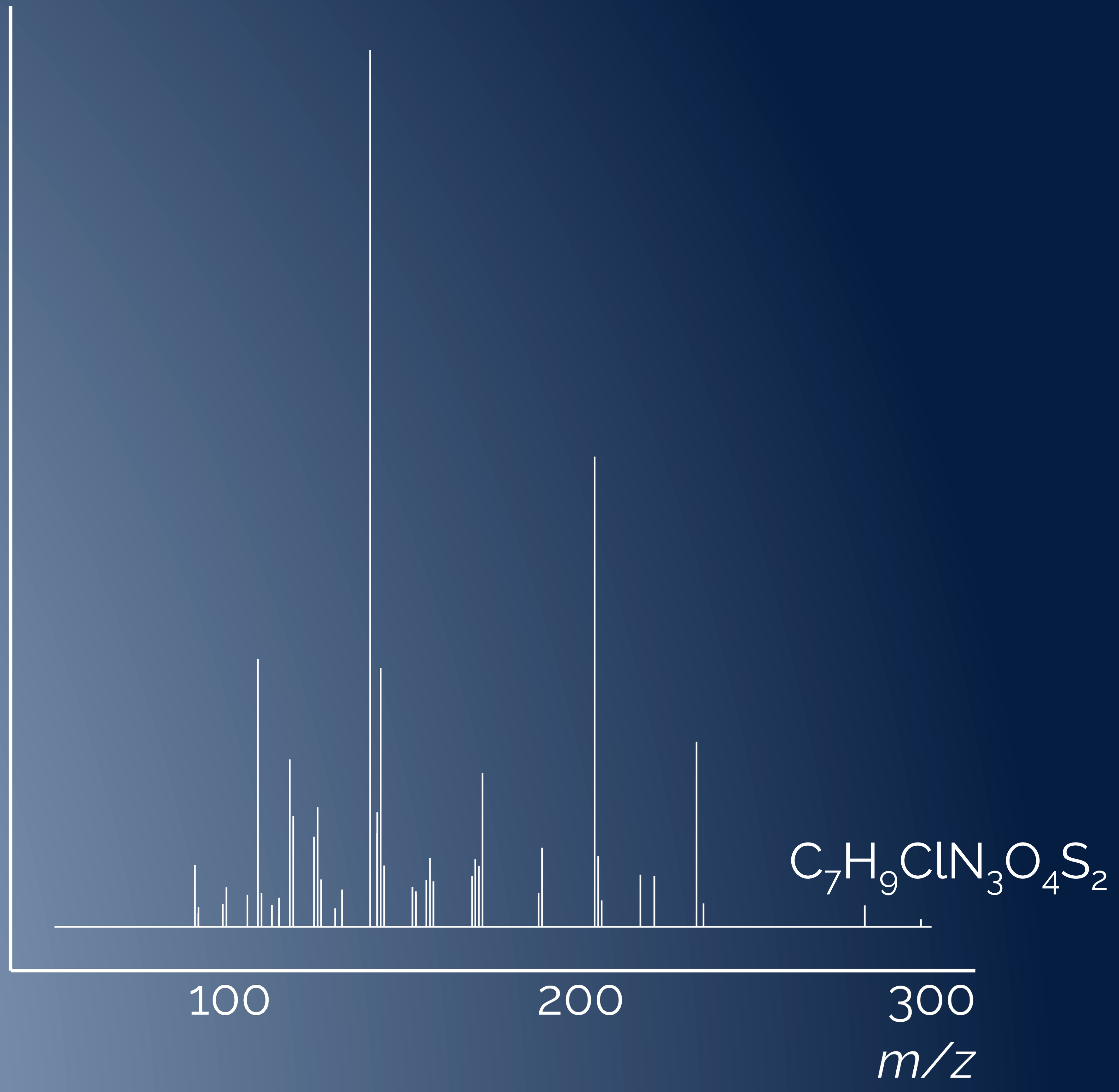
information available

in mass spectra



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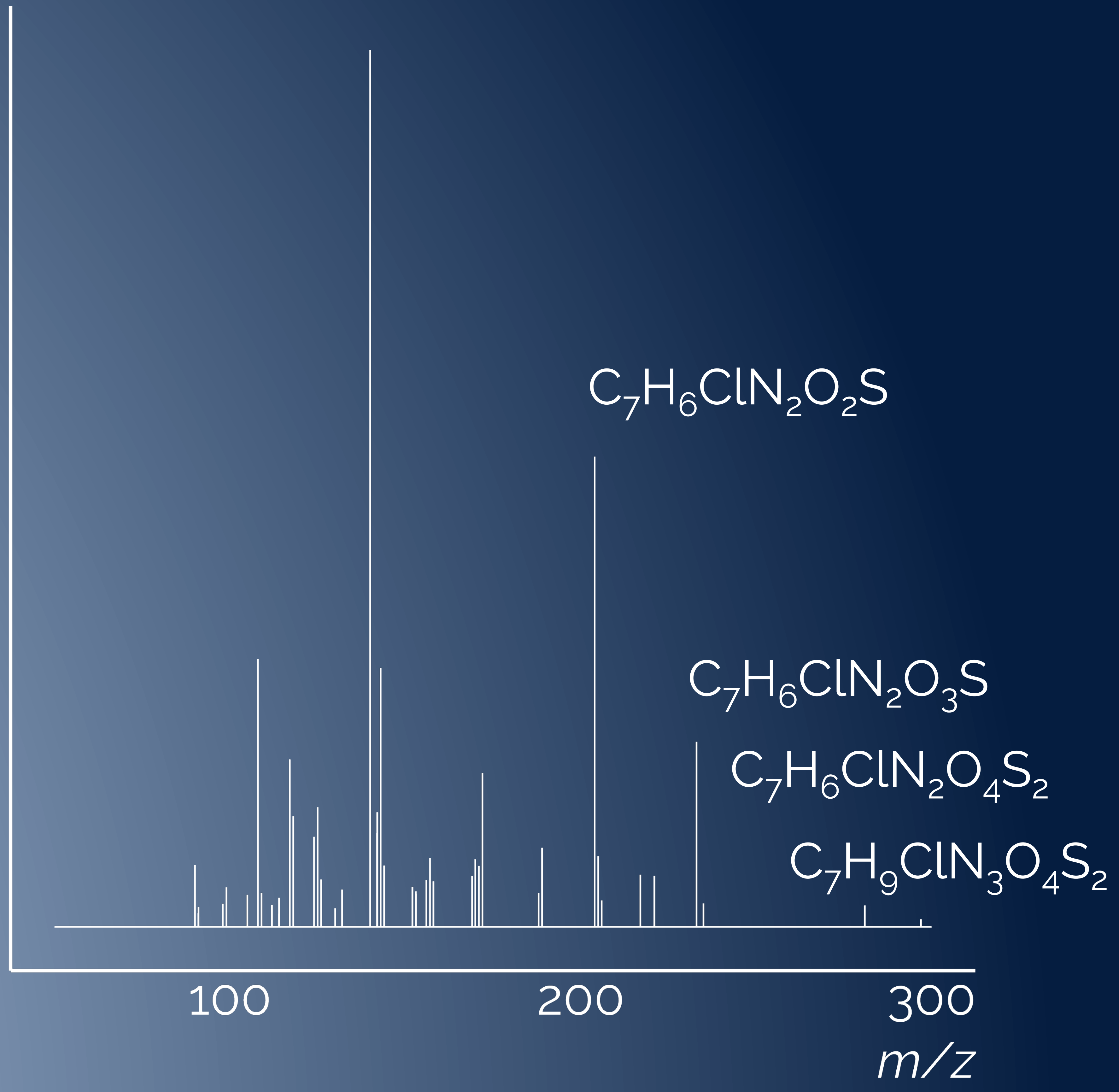

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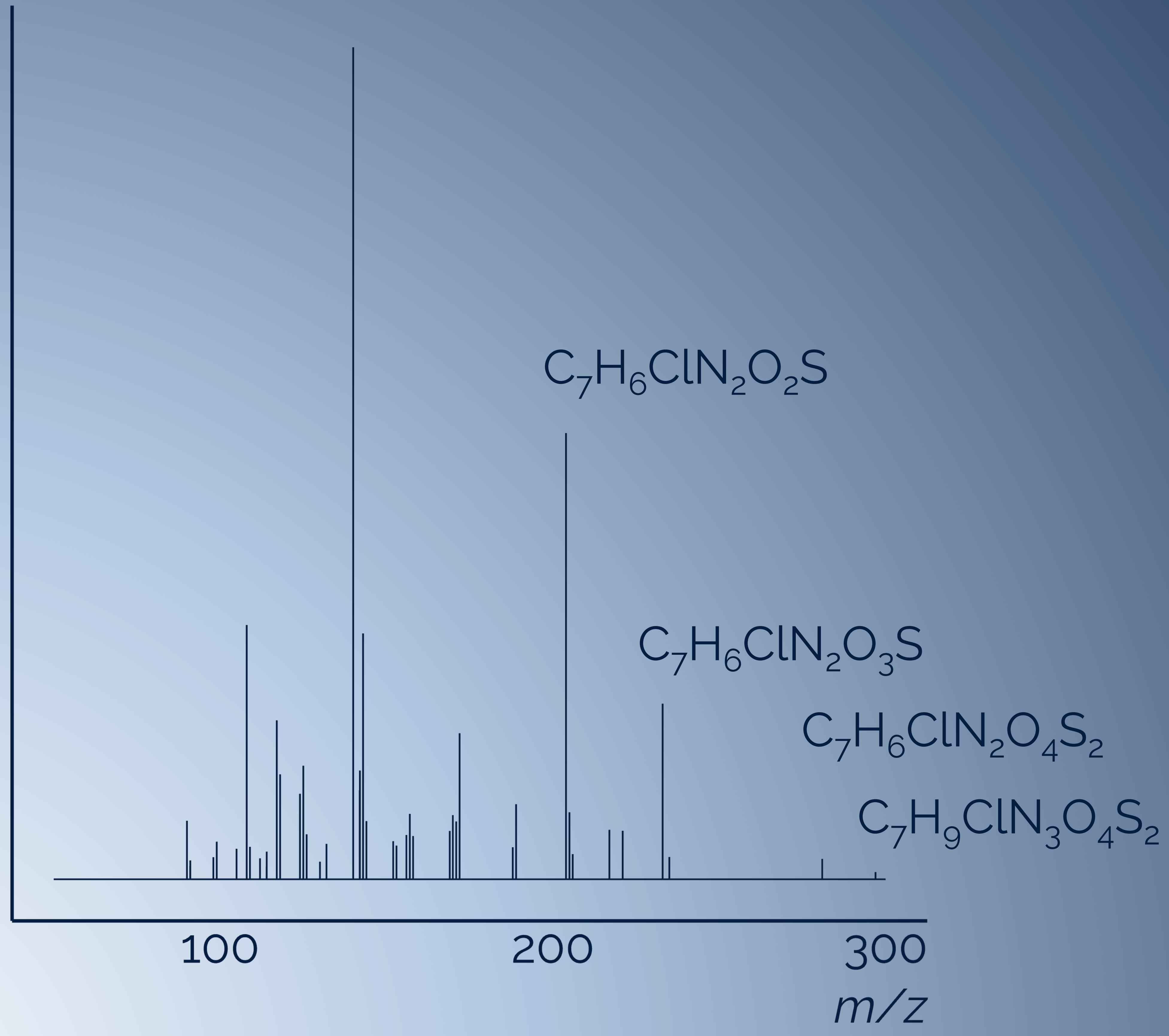
in mass spectra



