Kruvelab.com

Stockholm University

Selecting mobile phase and column for non-targeted LC/ESI/HRMS analysis

Amina Souihi
amina.souihi@mmk.su.se

LC/ESI/HRMS

Water sample

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Water sample

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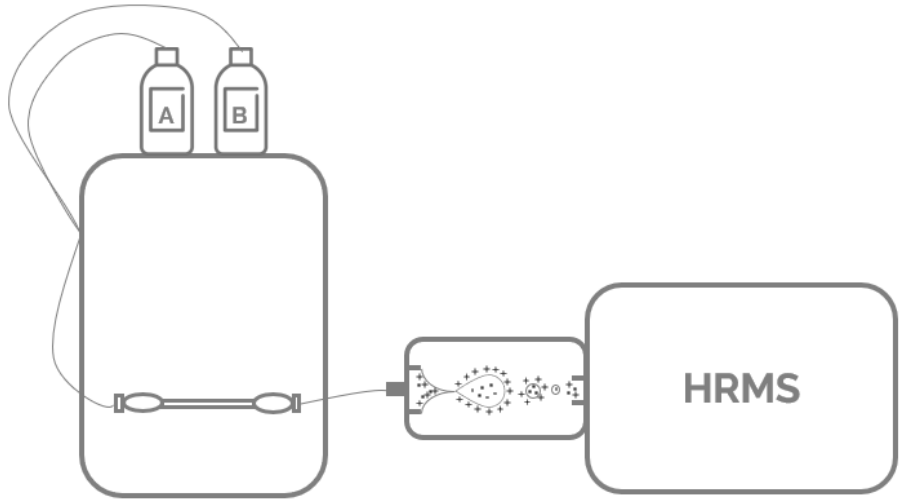
LC/ESI/HRMS