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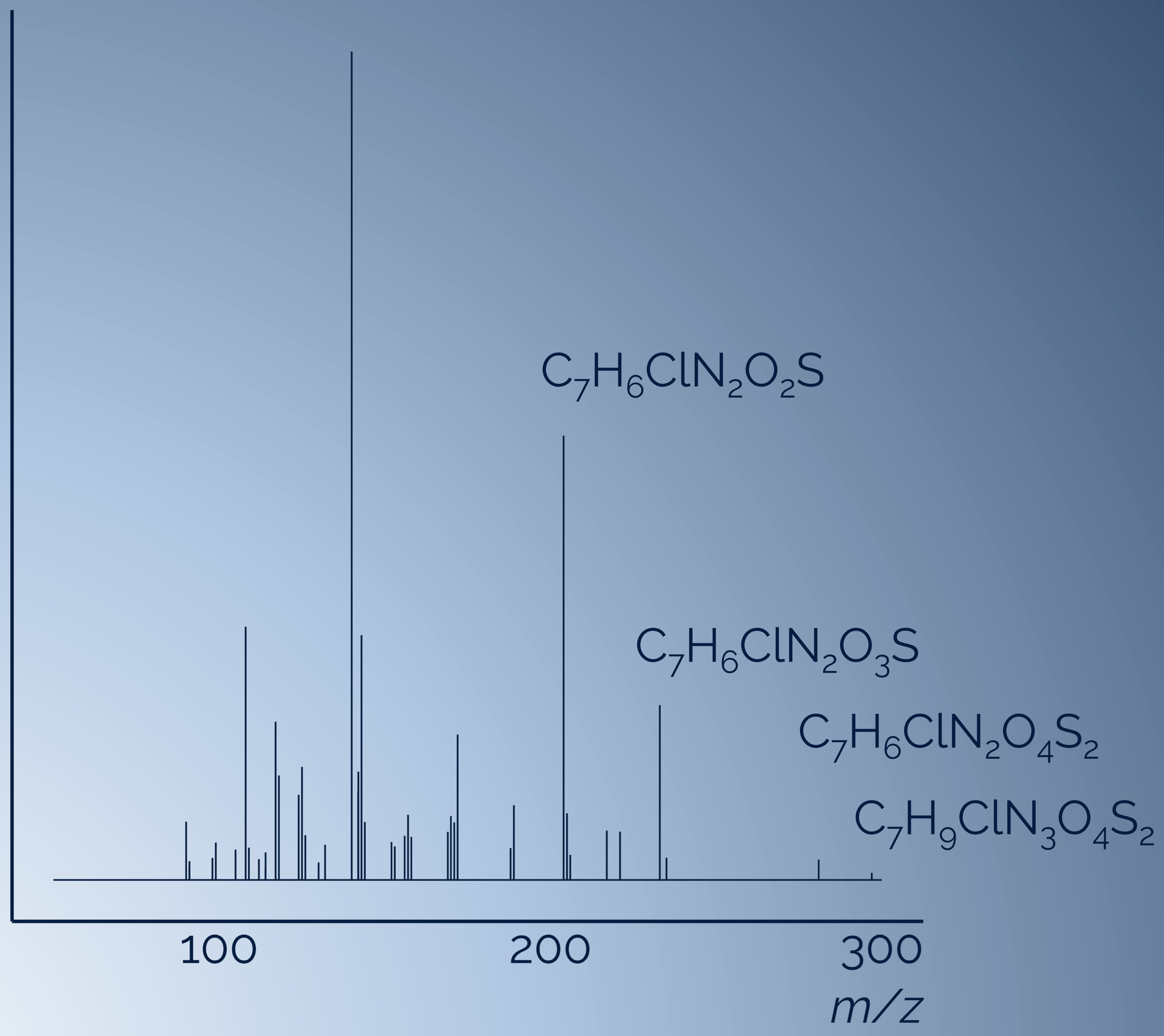

predict toxicity

for unknown chemicals

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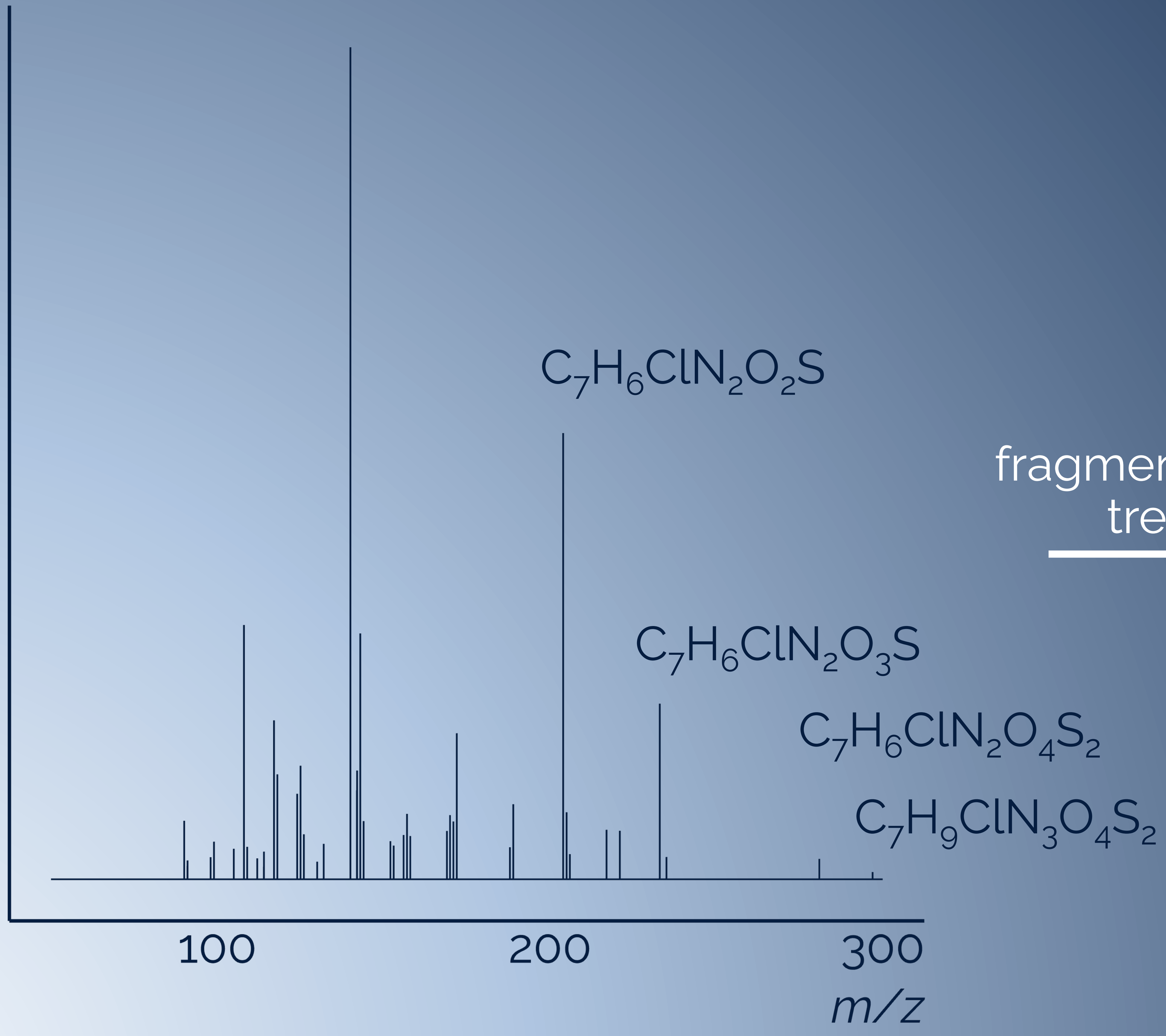
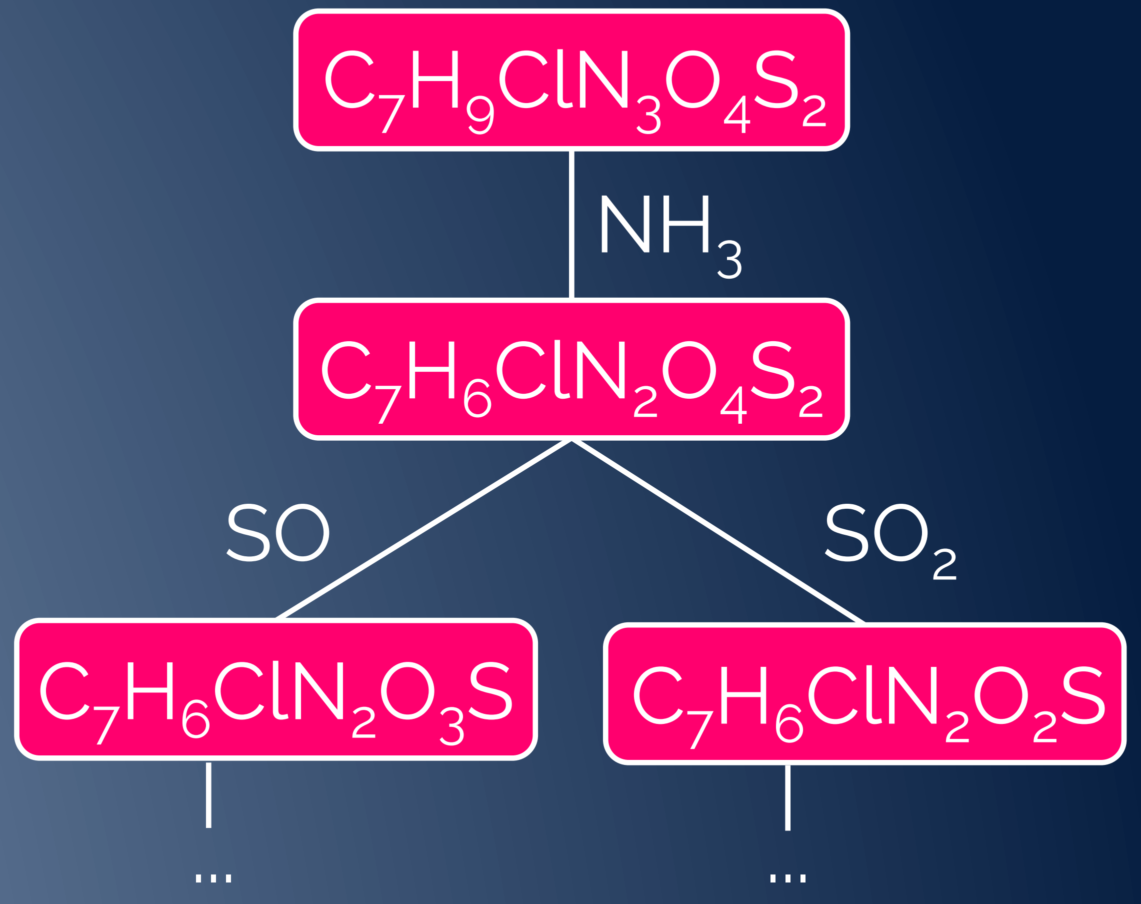


fragmentation tree



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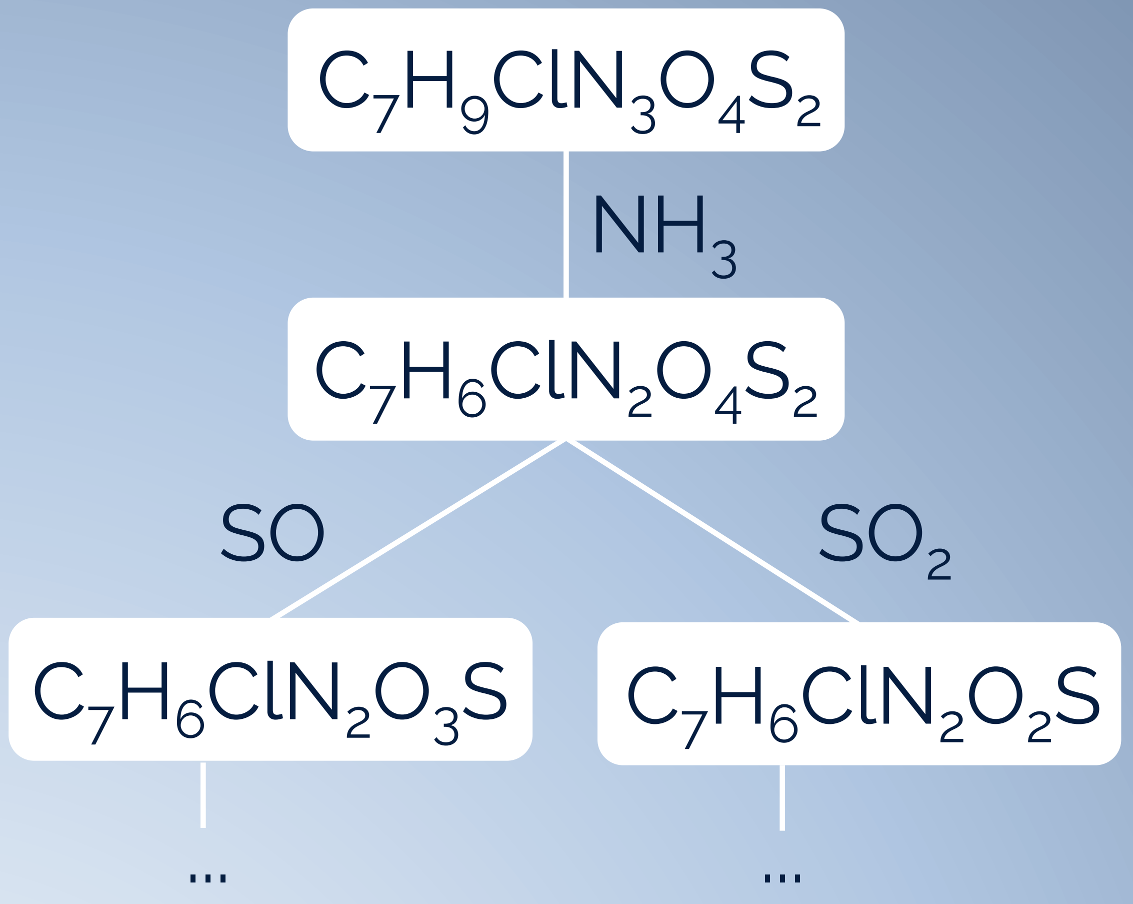

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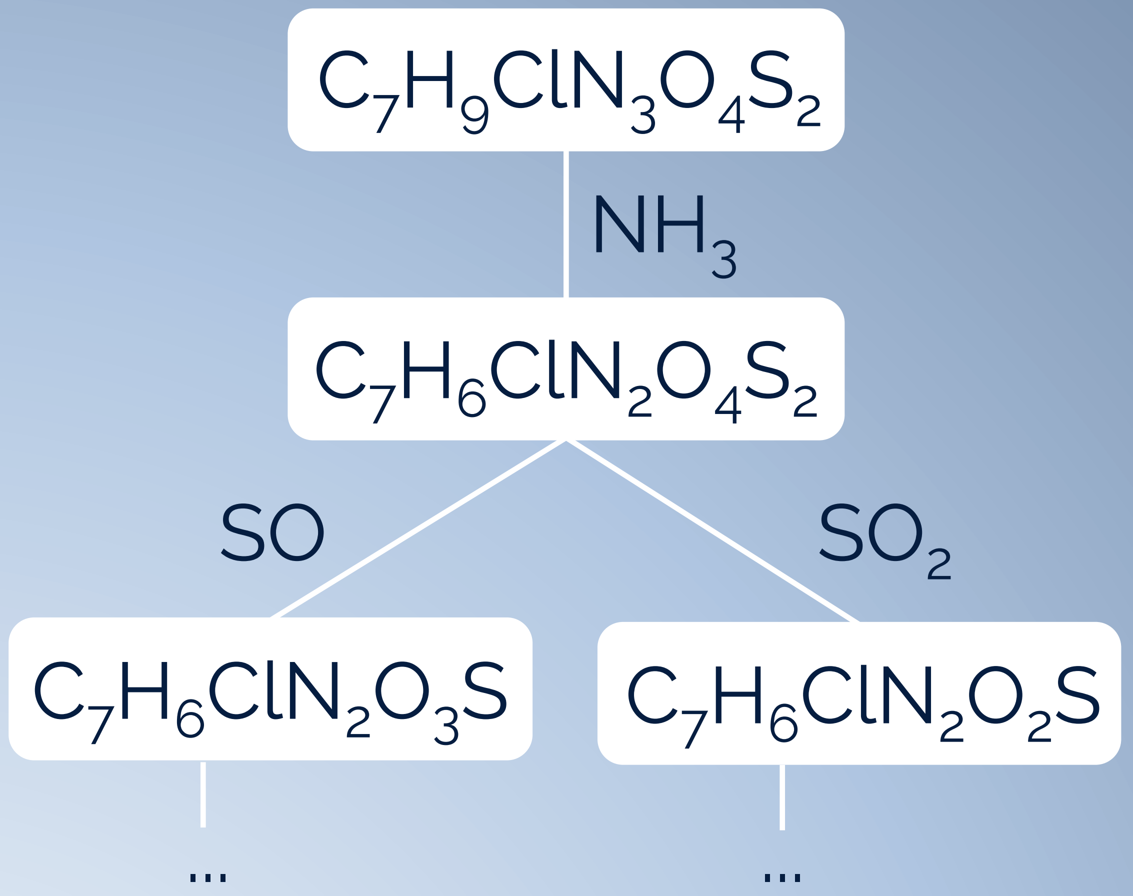

machine learning →

0	O-P
1	S=O
0	C-F
1	NH_2
1	O

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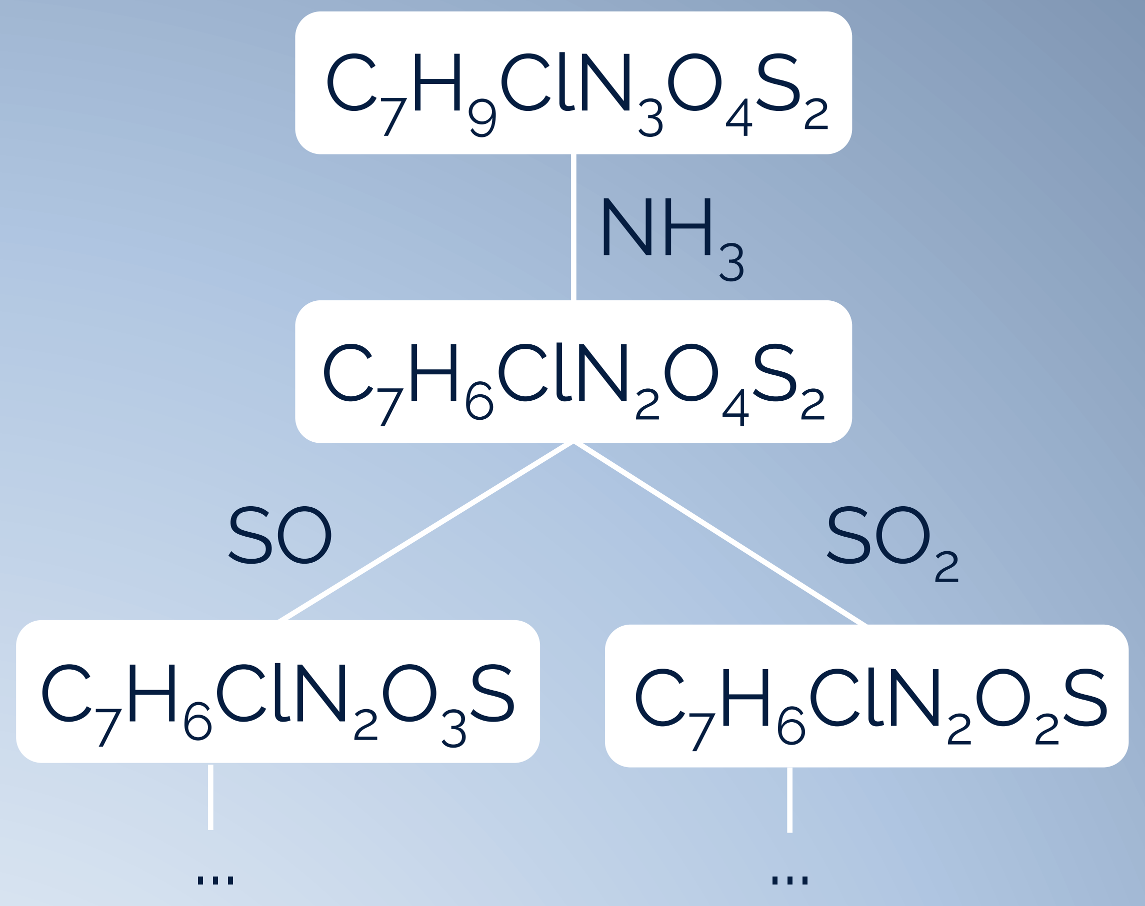

machine learning →

0	O-P
1	S=O
0	C-F
1	NH_2
1	O

machine learning →

p(AhR active)= 0.83

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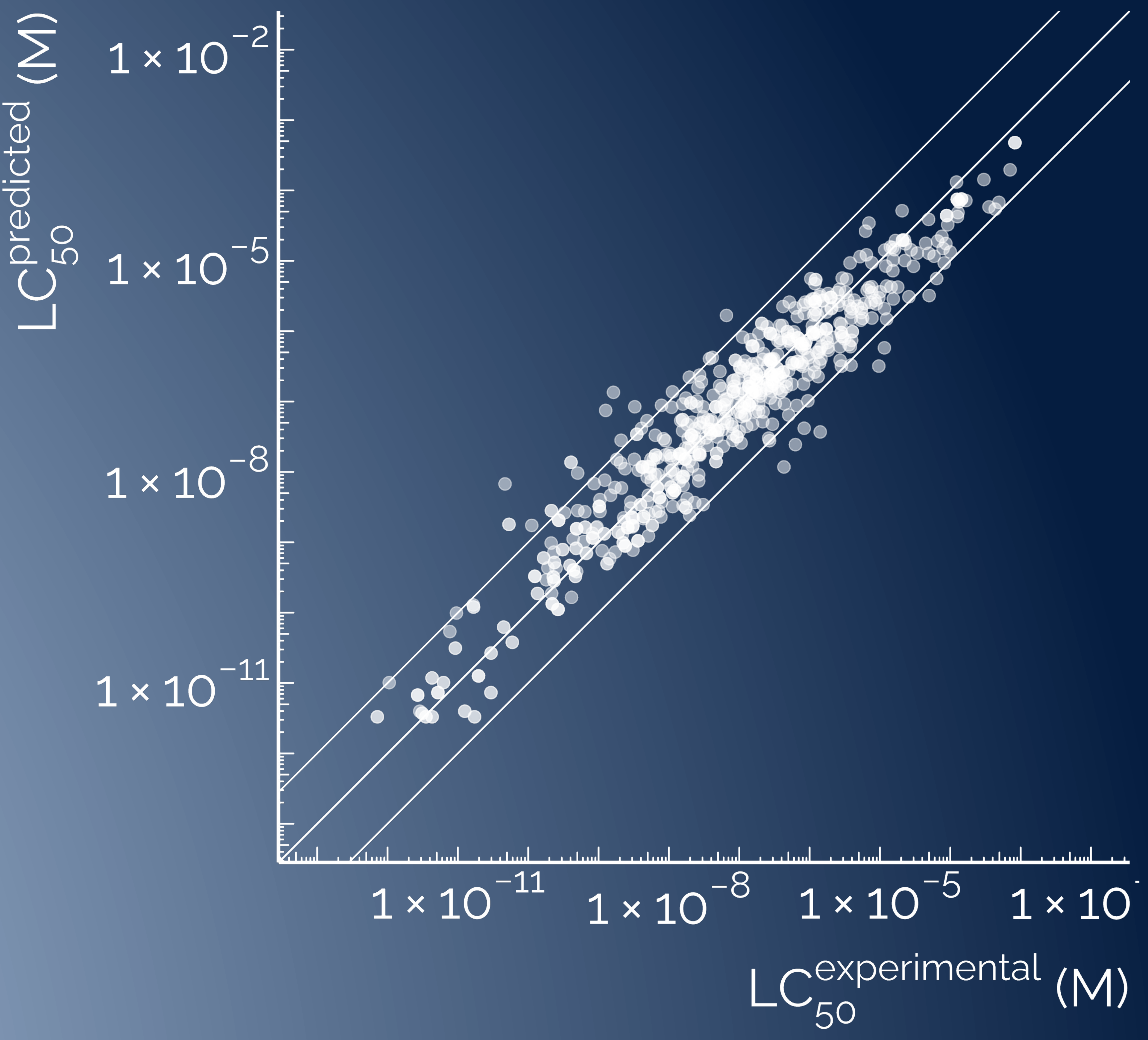
LC_{50} predictions

Peets et al. ES&T 2022

fish LC_{50}



test set

RMSE 0.78 log(M)

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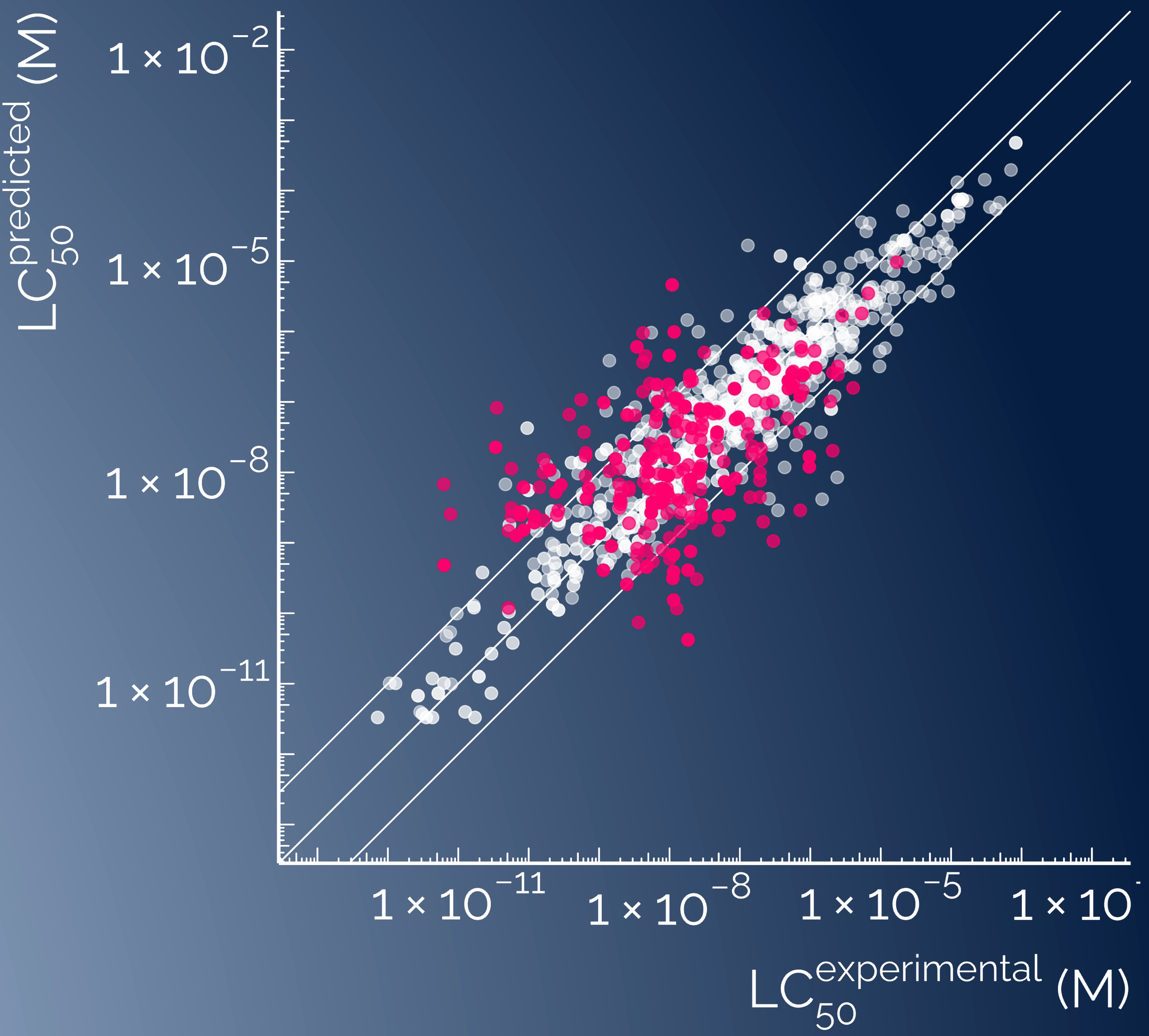
LC_{50} predictions

Peets et al. ES&T 2022

fish LC_{50}



validation on MassBank

$RMSE_{model}$ 0.88 log(M)

$SD_{experimental}$ 0.44 log(M)

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endocrine disruption

Rahu et al. JCIM 2024
Tox21 endpoints

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endocrine disruption

Rahu et al. JCIM 2024
Tox21 endpoints

true label	
active	non-active

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endocrine disruption

Rahu et al. JCIM 2024
Tox21 endpoints

		true label	
		active	non-active
prediction	active		
	non-active		

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endocrine disruption

Rahu et al. JCIM 2024
Tox21 endpoints

		true label	
		active	non-active
prediction	active	TP	
	non-active		TN

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endocrine disruption

Rahu et al. JCIM 2024
Tox21 endpoints

		true label	
		active	non-active
prediction	active	TP	FP
	non-active		TN

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endocrine disruption

Rahu et al. JCIM 2024
Tox21 endpoints

		true label	
		active	non-active
prediction	active	TP	FP
	non-active	FN	TN

which is more dramatic:
FP or FN?