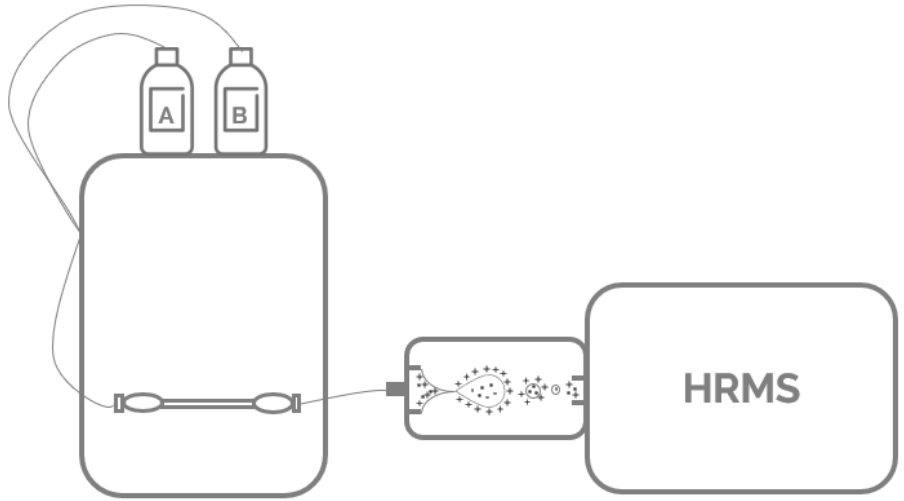
Water sample



Non-targeted LC/ESI/HRMS

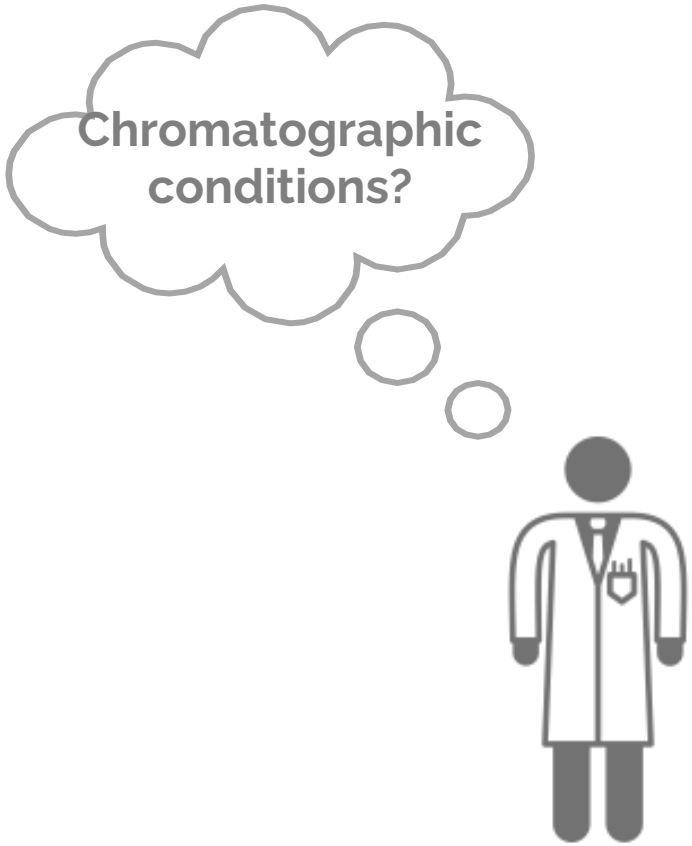
LC/ESI/HRMS

Water sample

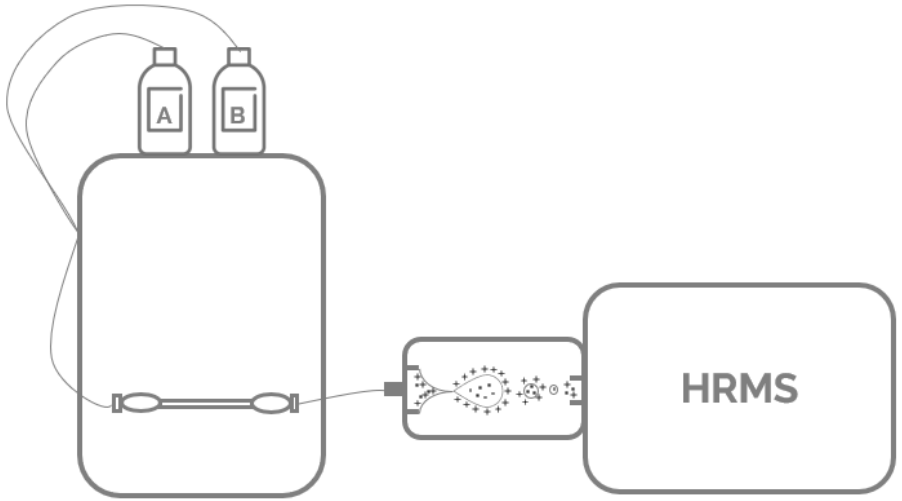


Non-targeted LC/ESI/HRMS

LC/ESI/HRMS

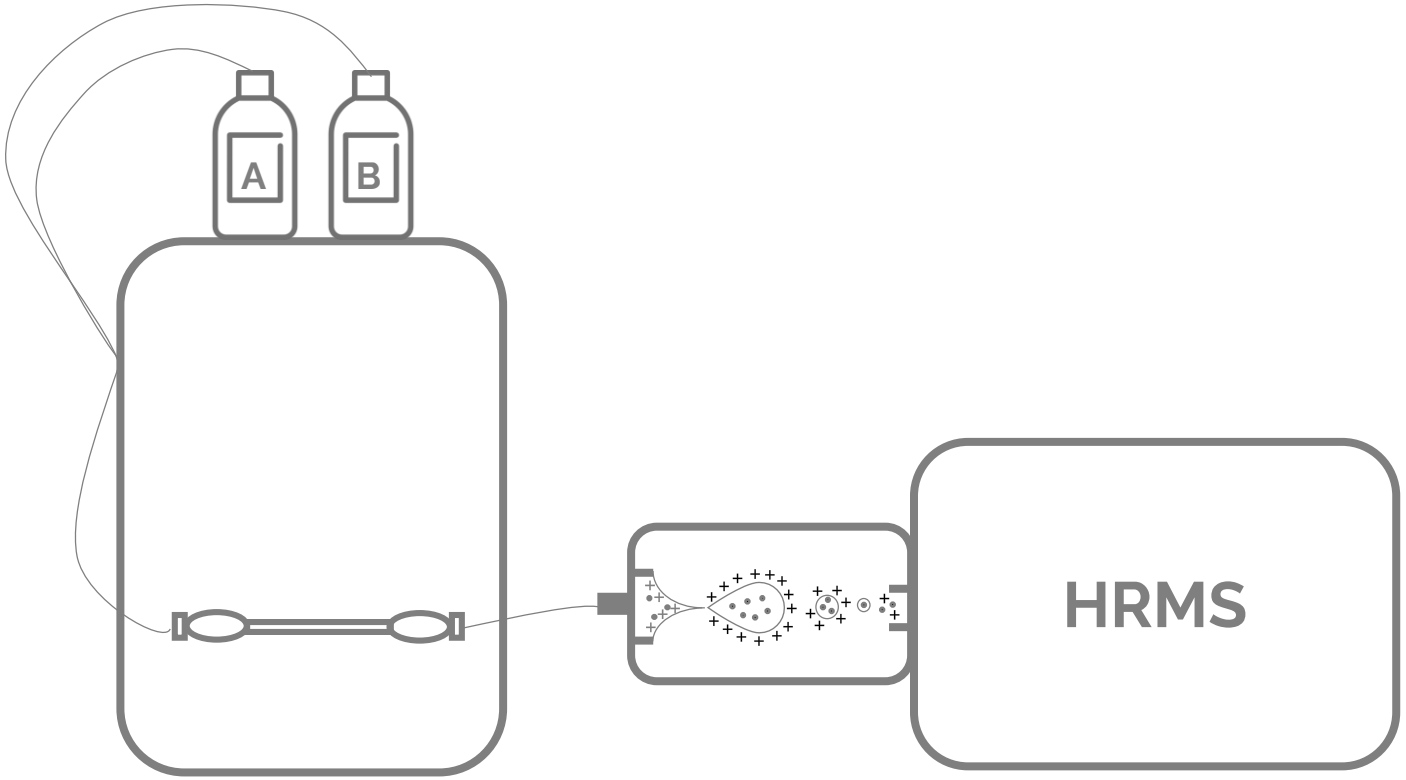


Water sample

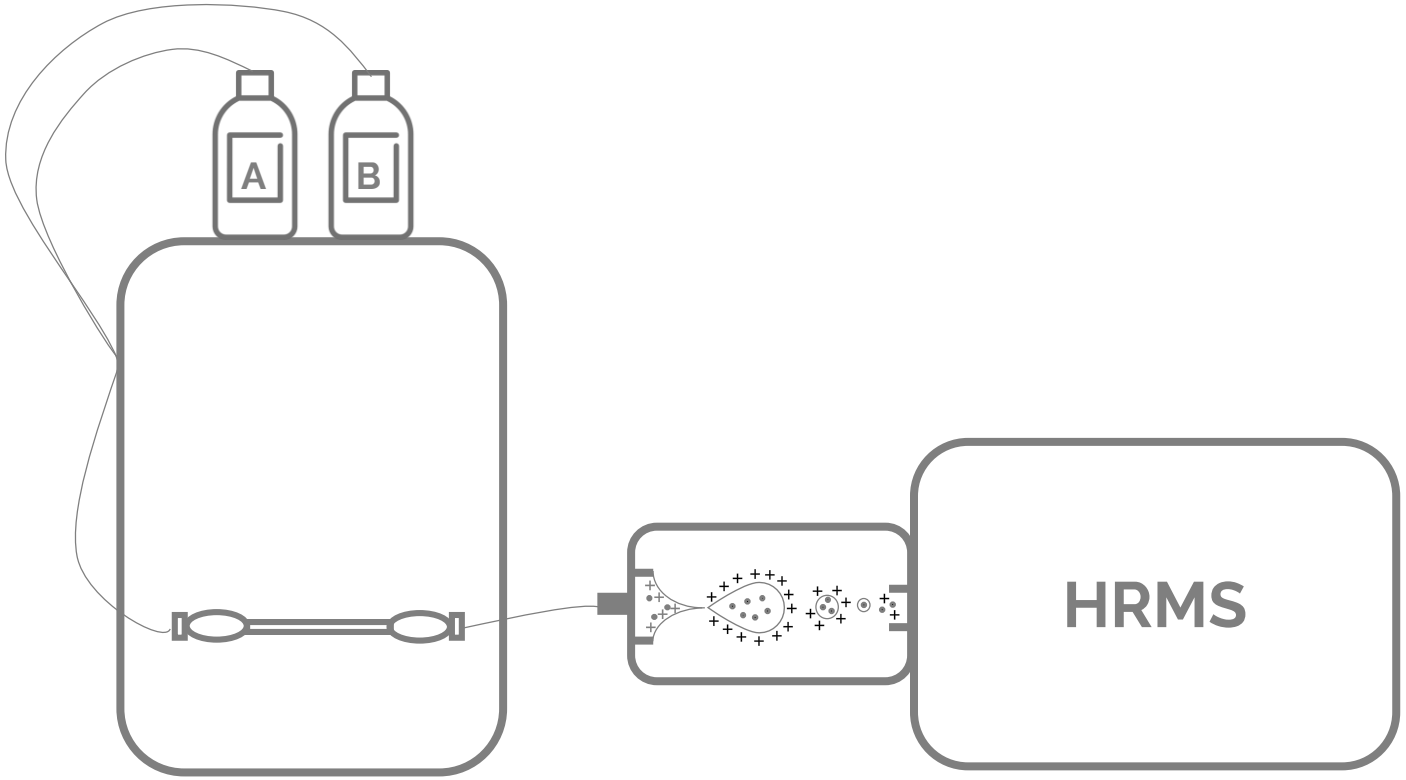


Non-targeted LC/ESI/HRMS

AIMS

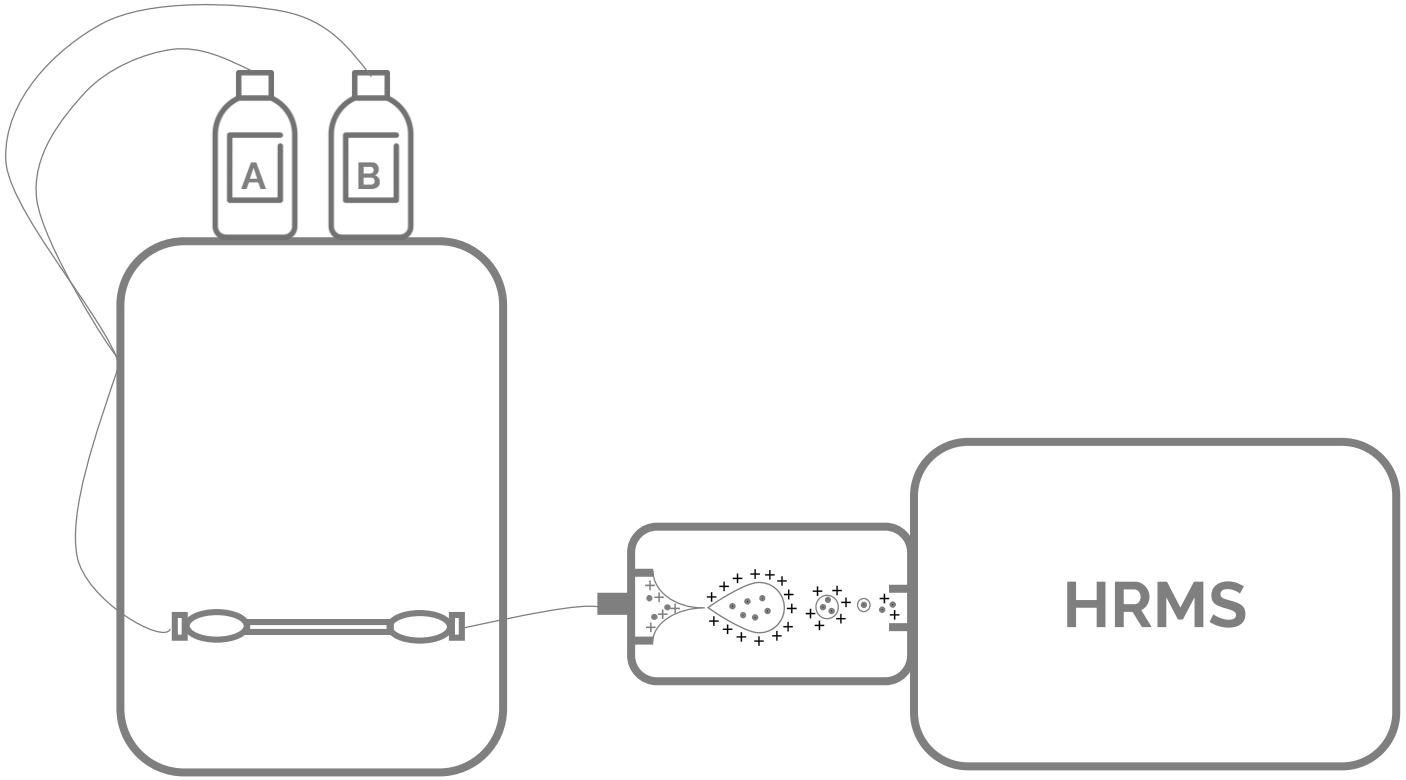


AIMS



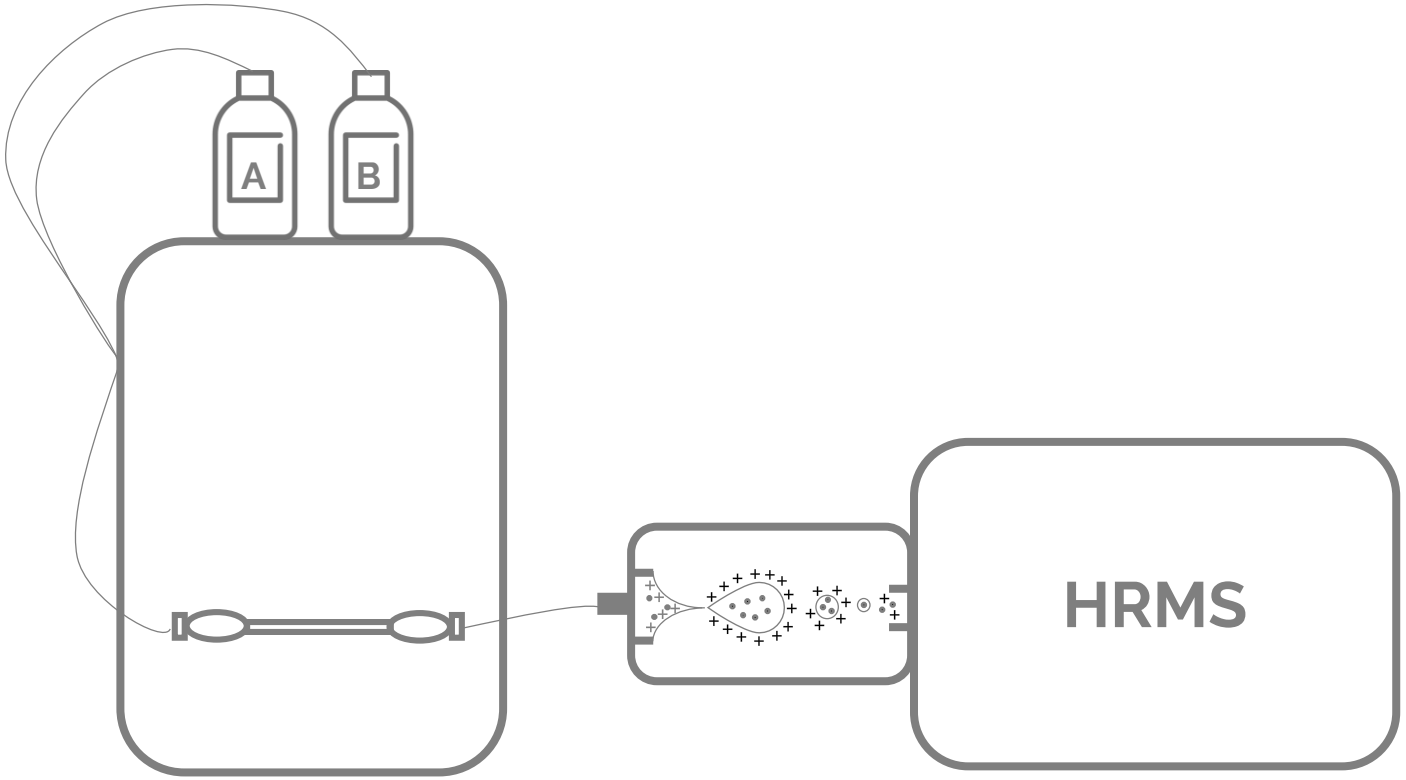
Column?

AIMS



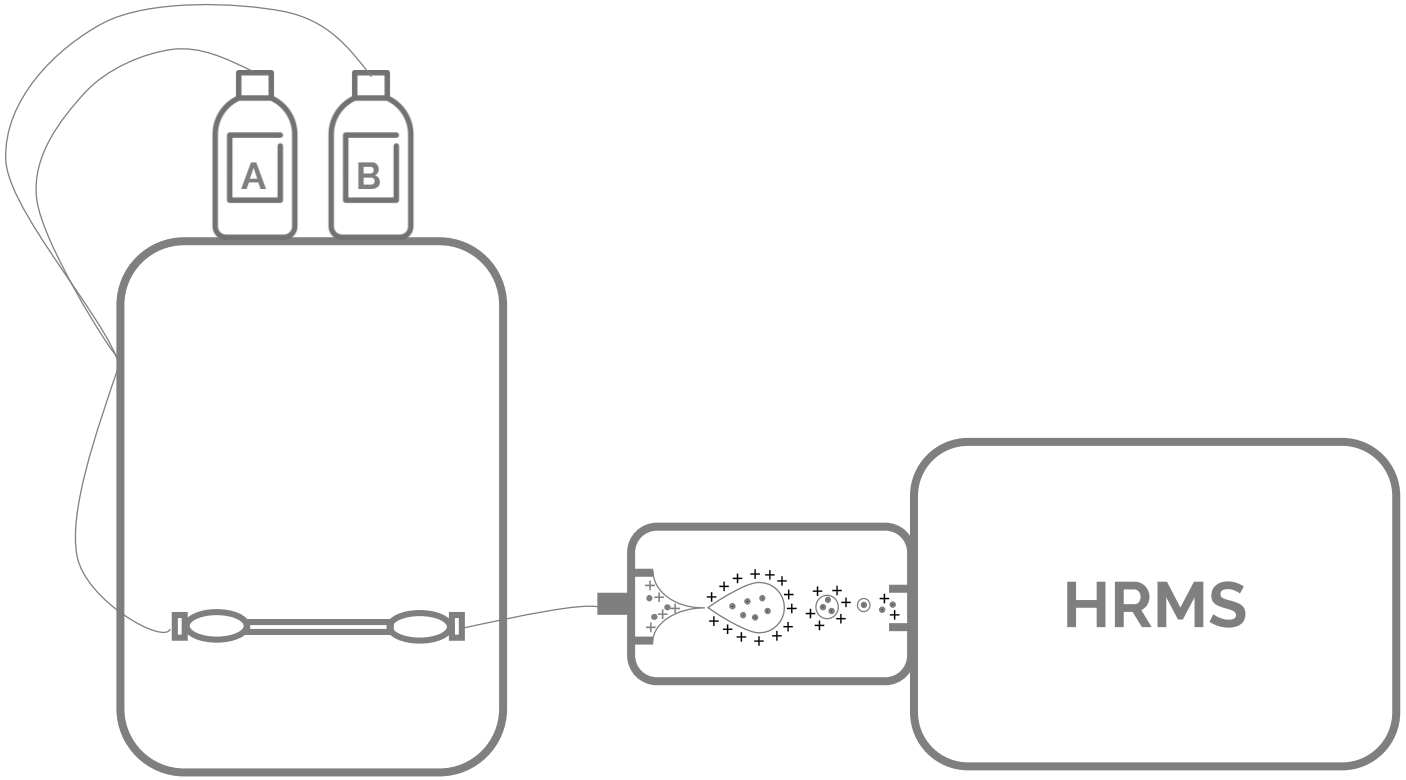
Column?
Organic modifier?

AIMS



Column?
Organic modifier?
pH?

AIMS



Column?
Organic modifier?
pH?
Additive?