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endocrine disruption

Rahu et al. JCIM 2024
Tox21 endpoints

		true label	
		active	non-active
prediction	active	TP	FP
	non-active	FN	TN

FPR @ TPR = 0.9

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endocrine disruption

Rahu et al. JCIM 2024
Tox21 endpoints

bioassay	FPR
sr.mmp	25.1%
sr.p53	25.4%
nr.ahr	41.8%
...	...
nr.ar	82.4%
nr.er	85.0%

MassBank & MoNA
748 compounds

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alternative approaches

Kreutzer et al. in preparation
Tox21 endpoints

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molecular networks

Kreutzer et al. in preparation
Tox21 endpoints

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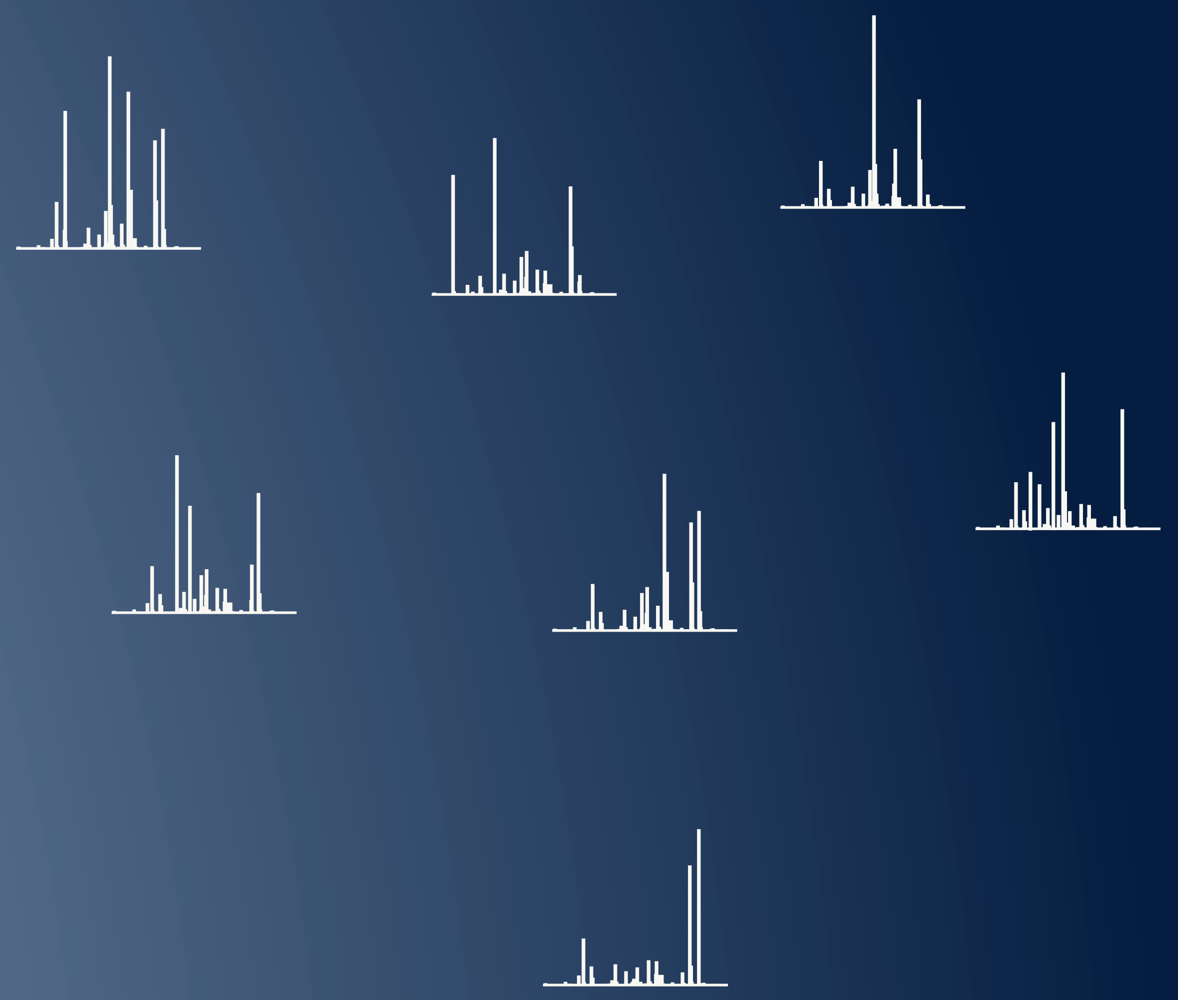
molecular networks

Kreutzer et al. in preparation
Tox21 endpoints



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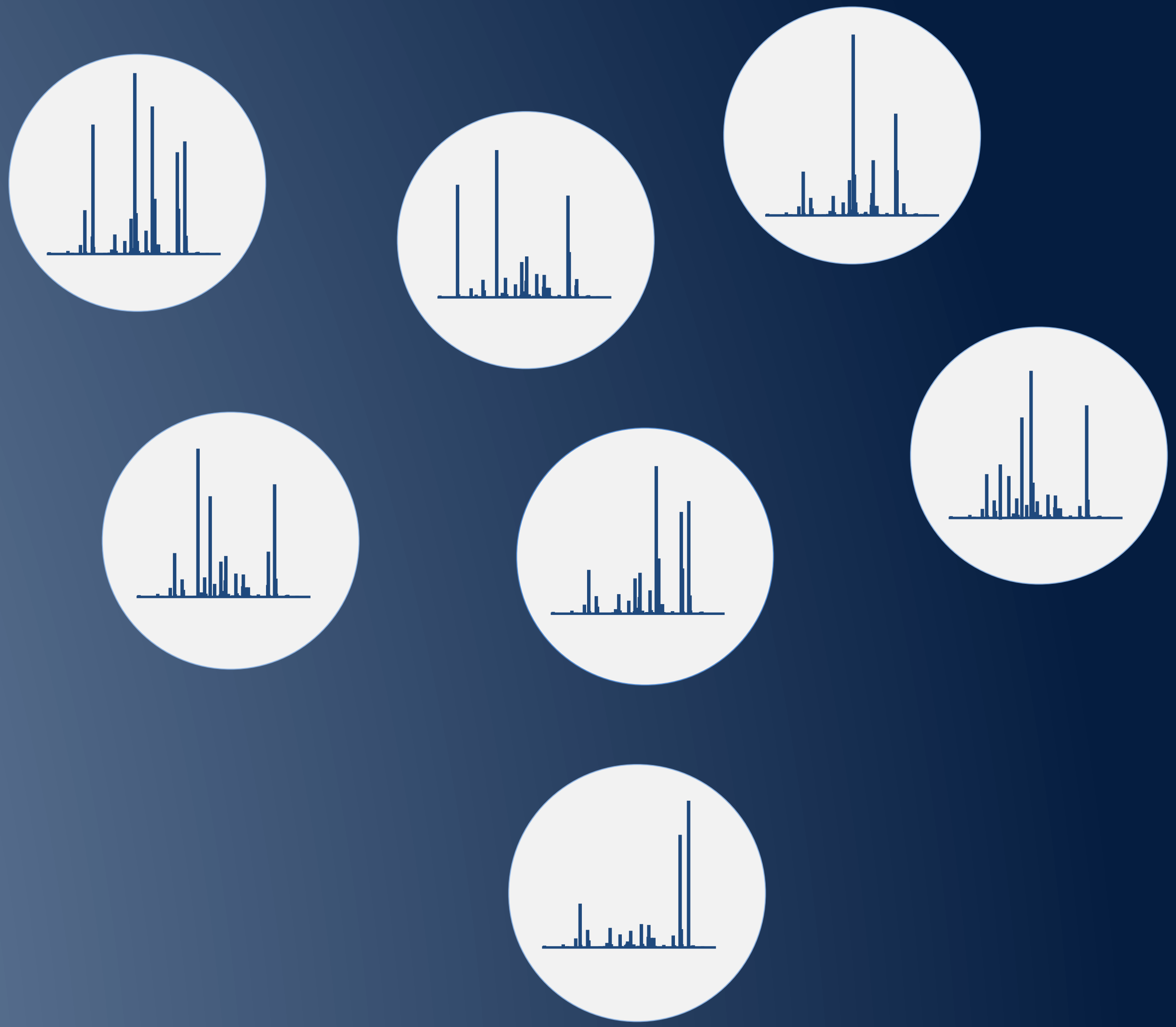
molecular networks

Kreutzer et al. in preparation
Tox21 endpoints



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molecular networks

Kreutzer et al. in preparation
Tox21 endpoints

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molecular networks

Kreutzer et al. in preparation
Tox21 endpoints

Active
Inactive
Inconclusive

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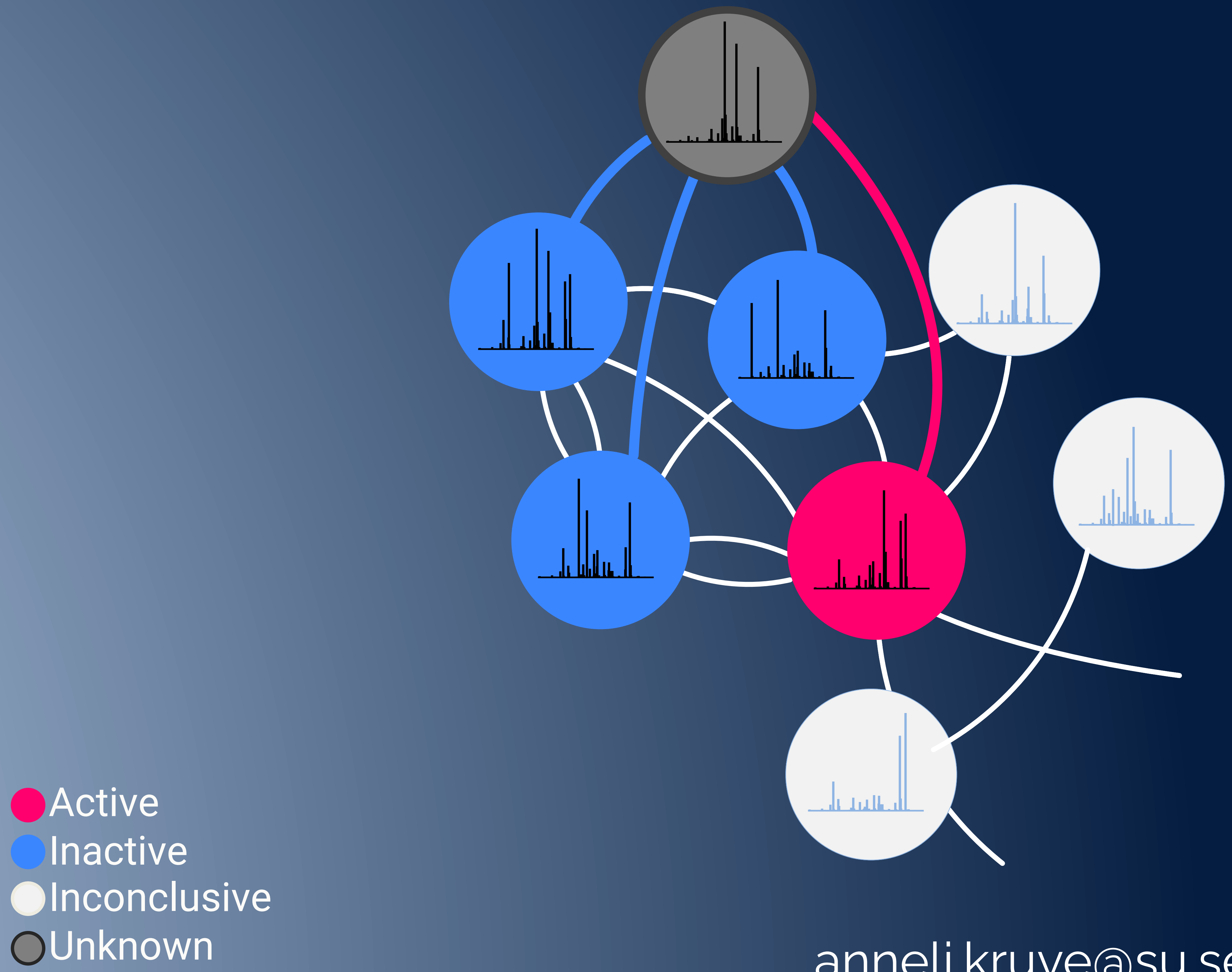
molecular networks

Kreutzer et al. in preparation
Tox21 endpoints



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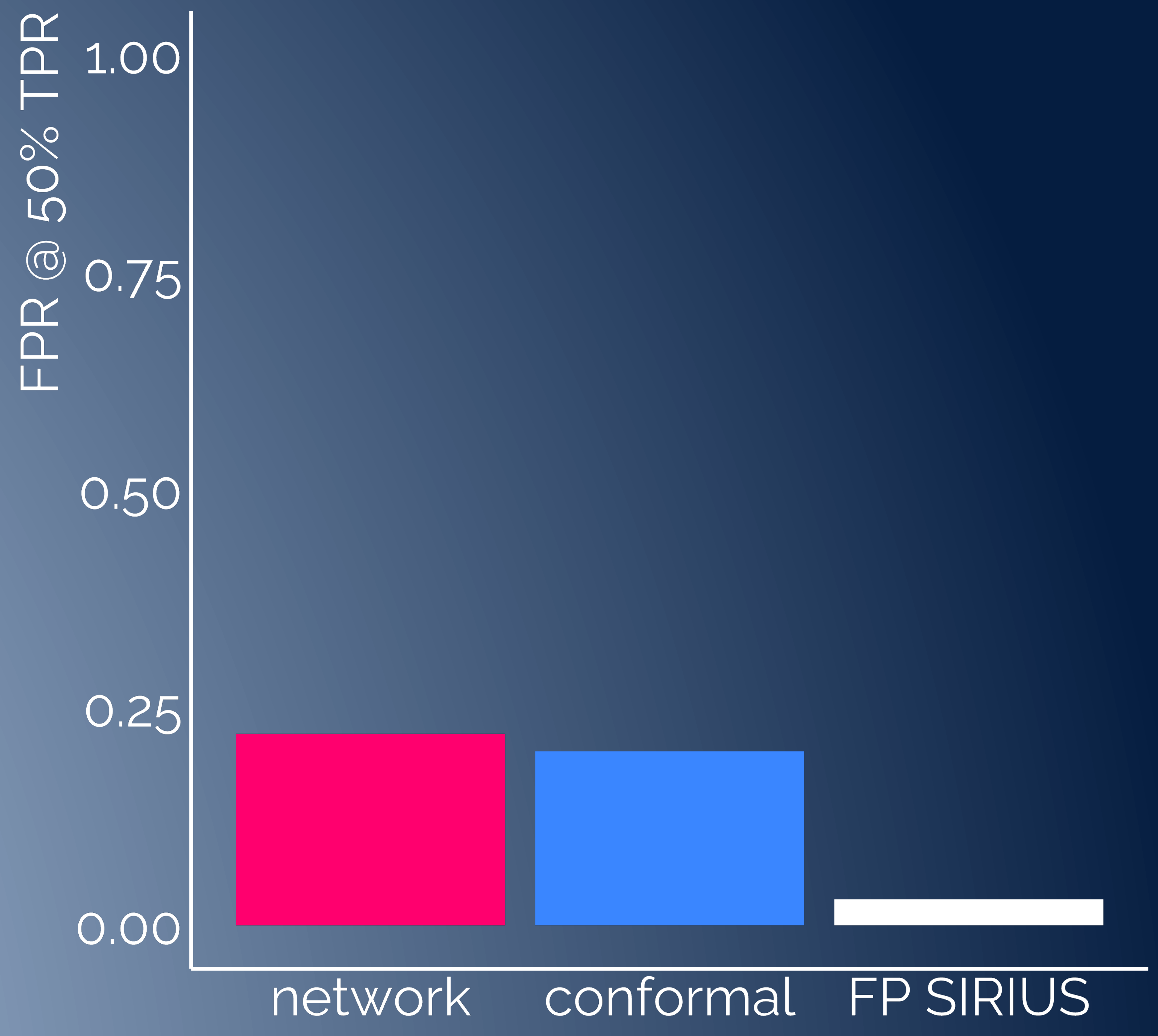
AhR activity predictions

Kreutzer et al. in preparation
Tox21 endpoints



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case study:
interlaboratory comparison

water analysis

thousands of chemicals detected

in the environment

Machine Translated by Google

BioCell ANALYTICA

Table 1. The following BEQ values were measured in the current samples:

	Nrf2 activity	AntiÿAR activity	AR activity	ER activity	AhR activity
	µg/L (tBHQ equivalents)	ng/L (OHF equivalents)	ng/L (DHT equivalents)	pg/L (E2 equivalents)	ng/L (TCDD equivalents)
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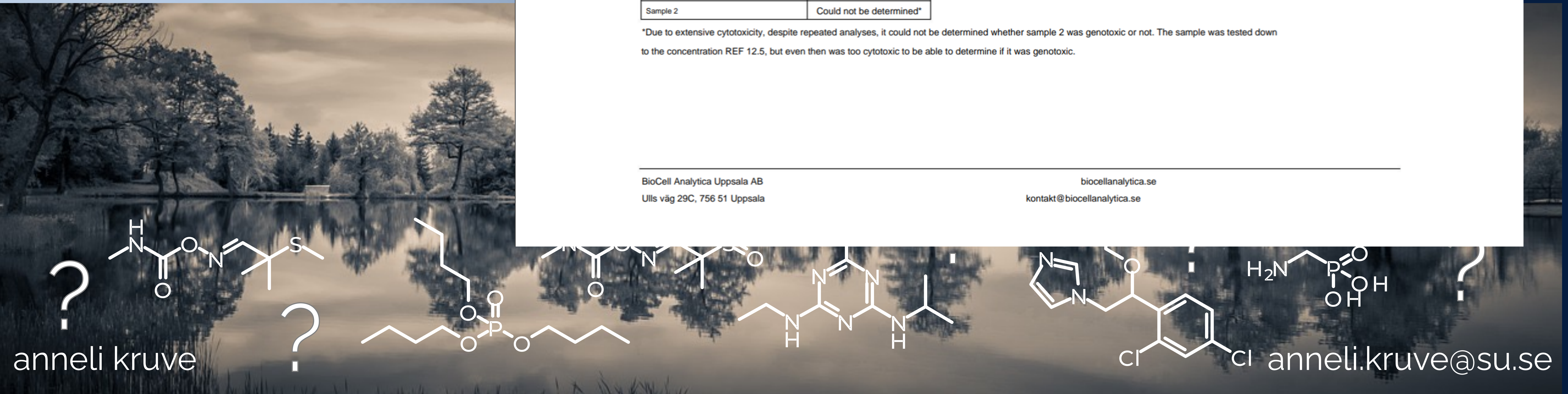
Sample 2	992	2670	<LOD*	<LOD*	<LOD*
Detection limit	8.34	43.8	0.122	12.5	0.0196
detection limit*			6.93	50.0	0.156

Table 2. Genotoxicity.

	Genotoxic?
Reference to sample 1	No
Sample 1	No
Reference to sample 2	No
Sample 2	Could not be determined*

*Due to extensive cytotoxicity, despite repeated analyses, it could not be determined whether sample 2 was genotoxic or not. The sample was tested down to the concentration REF 12.5, but even then was too cytotoxic to be able to determine if it was genotoxic.



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interlaboratory comparison

Sandberg, Rahu, in preparation

SAMPLES

spiked water samples

ANALYSIS

HRMS, etc. characterization

DATA PROCESSING

AhR activity

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results

4700

LC/HRMS features detected

238

features with MS^2 spectra

55

features predicted active

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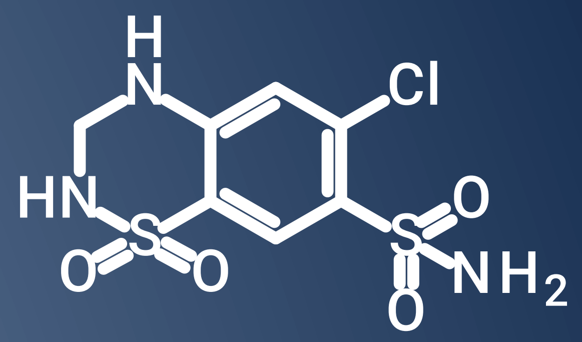
AhR active