EXPERIMENTAL

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NORMAN compounds in MassBank(S1)

(1295 compounds in positive ESI)

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1218 PaDEL descriptors + log*P* + number of acidic and basic functional groups (rcdk)

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Hierarchical clustering

EXPERIMENTAL

NORMAN compounds in MassBank(S1)
(1295 compounds in positive ESI)

↓

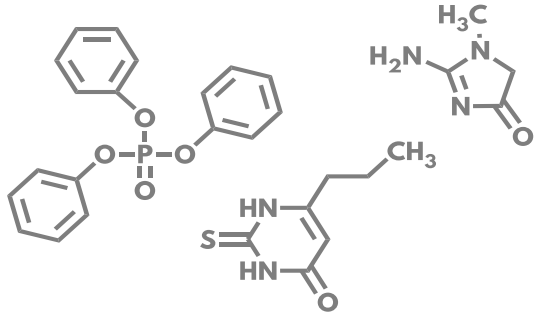
1218 PaDEL descriptors + log*P* + number of acidic and basic functional groups (rcdk)

↓

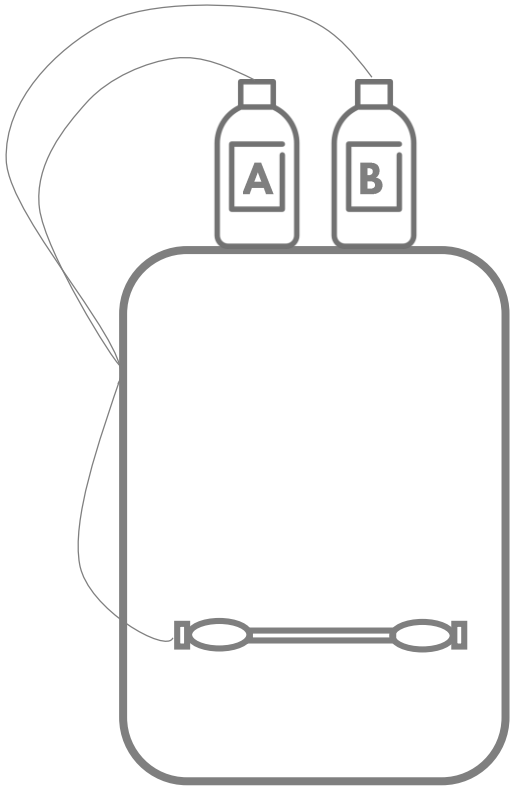
Hierarchical clustering

↓

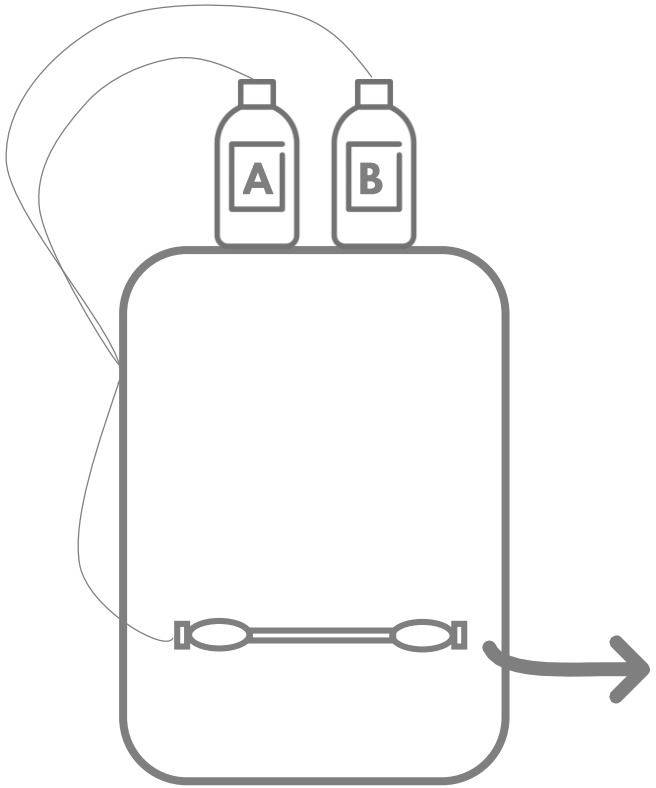
102 Selected compounds



EXPERIMENTAL

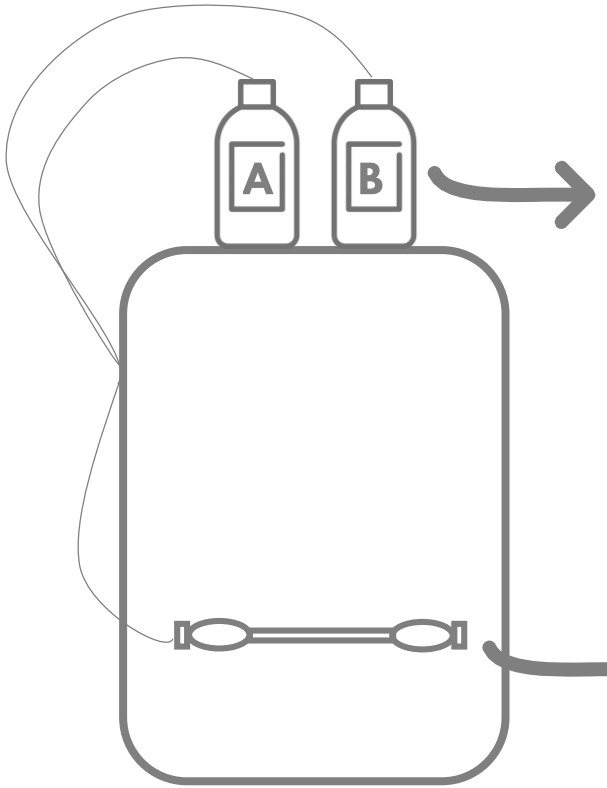


EXPERIMENTAL



Columns:
C18 reversed phase
Mixed mode
Biphenyl
HILIC

EXPERIMENTAL



Organic modifiers:
Acetonitrile
Methanol

Columns:
C18 reversed phase
Mixed mode
Biphenyl
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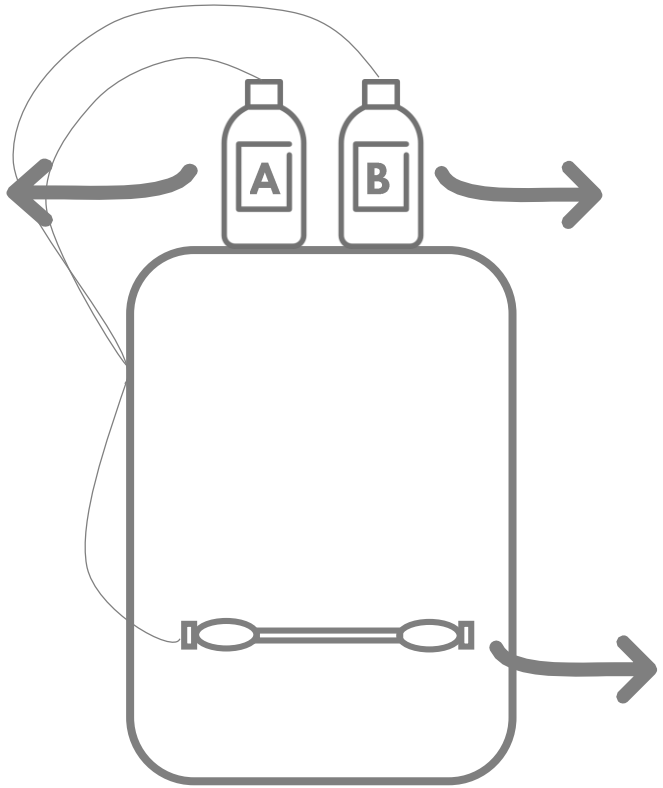
EXPERIMENTAL

Water phases:



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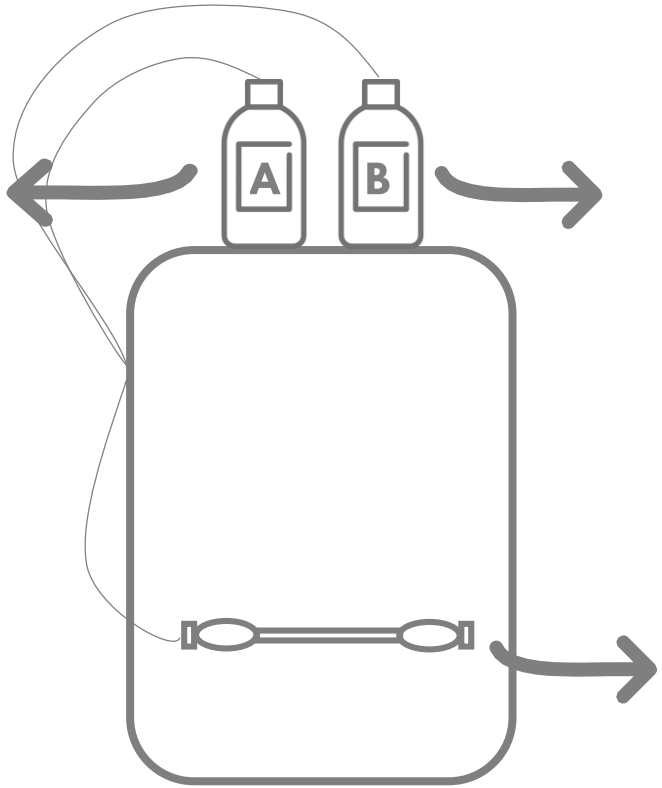
EXPERIMENTAL

Water phases:

pH



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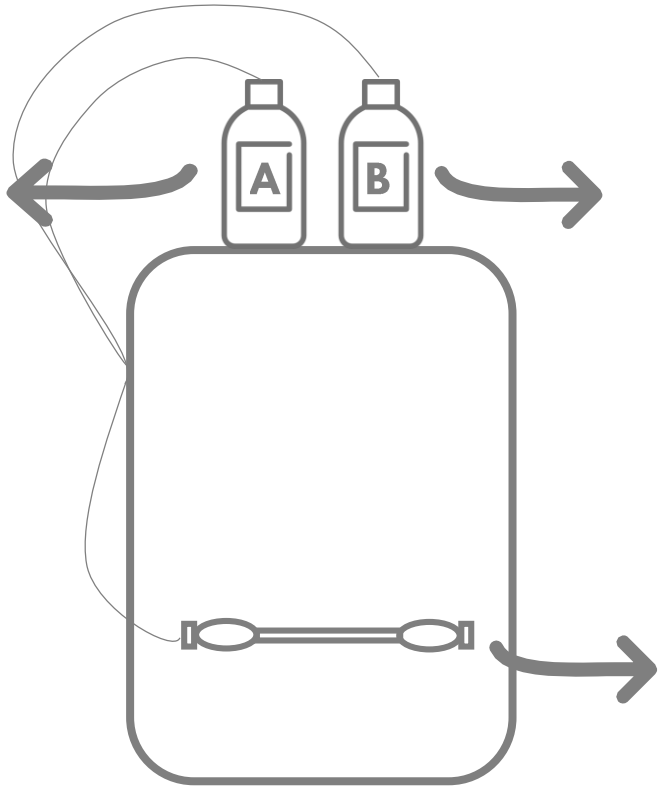
Water phases:

~2.1 0.1% TFA
~2.7 0.1% Formic acid
~2.9 0.1% Acetic acid

pH



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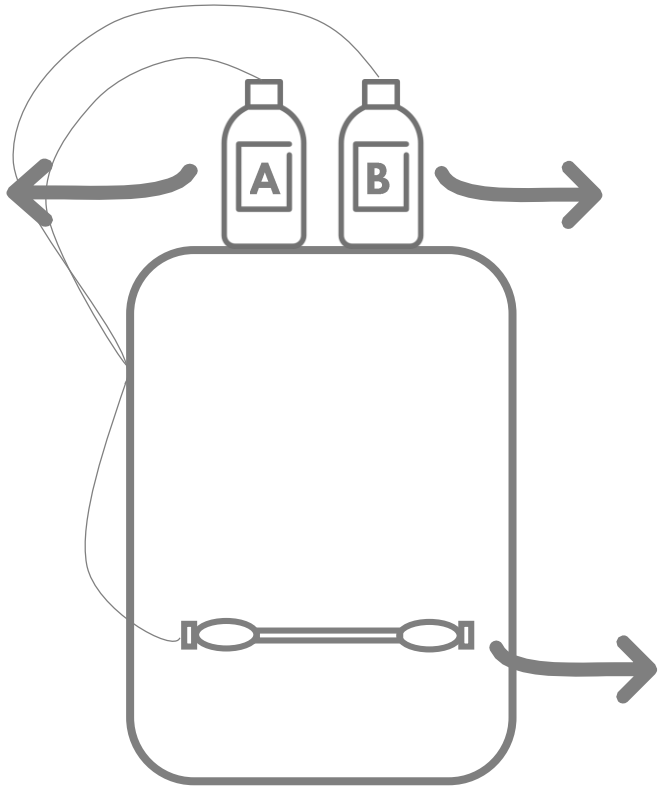