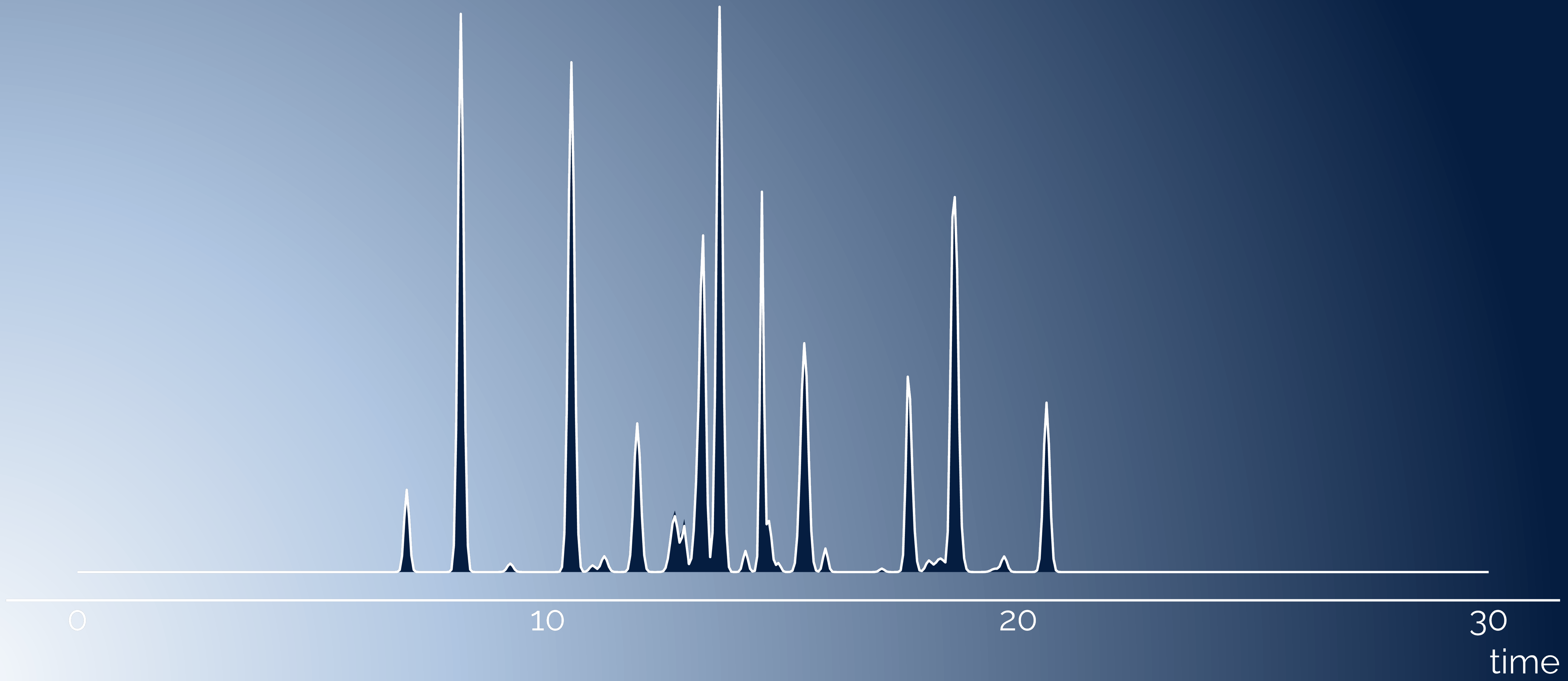
hydrochlorothiazide

quantification

of detected chemicals

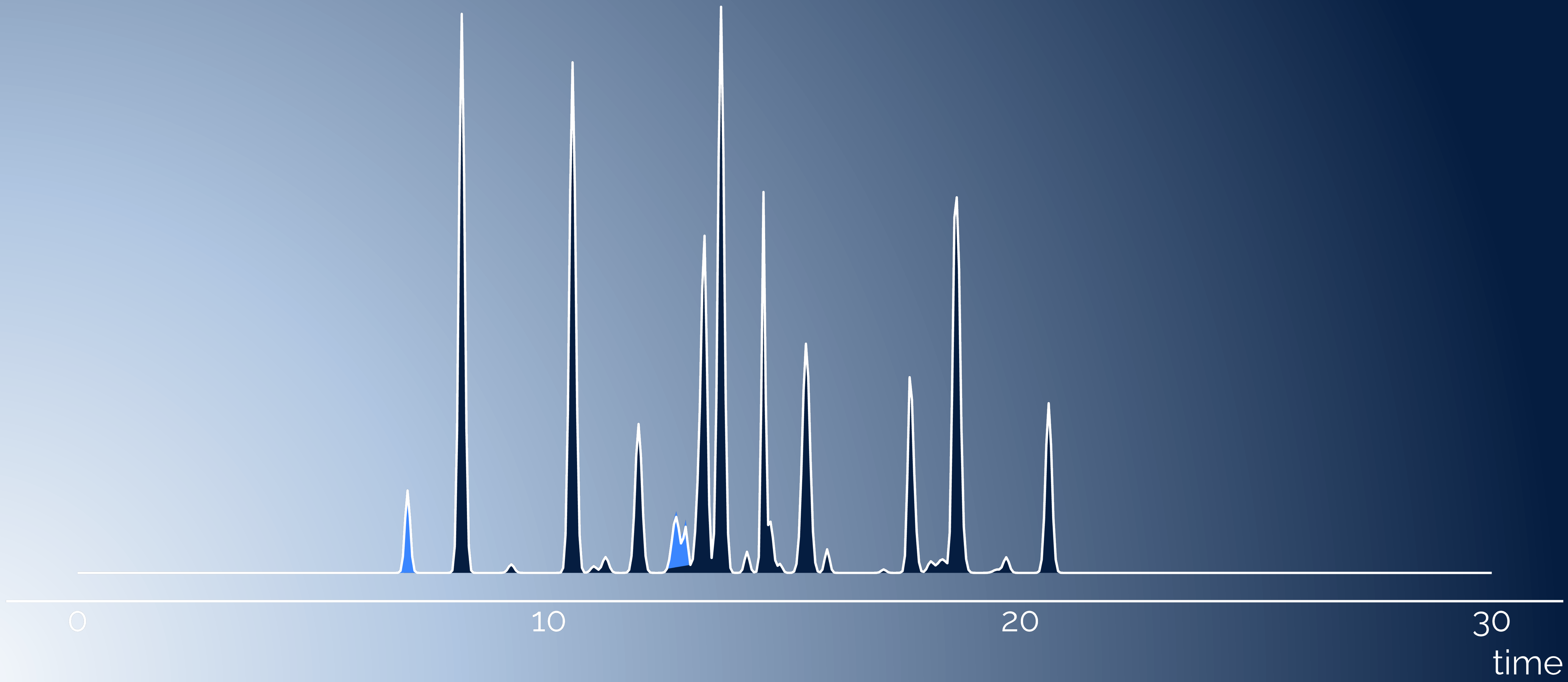
quantification in ESI/HRMS

Malm et al. Molecules 2021



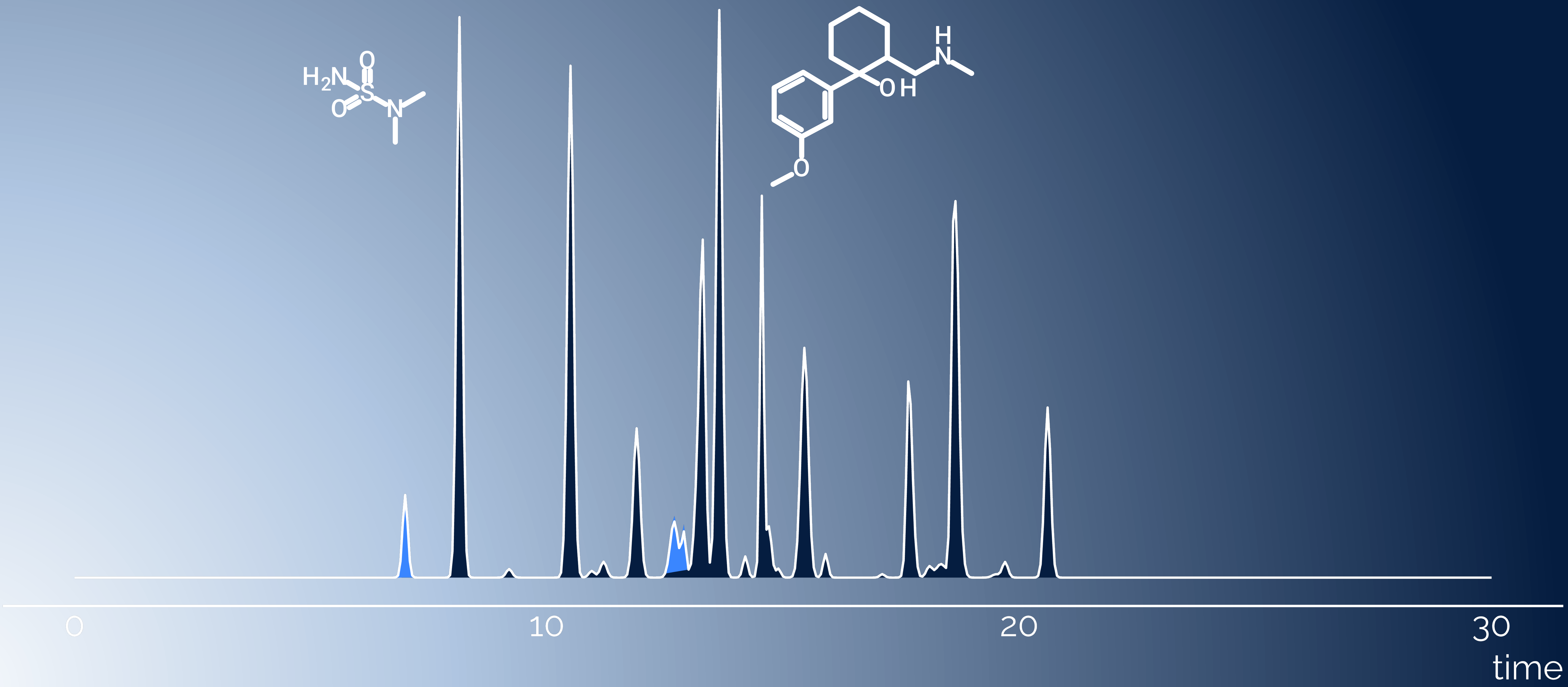
quantification in ESI/HRMS

Malm et al. Molecules 2021



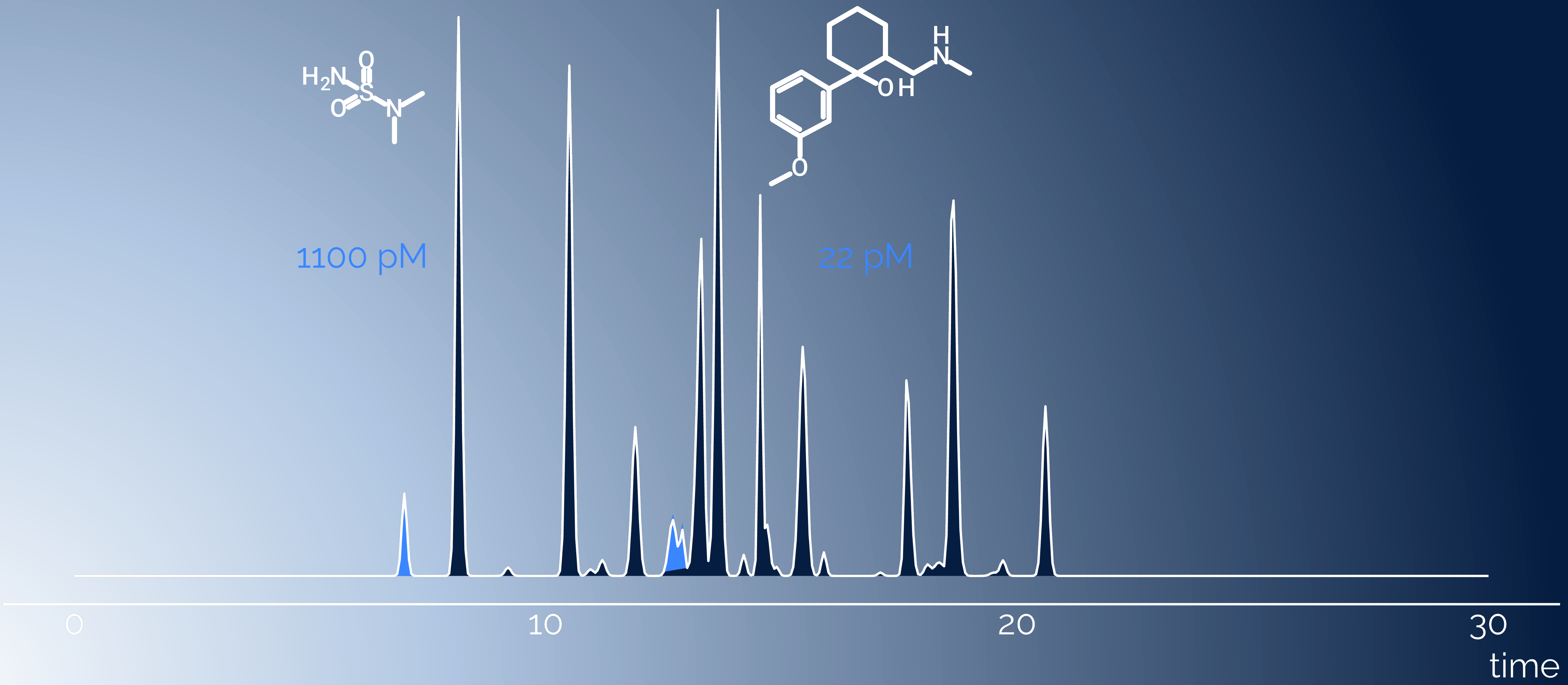
quantification in ESI/HRMS

Malm et al. Molecules 2021



quantification in ESI/HRMS

Malm et al. Molecules 2021



electrospray

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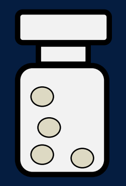
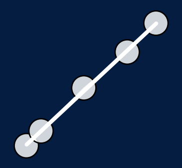
ionization efficiency



ANALYSIS

flow injections or LC



DATA

calibration graphs

$$\frac{slope_1}{slope_2} \rightarrow IE$$

IONIZATION EFFICIENCY

relative measurements

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ionization efficiency

one solvent, purely analyte properties

377 chemicals

10,000,000x difference in *IE*

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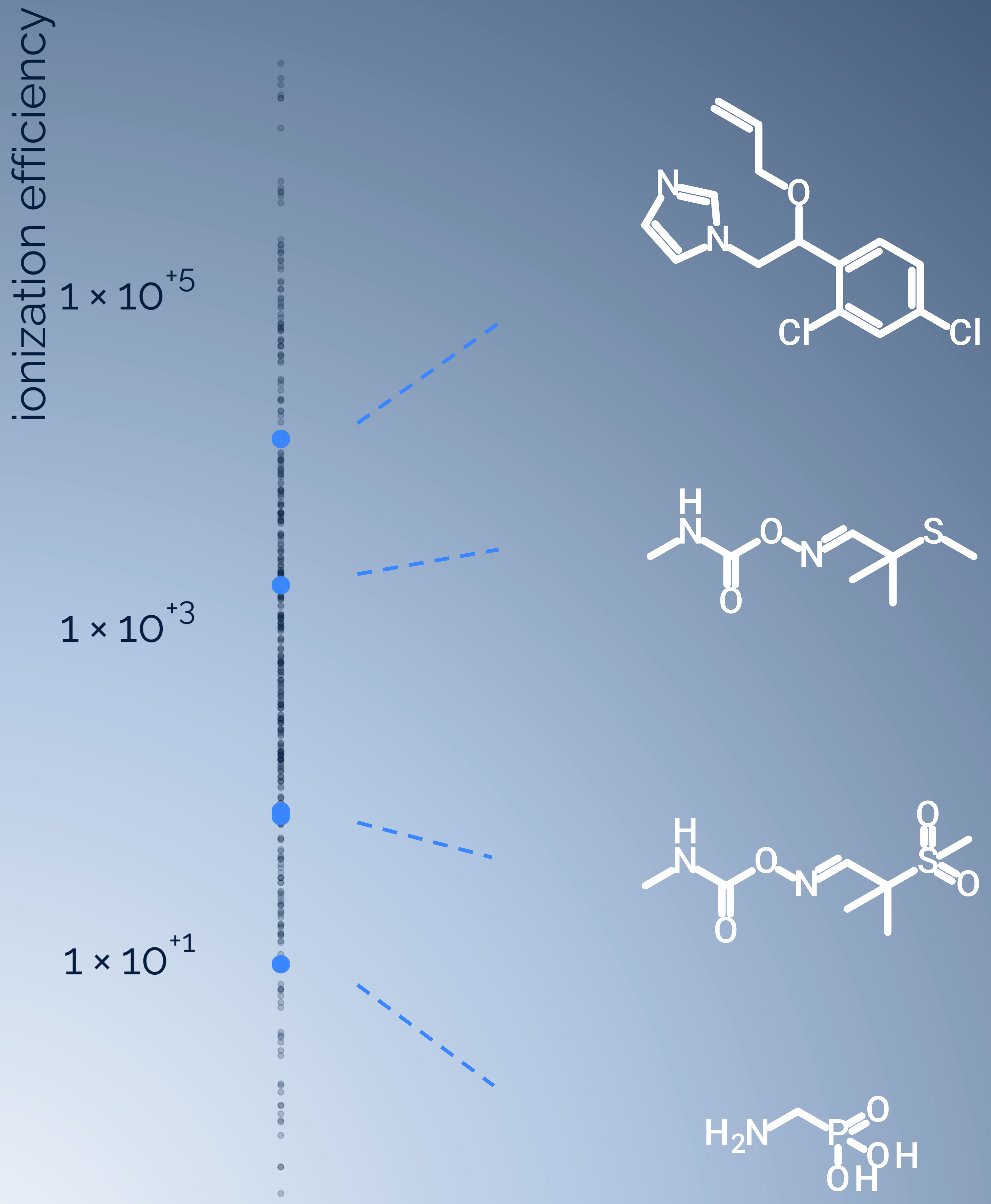