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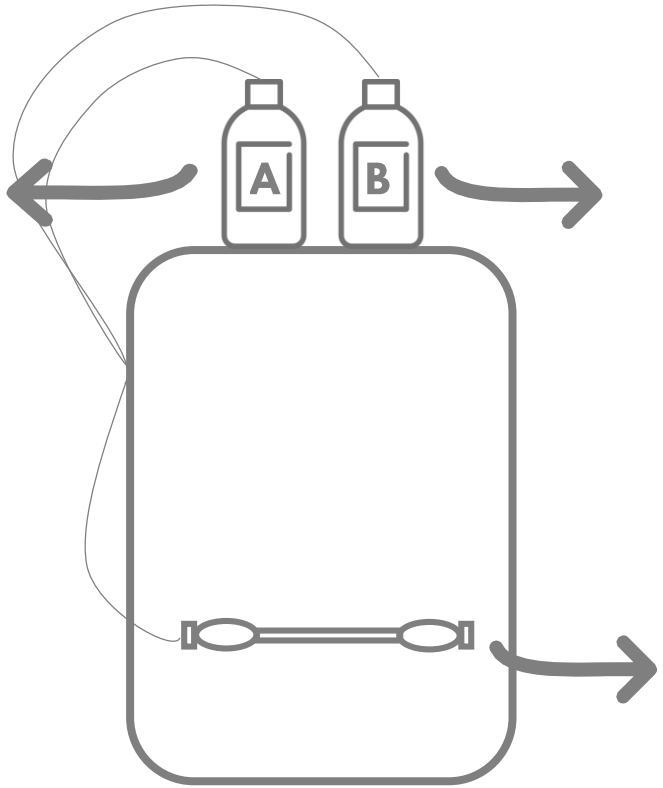
3.2 5 mM Ammonium formate
3.5 5 mM Ammonium acetate

pH



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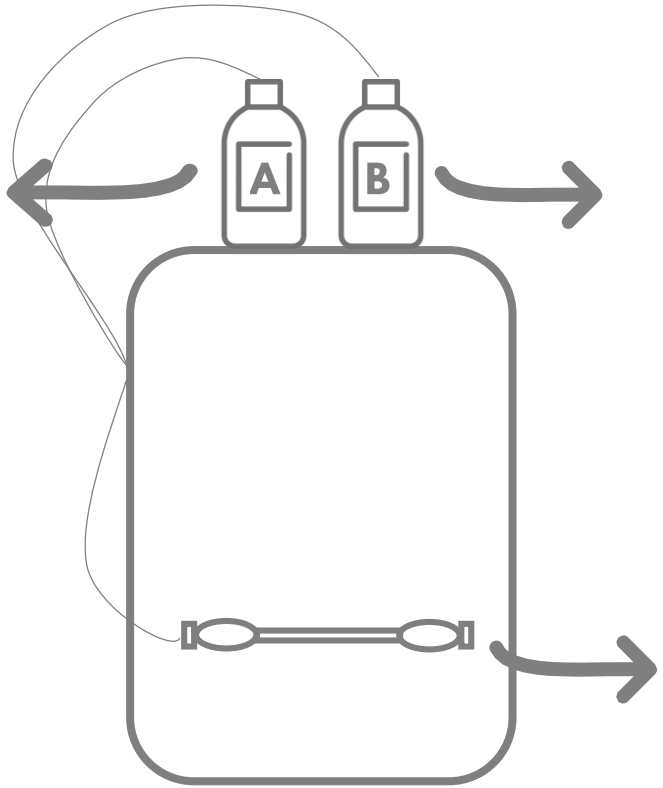

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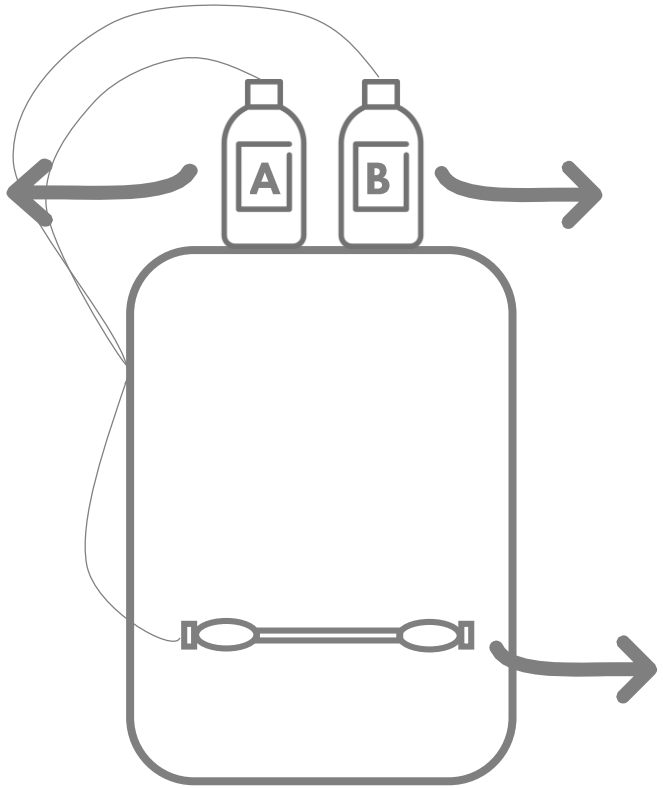
3.2	5 mM Ammonium formate
3.5	5 mM Ammonium acetate
5.0	5 mM Ammonium formate/acetate

pH



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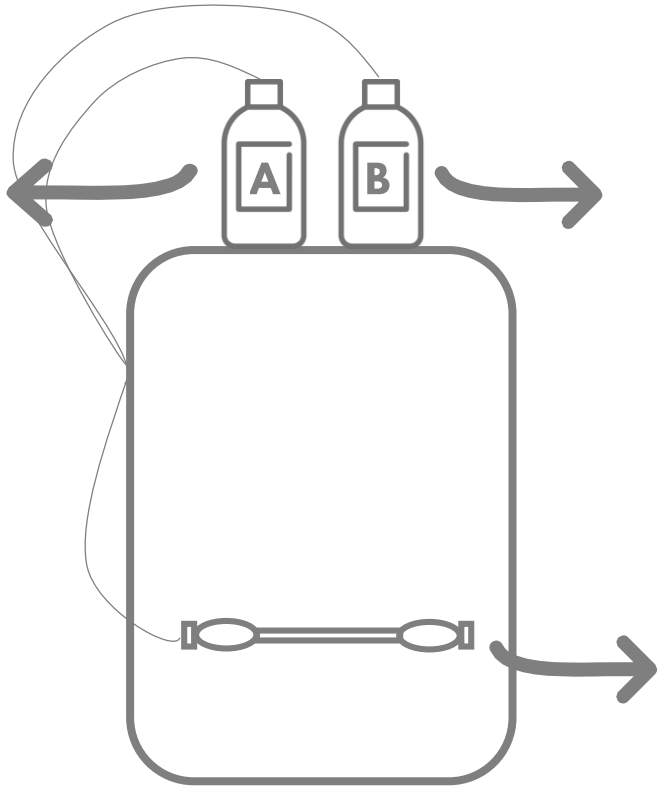

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3.2 5 mM Ammonium formate
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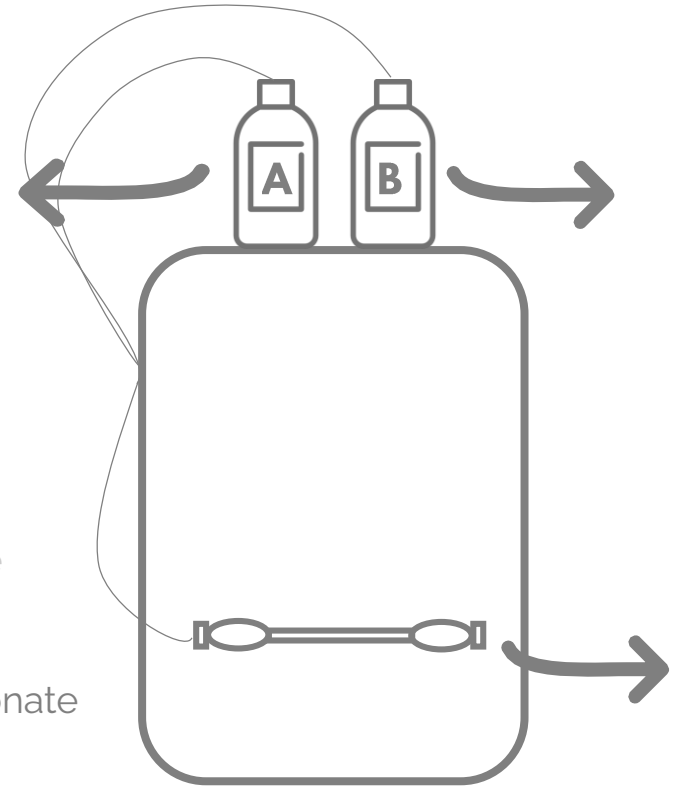
5.0 5 mM Ammonium formate/acetate

8.0 5 mM Ammonium formate/bicarbonate

pH



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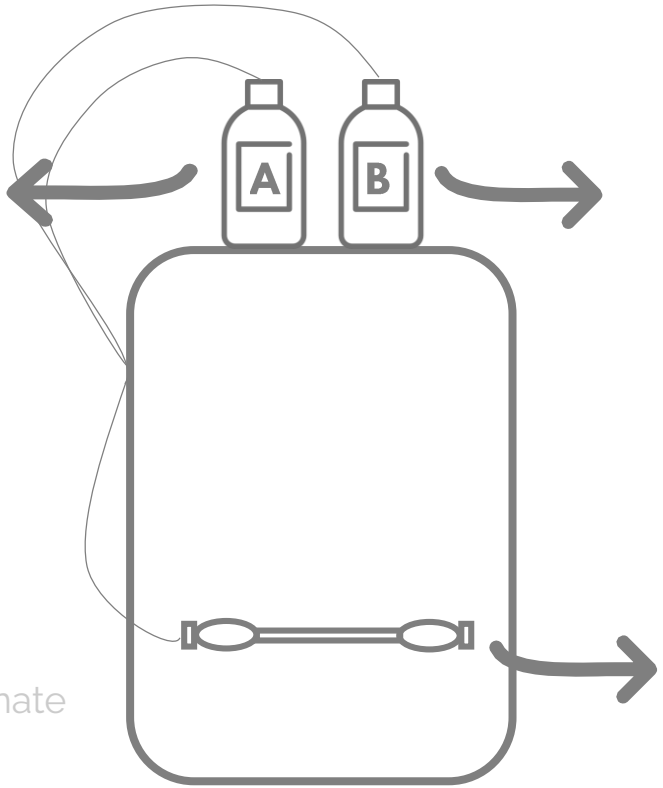

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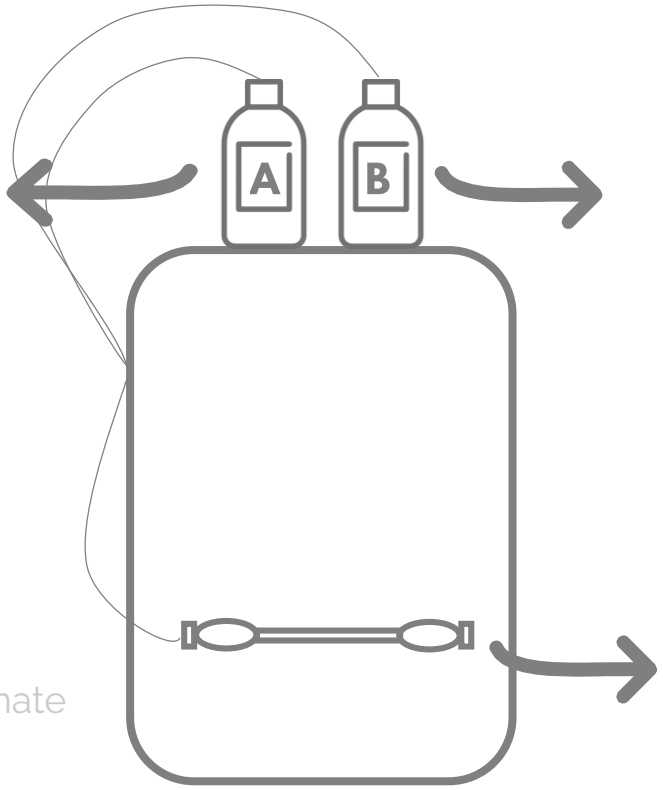
10.0	0.1% Ammonia

pH



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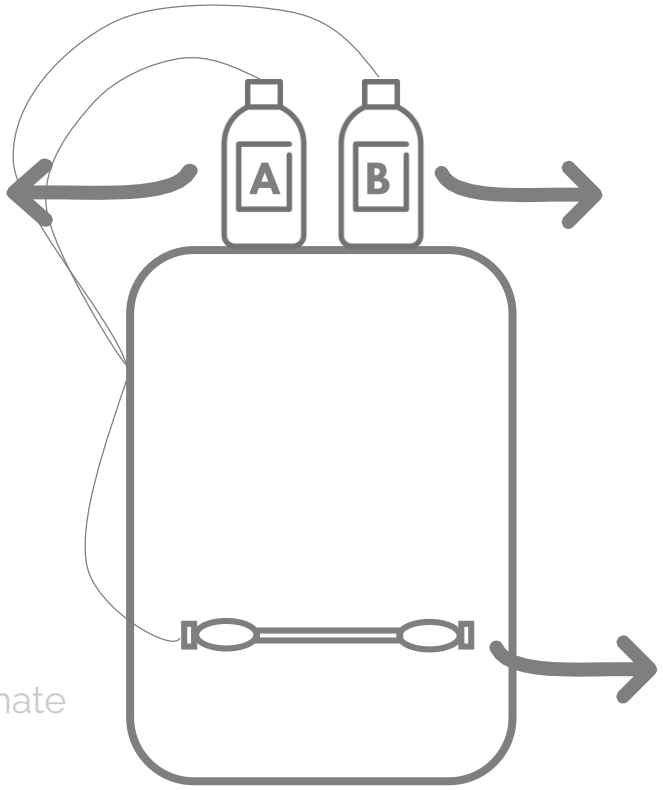
10.0	0.1% Ammonia



Organic modifiers:
Acetonitrile
Methanol

Columns:
C18 reversed phase
Mixed mode
Biphenyl
HILIC

Acquisition: ESI+

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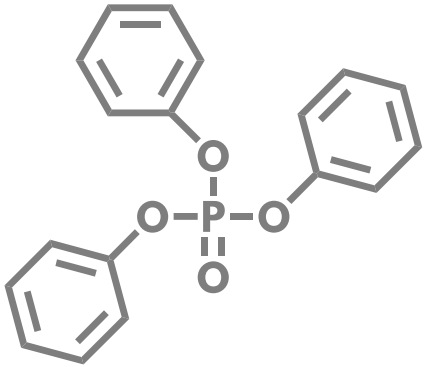
EXPERIMENTAL

Classification of compounds

EXPERIMENTAL

Classification of compounds

Neutral



Triphenylphosphate

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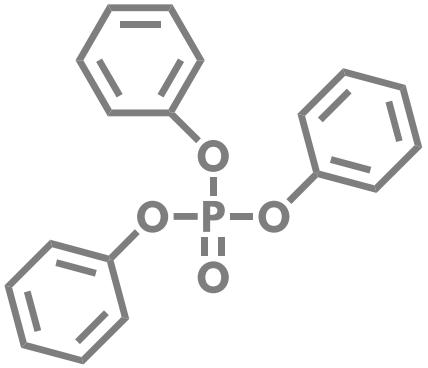
EXPERIMENTAL