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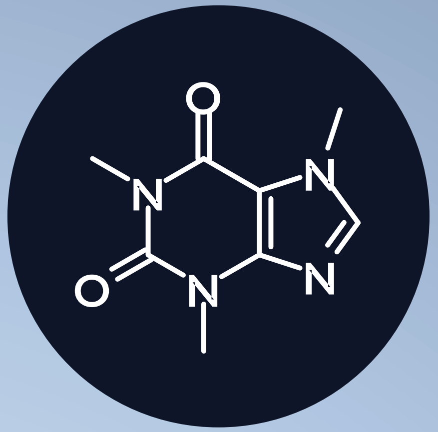
ionization efficiency



structure

mobile phase

instrument

matrix

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quantification

with machine learning

quantification approaches

CLOSE ELUTING

calibration graph

STRUCTURALLY SIMILAR

calibration graphs

IONIZATION EFFICIENCY

prediction model

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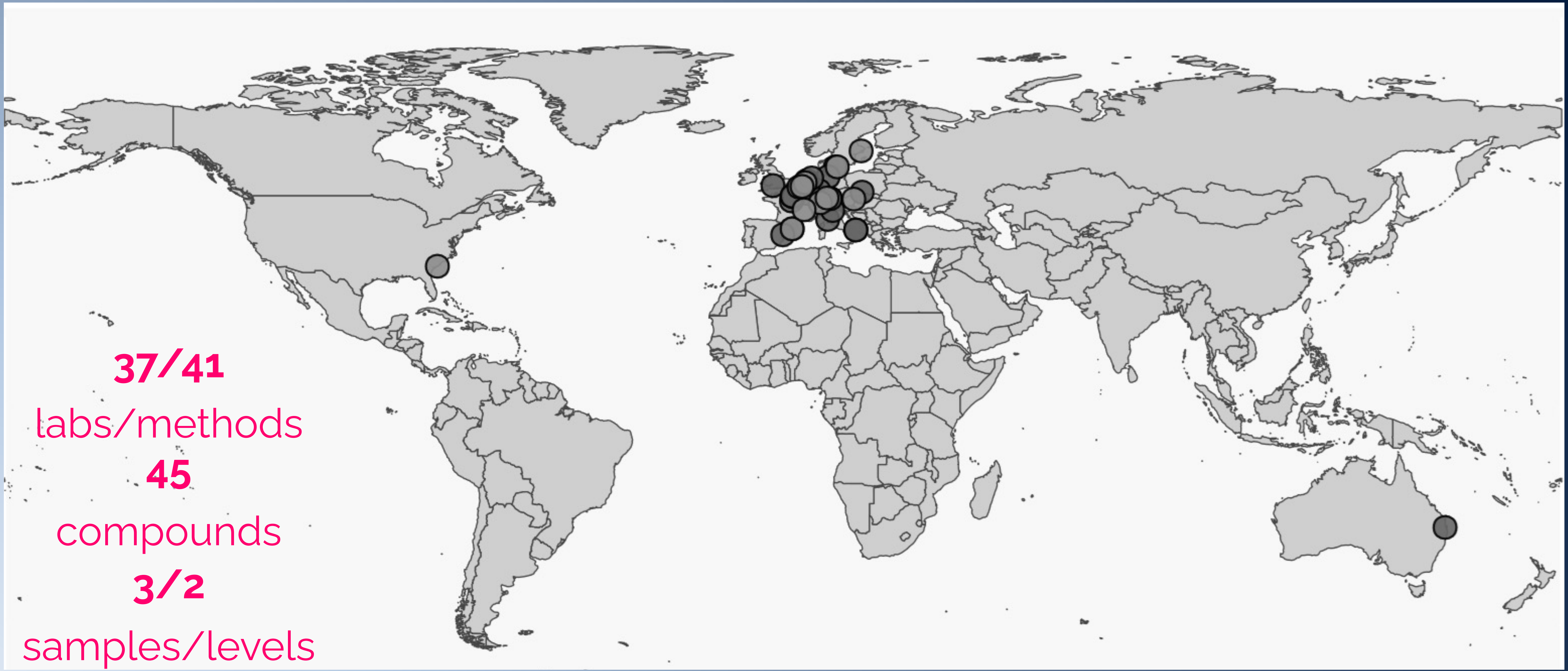
interlaboratory comparison

Malm et al. Anal Chem 2024



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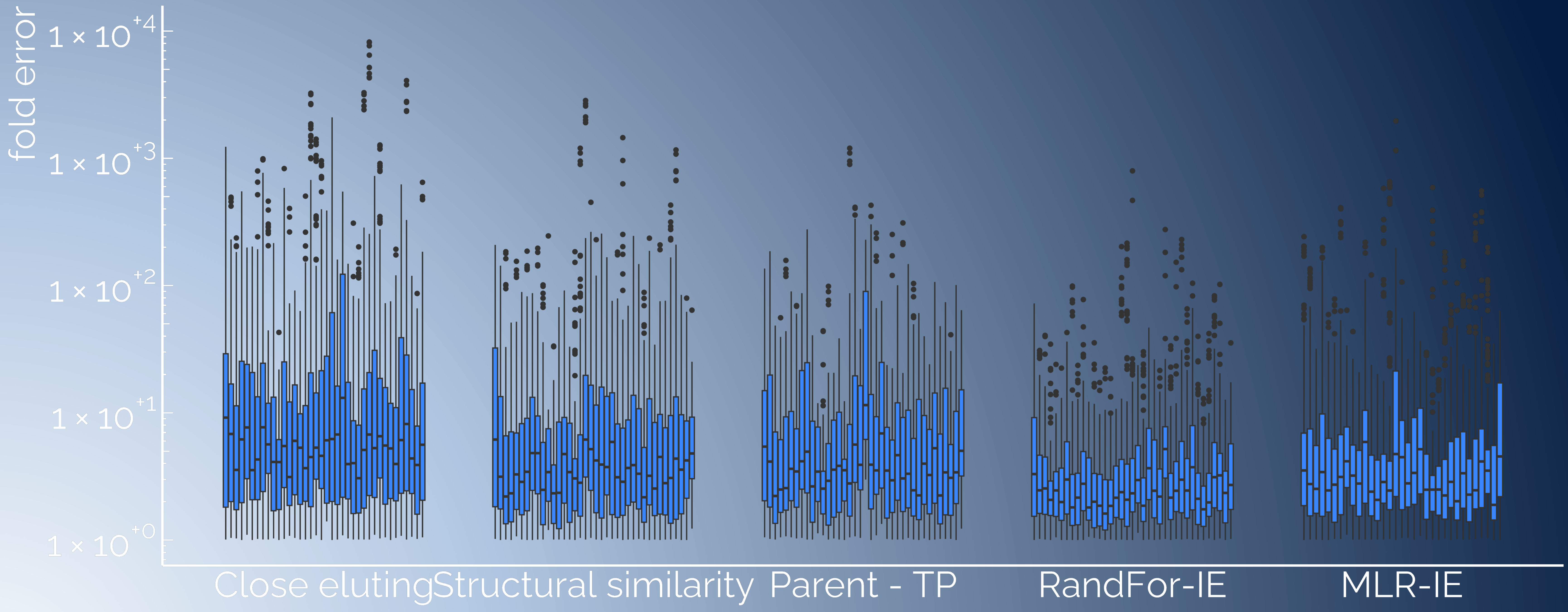
interlaboratory comparison

Malm et al. Anal Chem 2024



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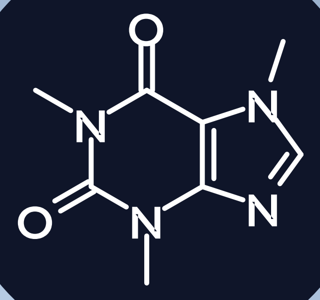
workflow



SMILES or MS²

+ mobile phase composition

features

PaDEL, Mordred, SIRIUS fingerprints

machine learning

RandomForest, xgbTree

model selection

Root Mean Square Error (RMSE)

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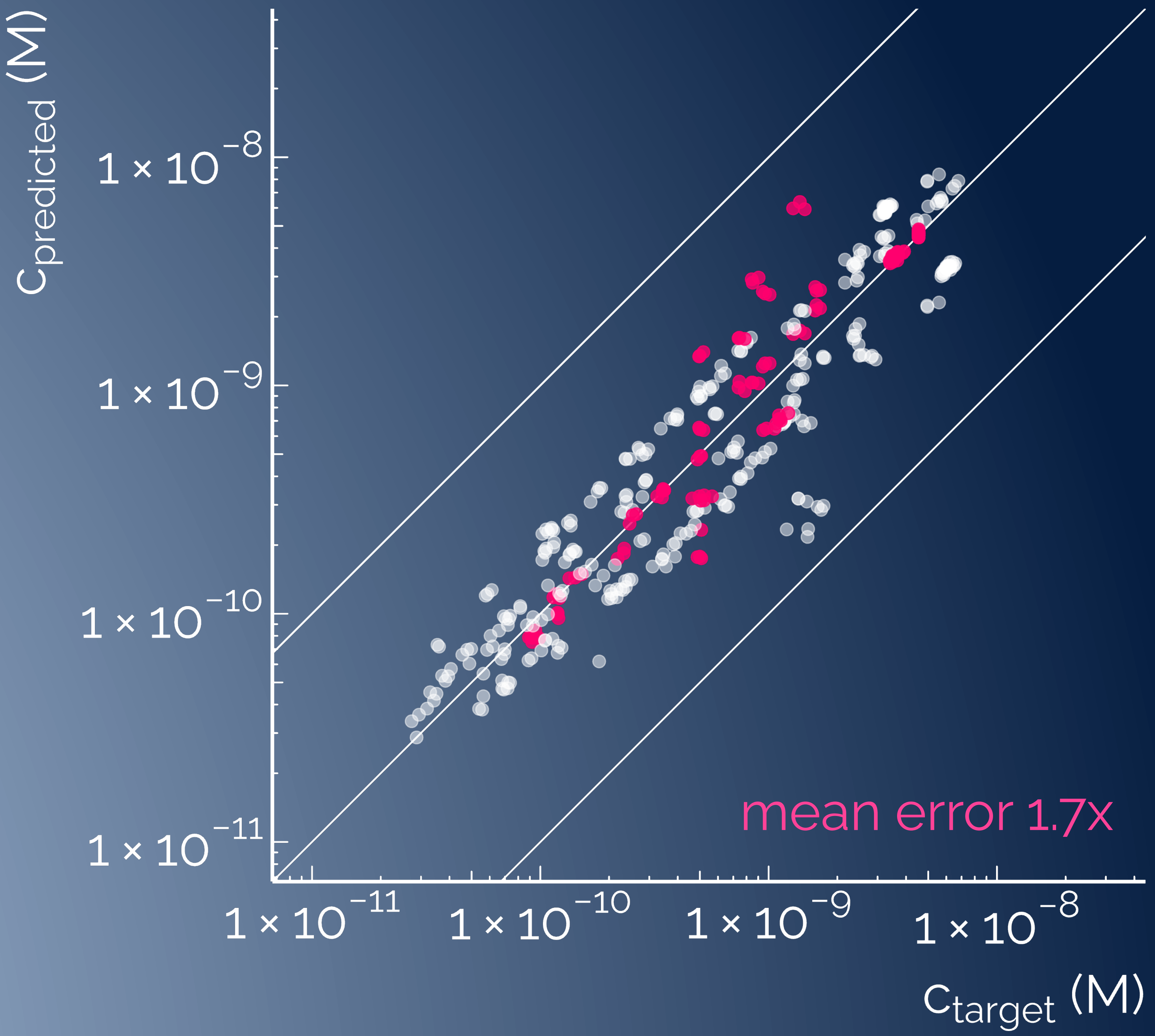
quantification

from structure

Been et al. Water Research 2021



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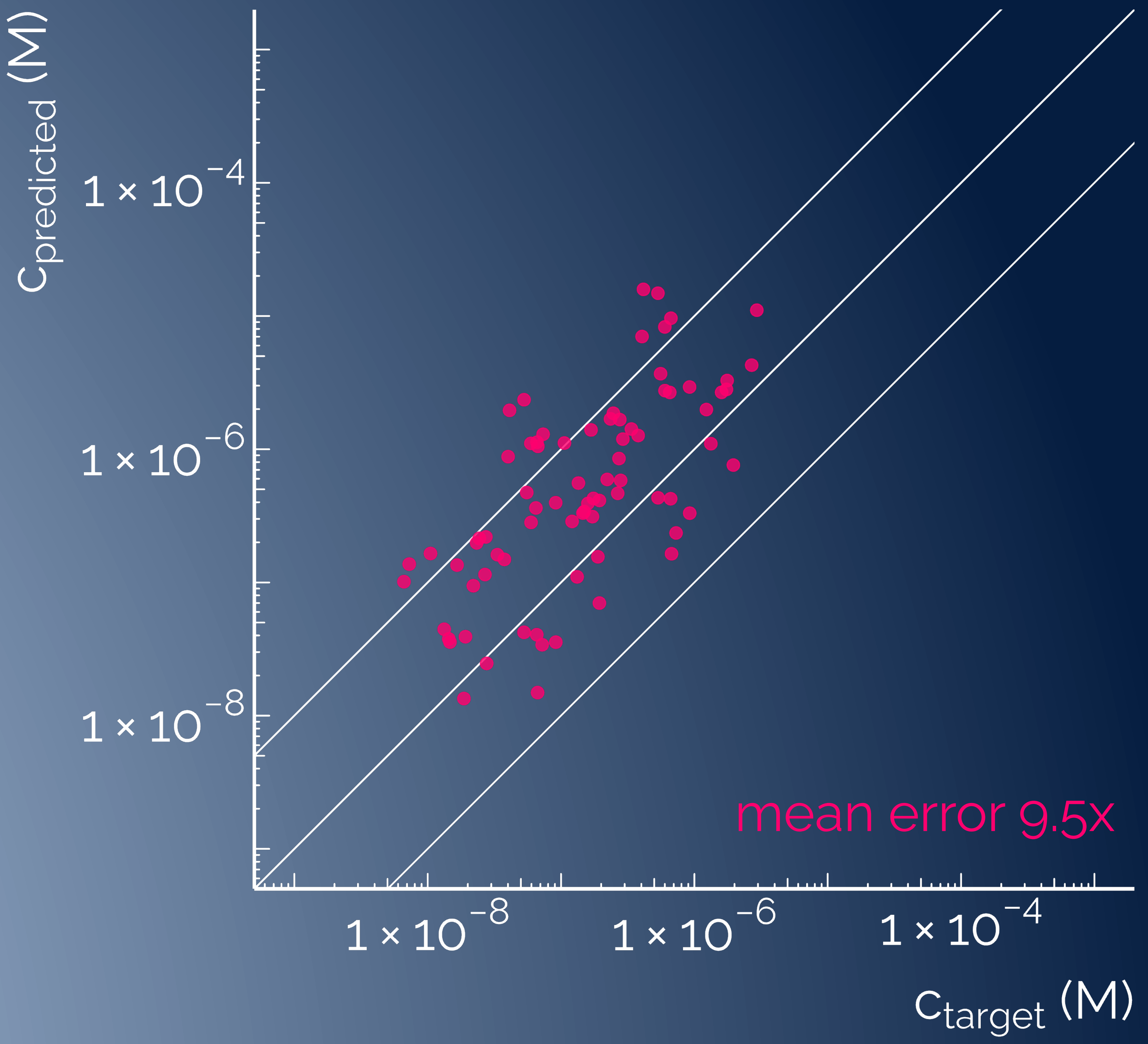
quantification