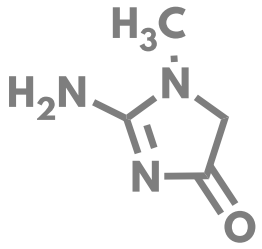
Classification of compounds

Neutral



Triphenylphosphate

Acids and bases

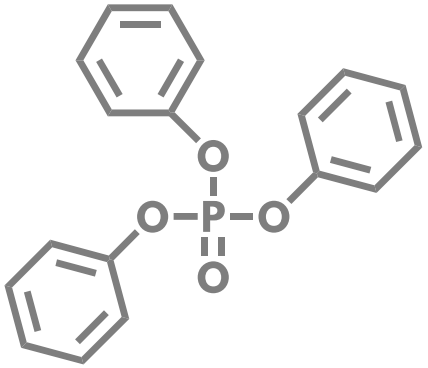


Creatinine

EXPERIMENTAL

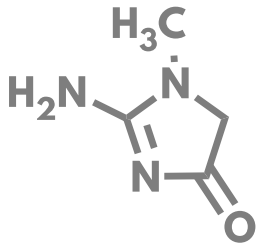
Classification of compounds

Neutral



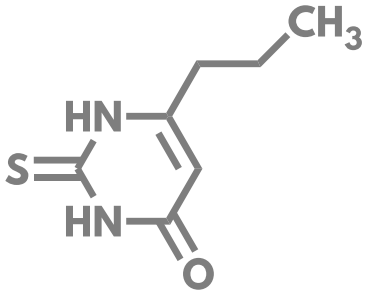
Triphenylphosphate

Acids and bases



Creatinine

Weak acids

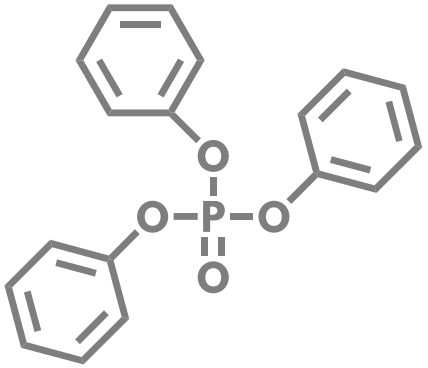


Propylthiouracil

EXPERIMENTAL

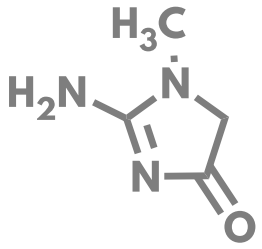
Classification of compounds

Neutral



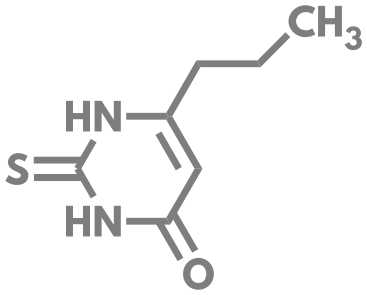
Triphenylphosphate

Acids and bases



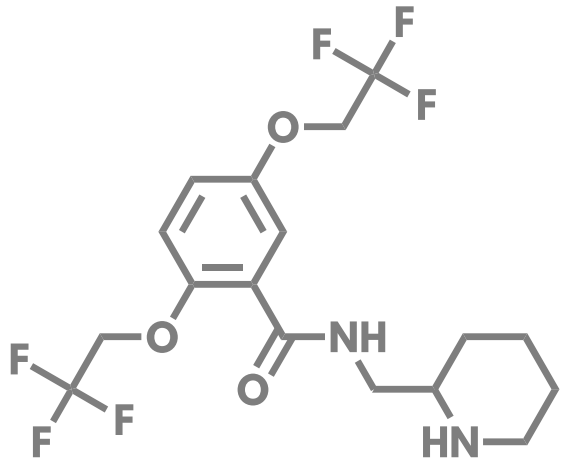
Creatinine

Weak acids



Propylthiouracil

Weak bases



Flecainide

EXPERIMENTAL

Comparison of response factor

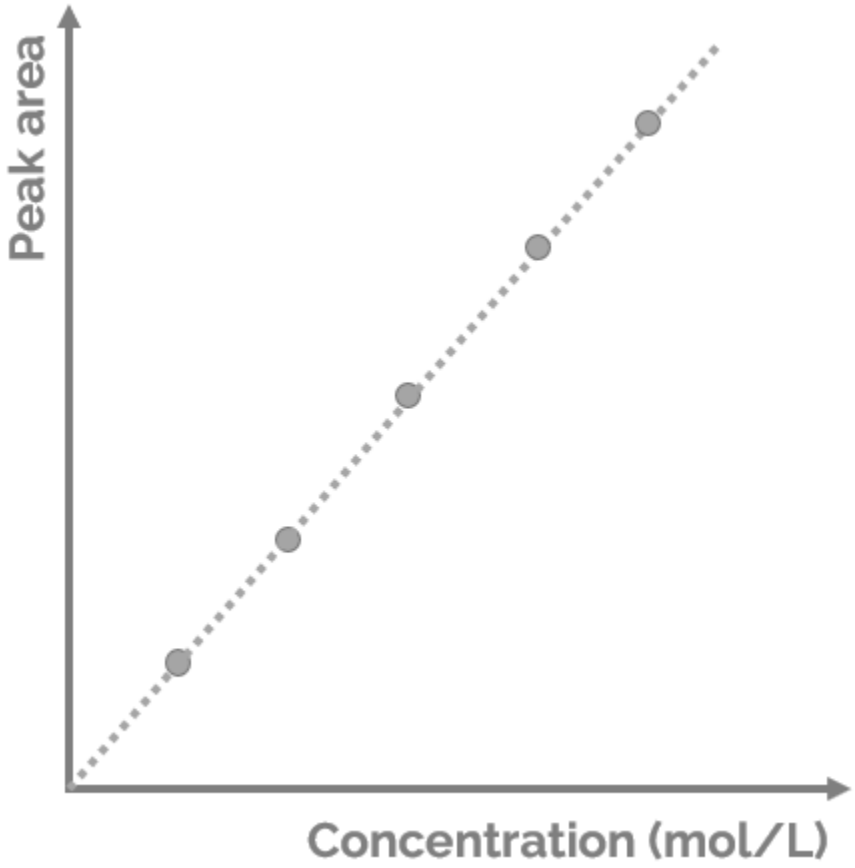
EXPERIMENTAL

Comparison of response factor

Response factor (RF) = **Peak area / Concentration**

EXPERIMENTAL

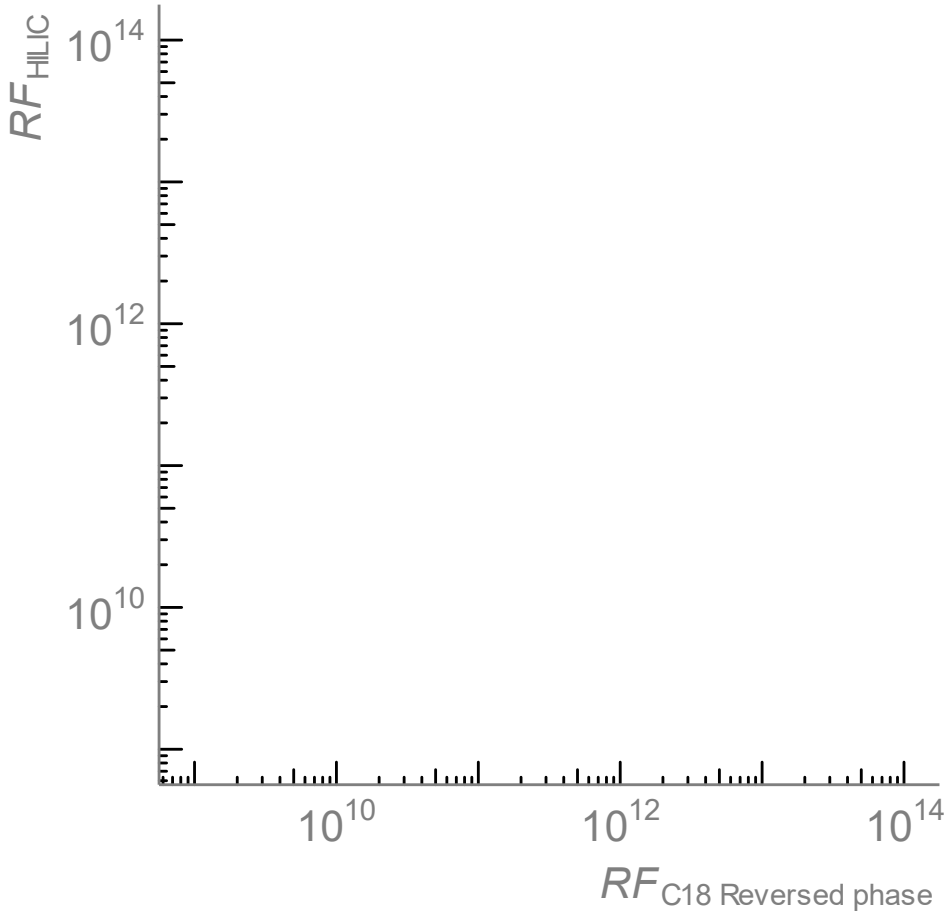
Comparison of response factor



Response factor (RF) = **Peak area / Concentration**

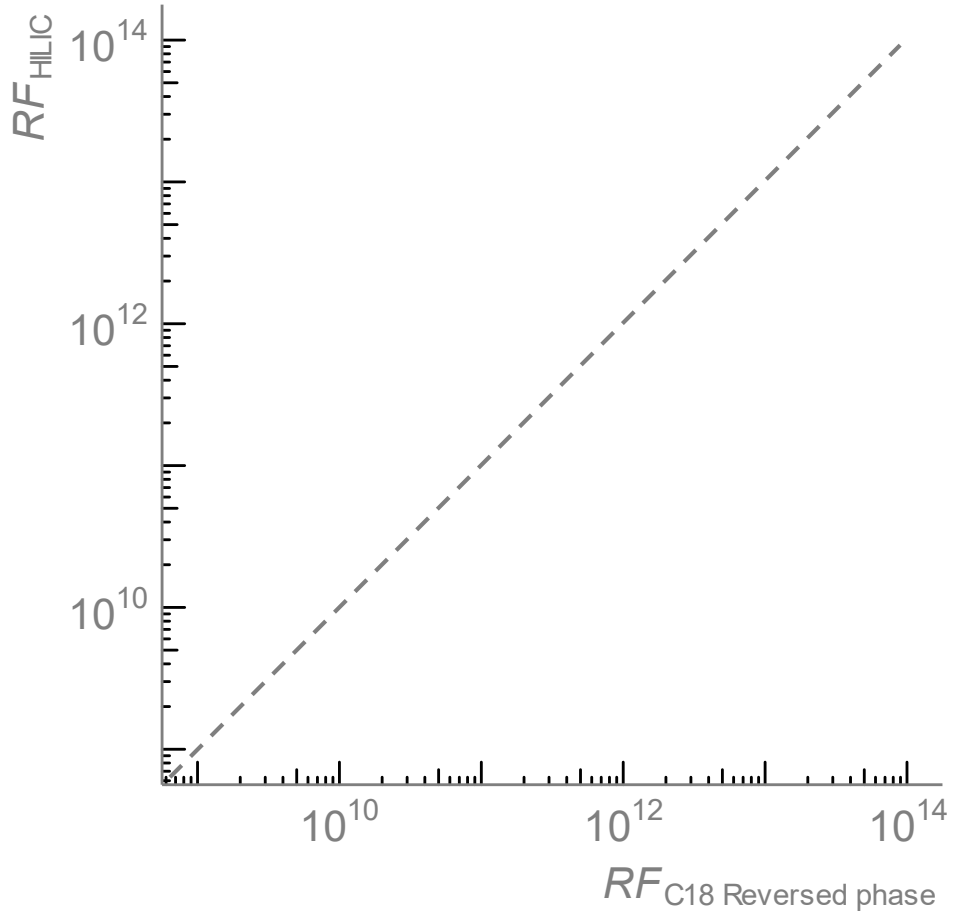
RESULTS

Column



RESULTS

Column



RESULTS

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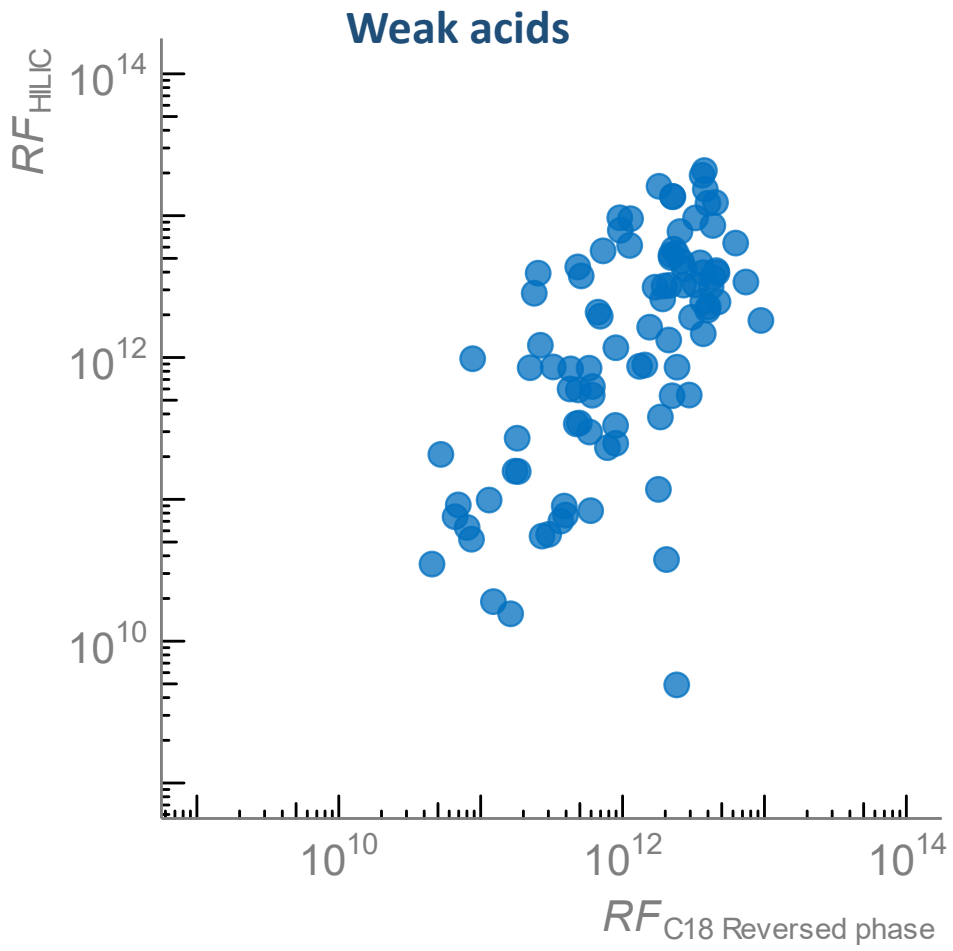

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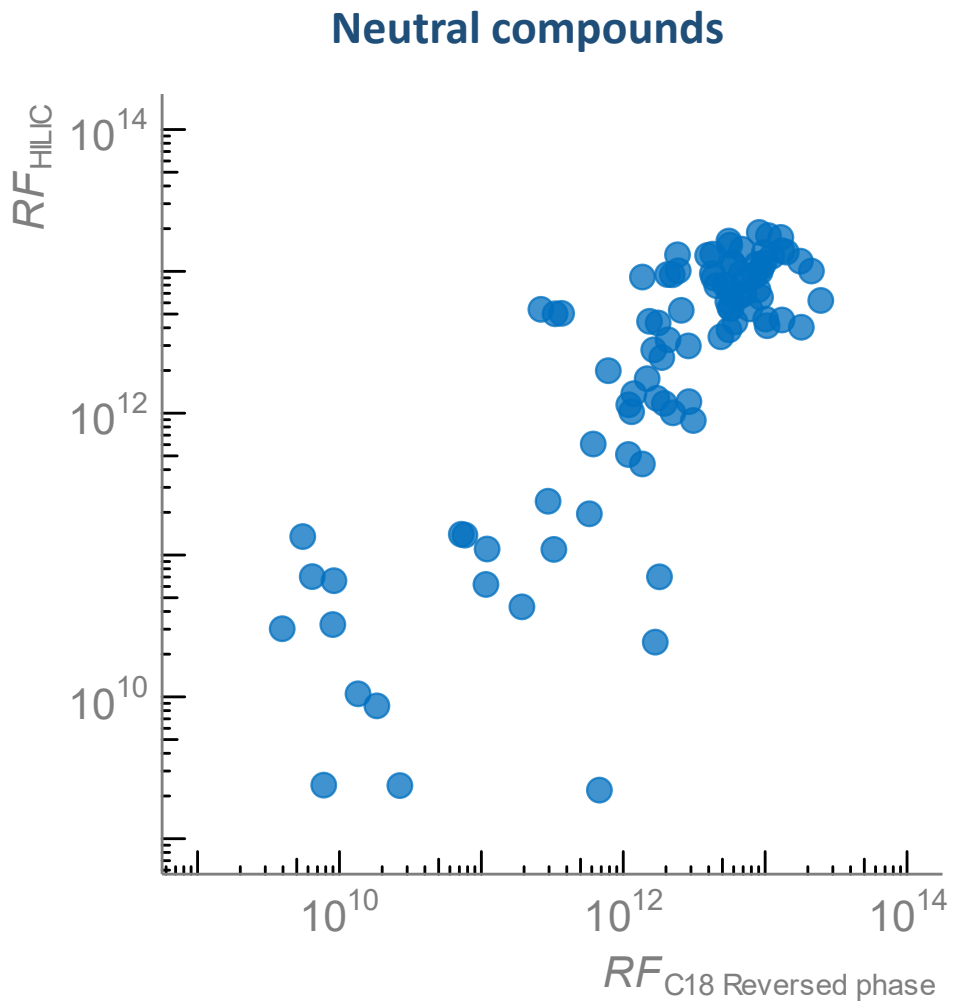

RESULTS

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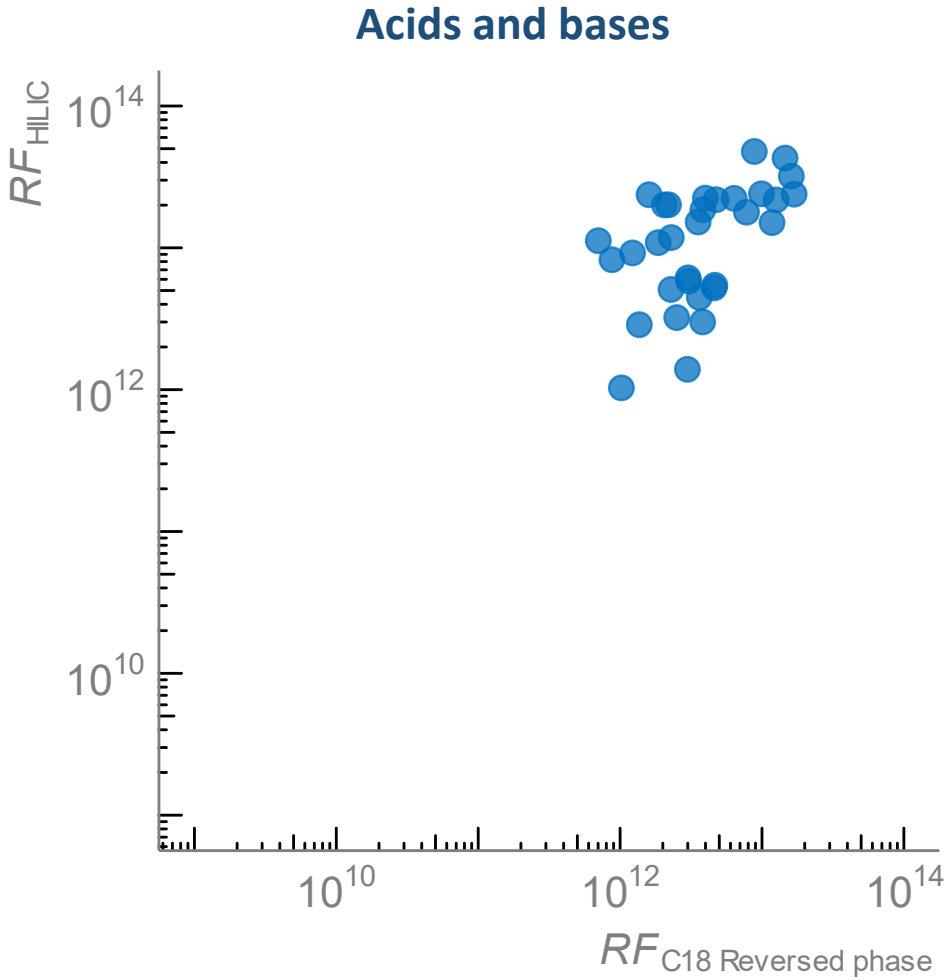

RESULTS

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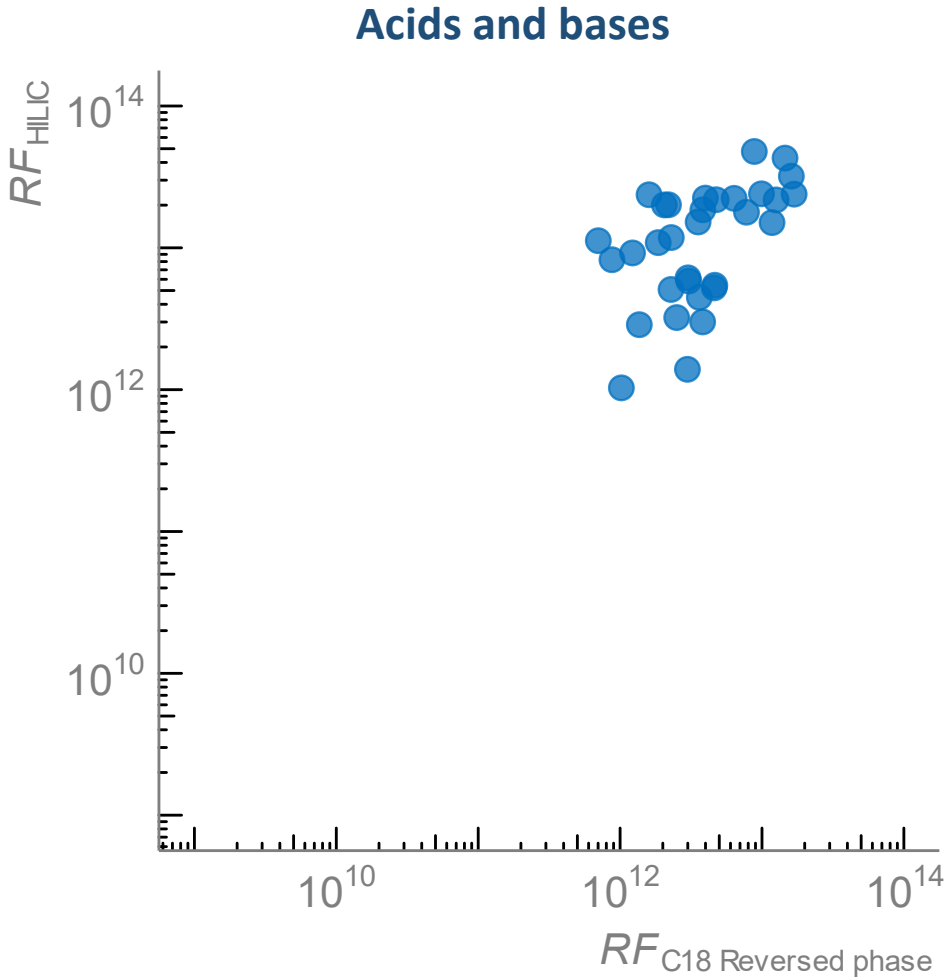
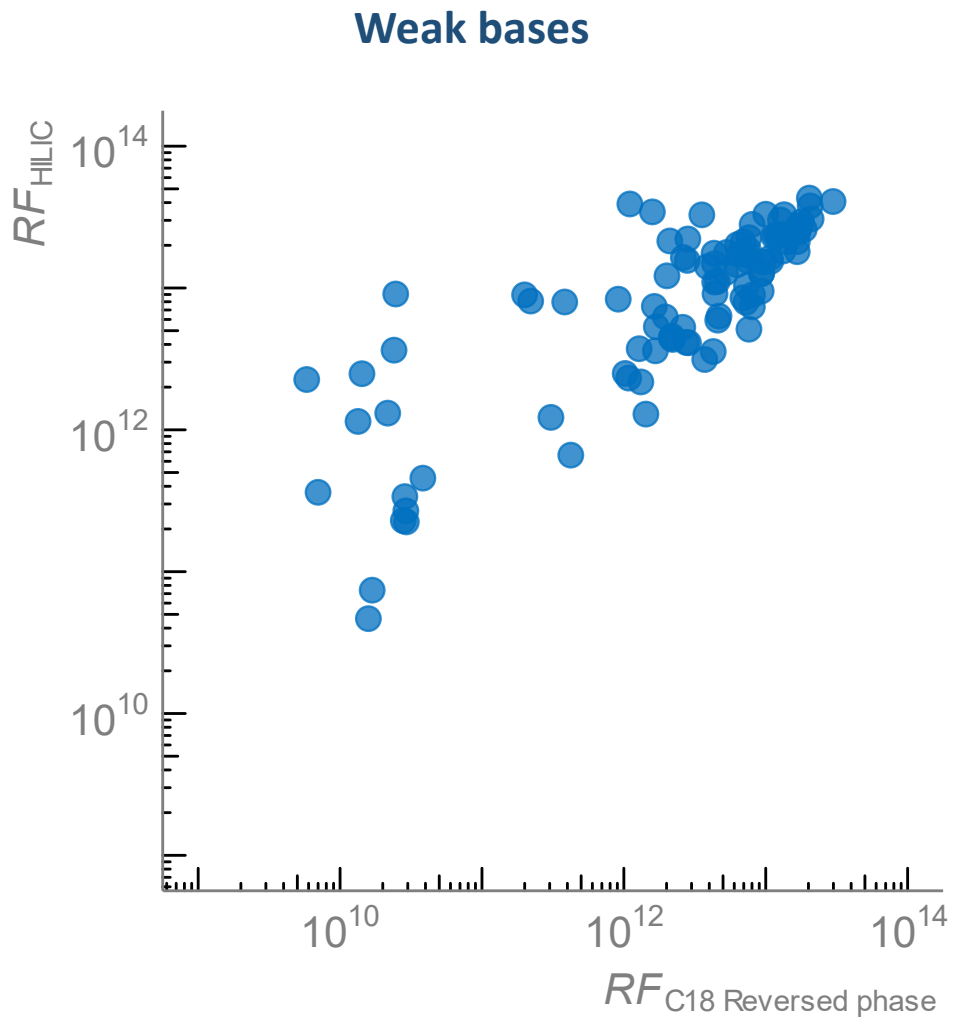


Compounds with acidic and basic groups are highly ionizable

RESULTS

Column



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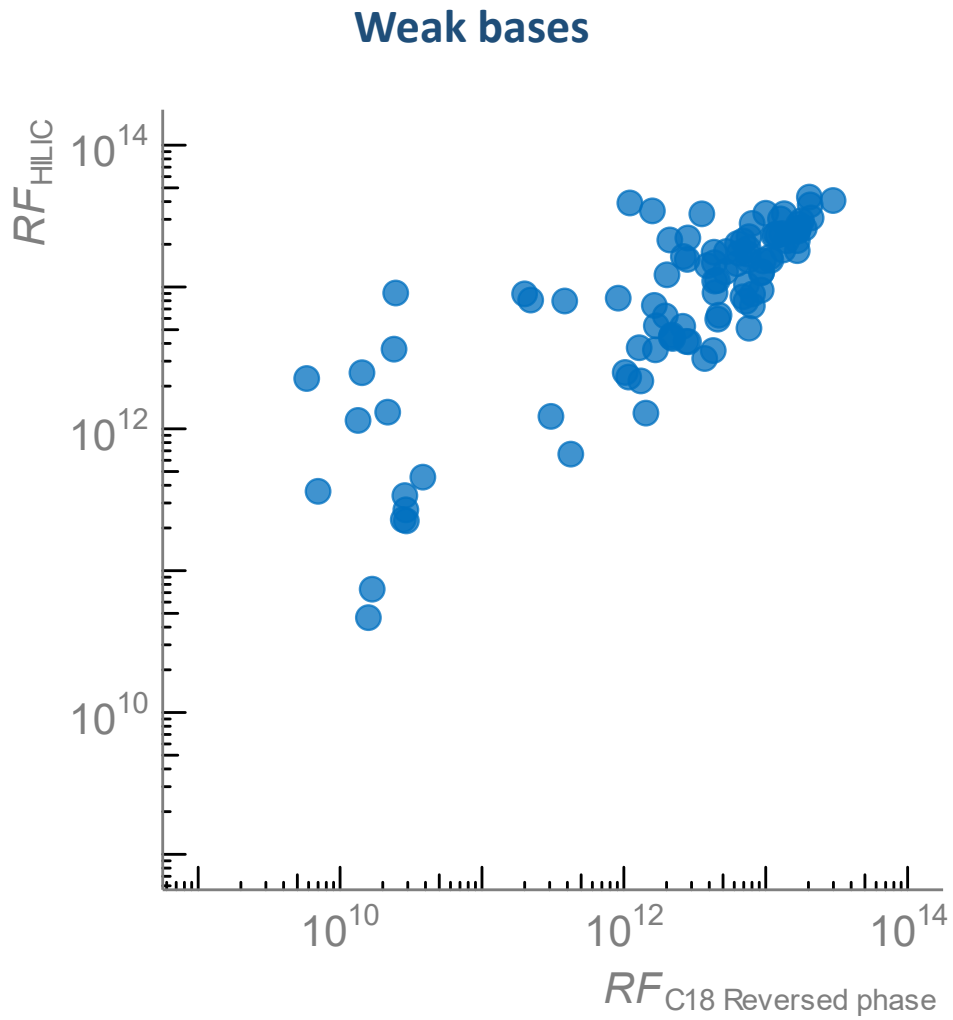
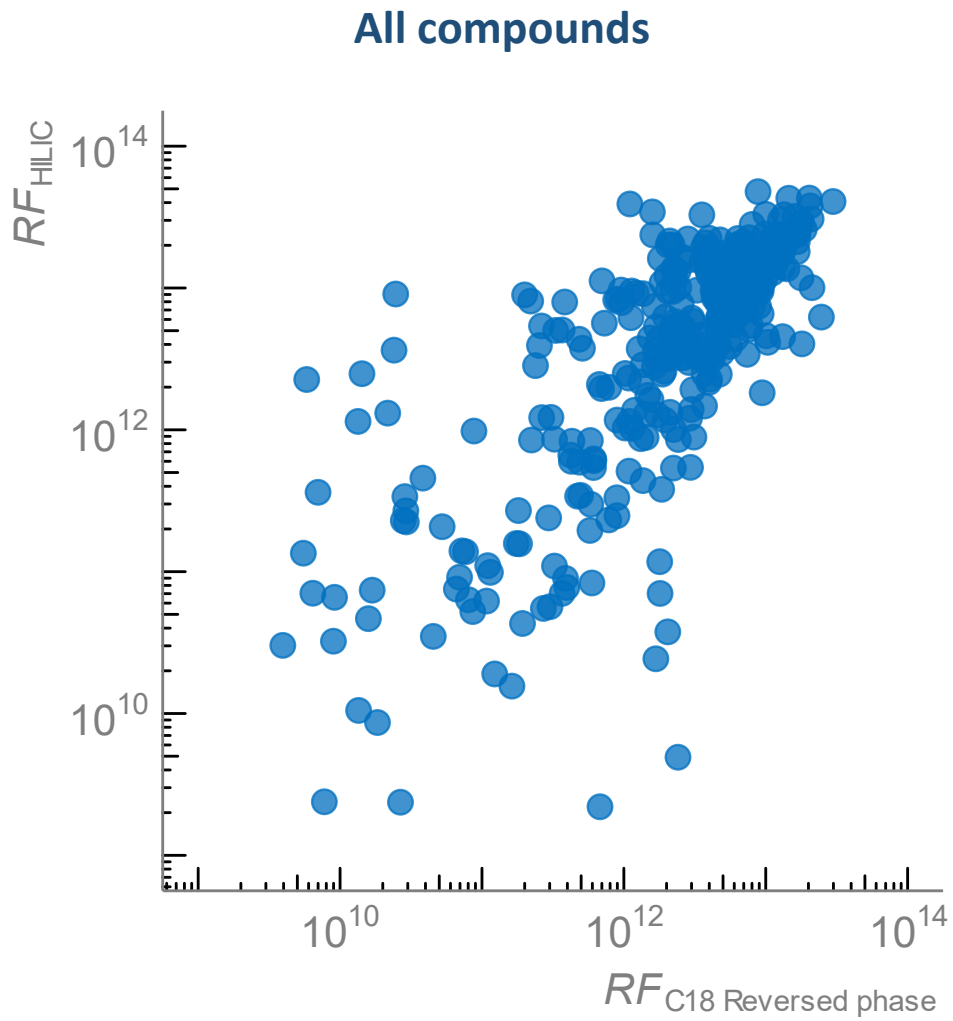