from MS² spectra

Sepman et al. Anal Chem 2023



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risk

based prioritization

prioritization of chemicals

TOXICITY

ecotoxicity and endocrine disruptors

CONCENTRATION

exposure to potentially toxic chemicals

RISK

$$\text{PriorityScore} = \frac{c_{\text{predicted}}}{\text{AC}_{50}^{\text{5th percentile}}}$$

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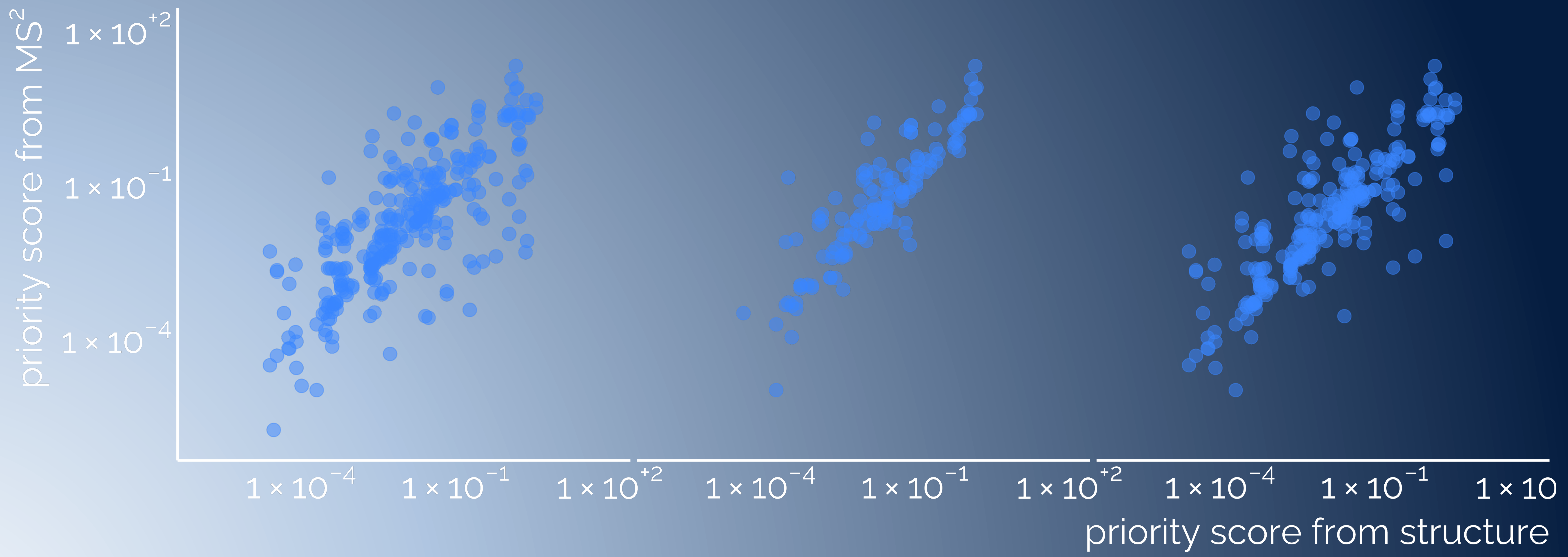
interlaboratory comparison

Sepman, in preparation



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how to …

PRIORITIZE

risk

IDENTIFY

structure

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how to …

PRIORITIZE

risk

IDENTIFY

structure

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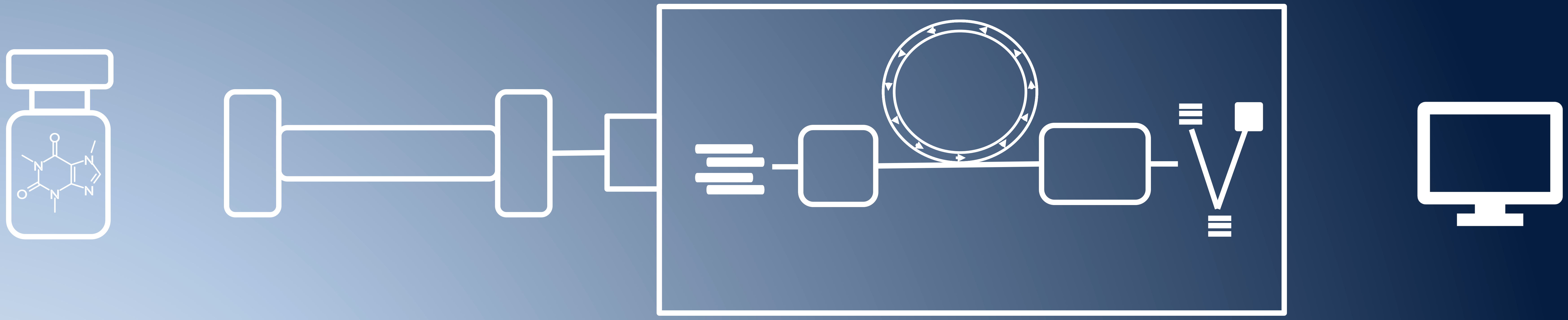
orthogonal separation

Akhlaqi et al. Anal Bioanal Chem 2023



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orthogonal separation

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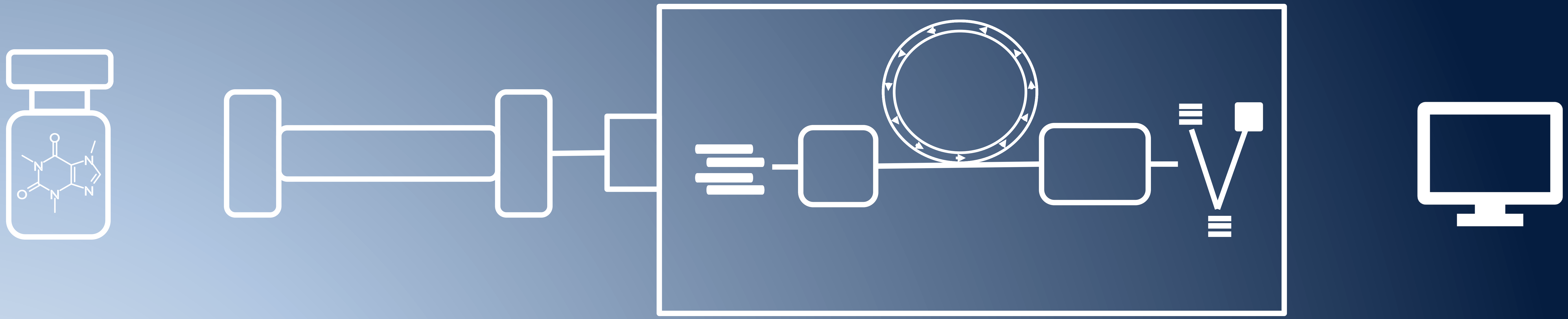

14 isomeric chemicals

C_{18} RP

Cyclic IMS & MS^2 with ToF

SIRIUS+ CSI:FingerID & CFM-ID

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orthogonal separation

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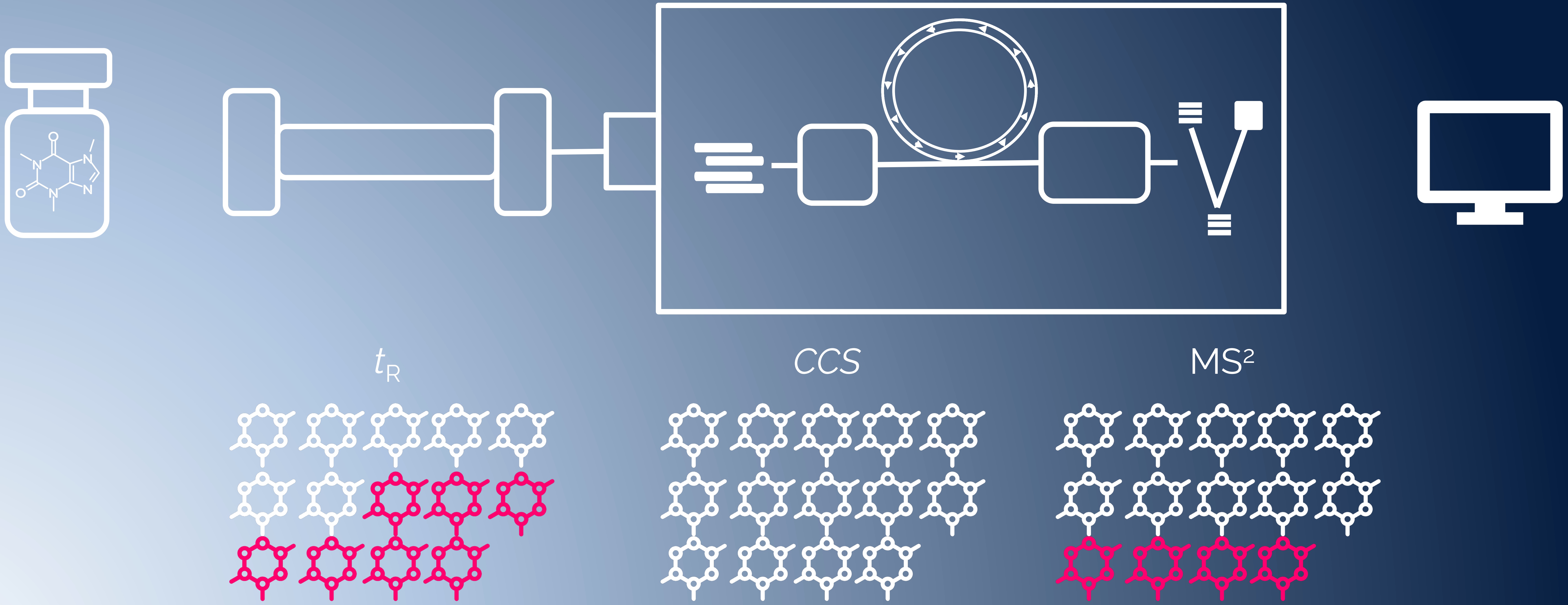

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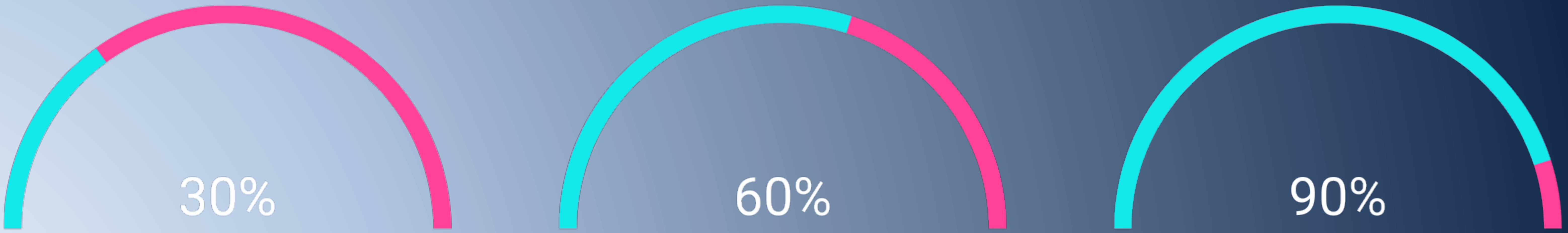
machine learning

for prioritization and identification in nontarget screening

TOXICITY QUANTIFICATION RETENTION TIME



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FORMAS

WISE Wallenberg Initiative Materials Science for Sustainability

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CARL TRYGGERS STIFTELSE
FÖR VETENSKAPLIG FORSKNING

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Exploring the research space...

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