Higher RF-s in HILIC for some weak bases

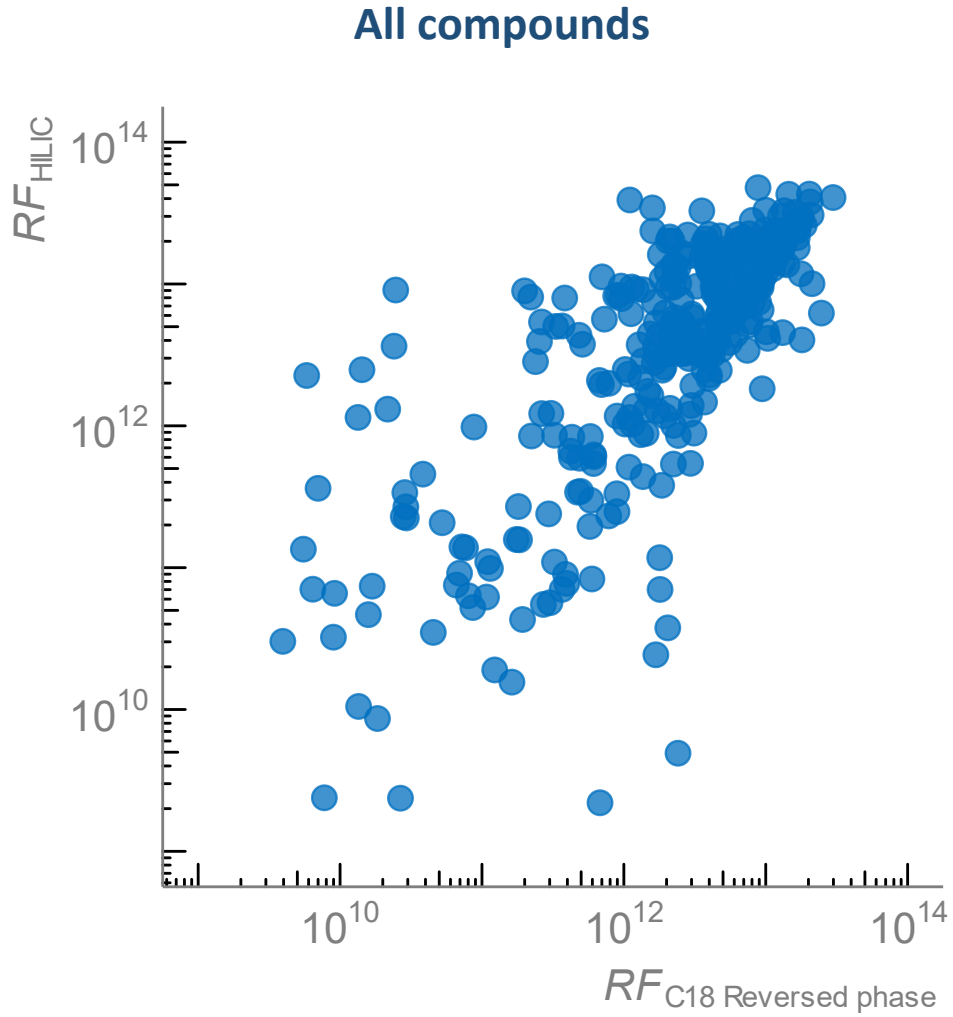
RESULTS

Column



RESULTS

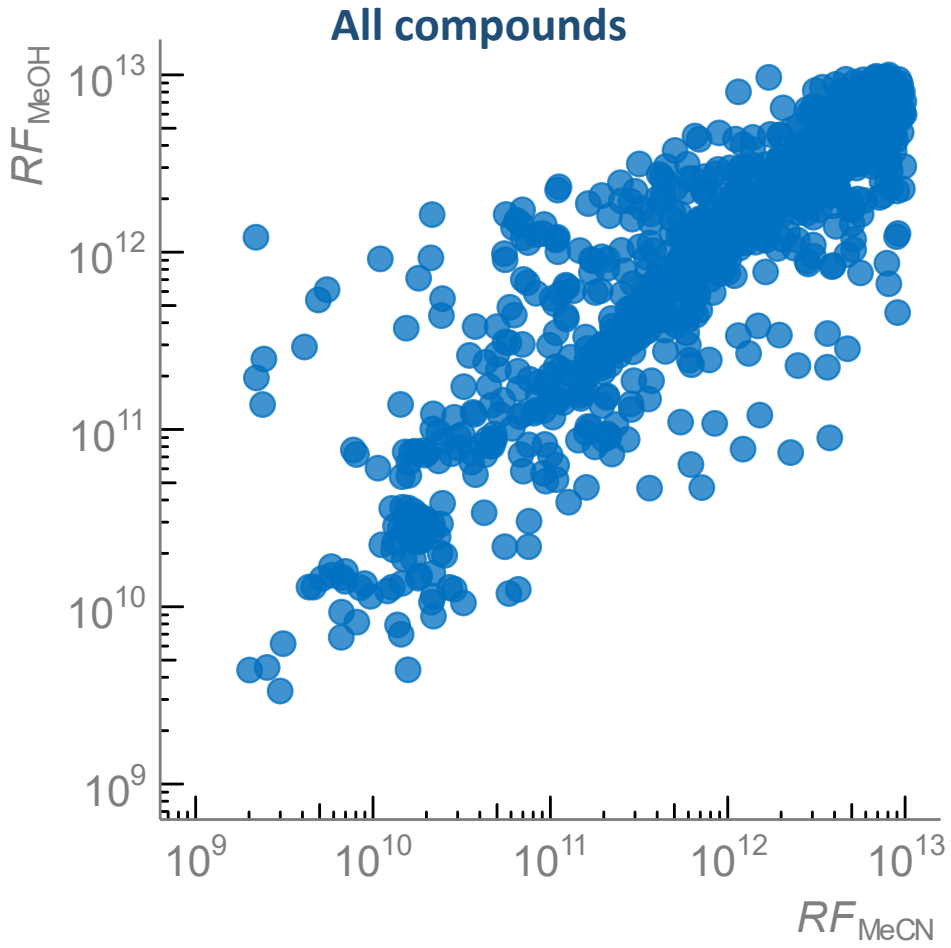
Column



Effect of column is not statistically significant in this case

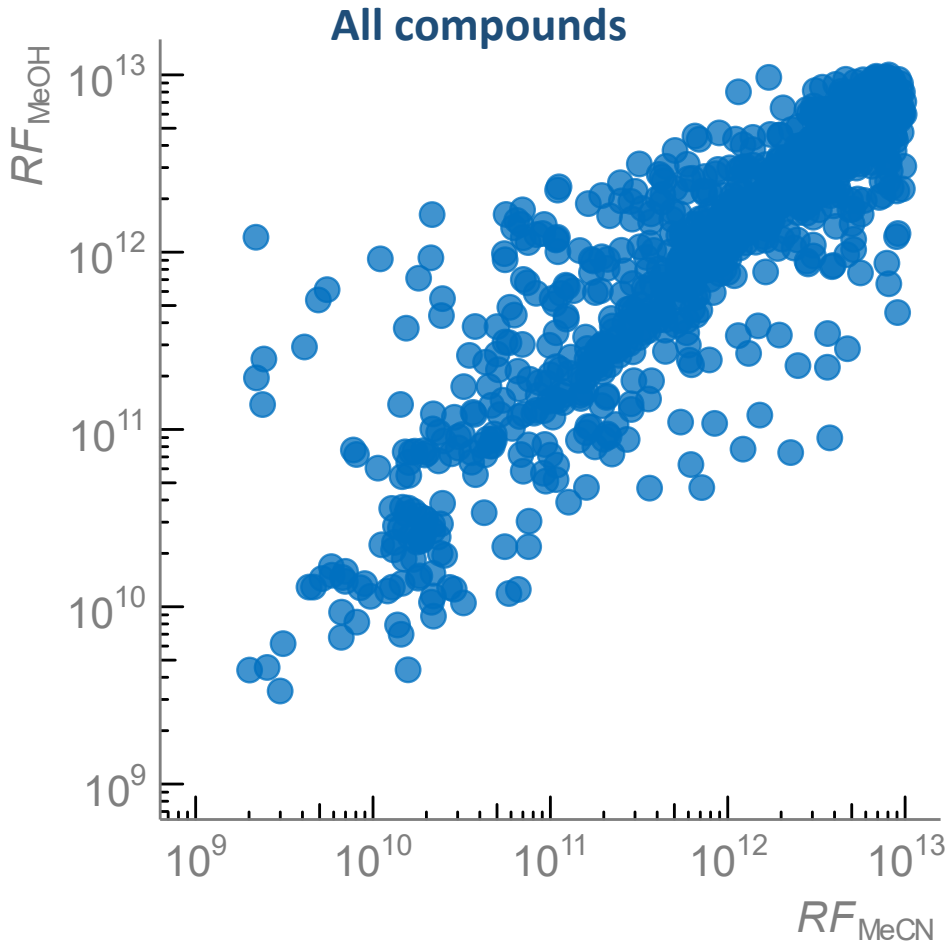
RESULTS

Organic modifier



RESULTS

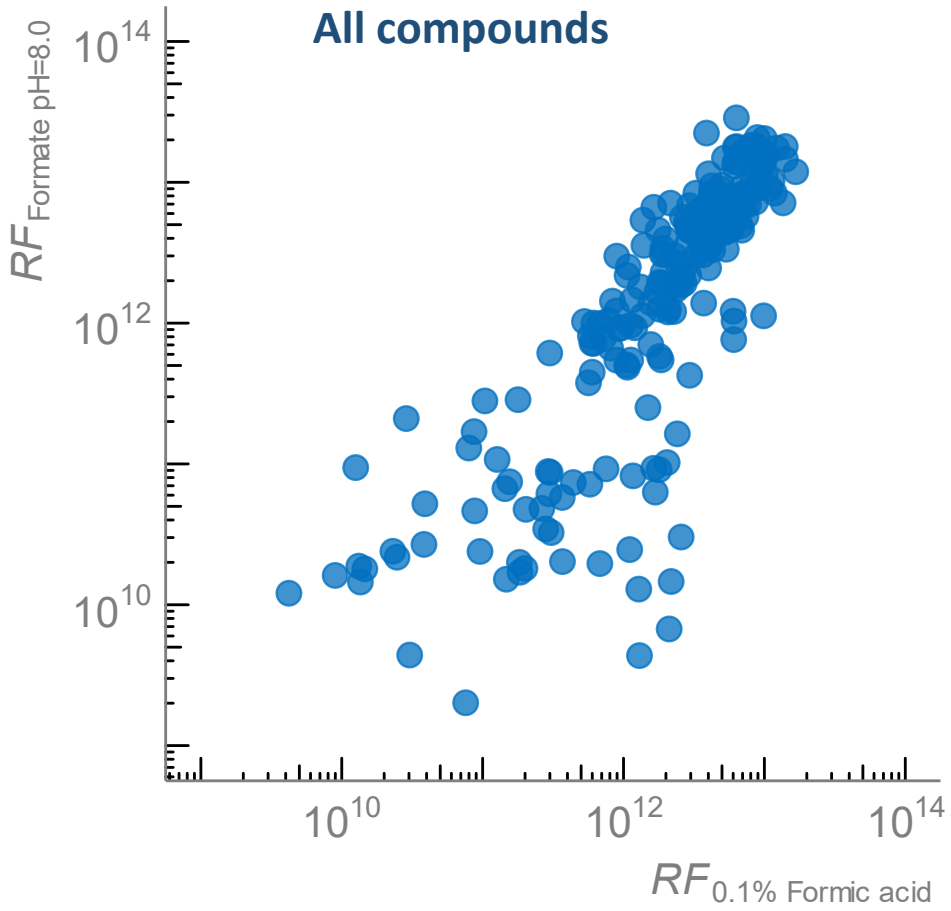
Organic modifier



The effect of organic modifier is not statistically significant

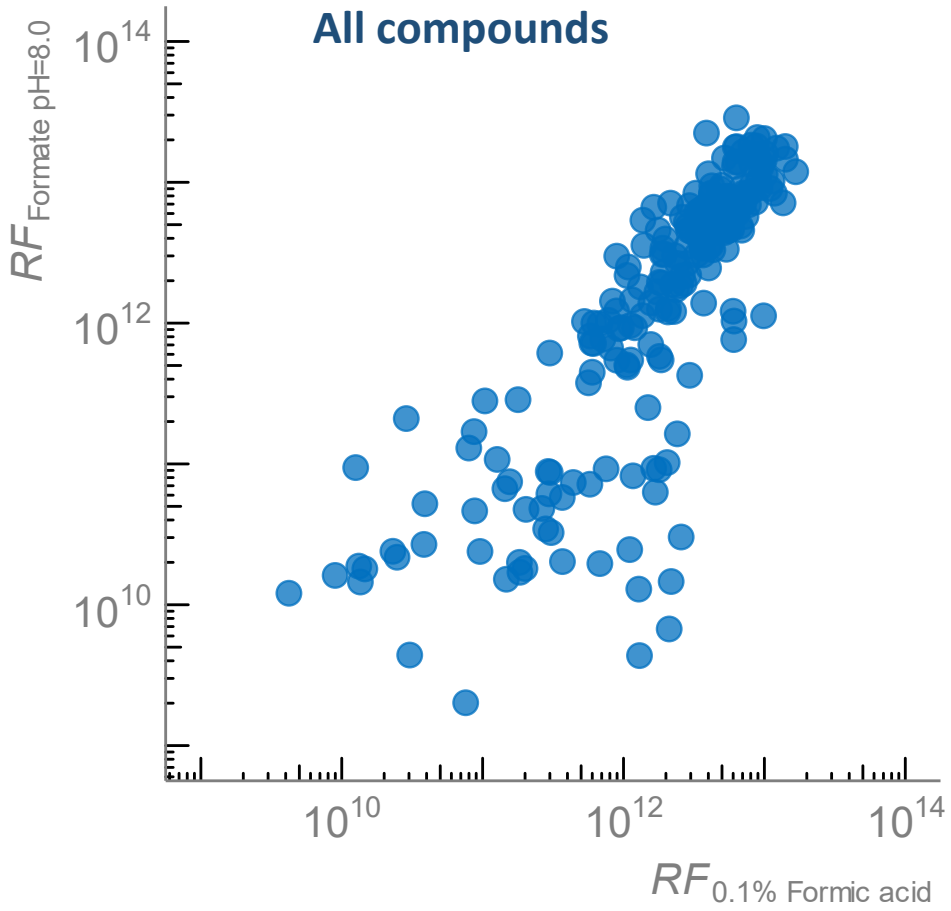
RESULTS

pH


All compounds
$RF_{Formate\ pH=8.0}$
10^{14}
10^{12}
10^{10}
10^{10}
10^{12}
10^{14}
$RF_{0.1\%\ Formic\ acid}$

RESULTS

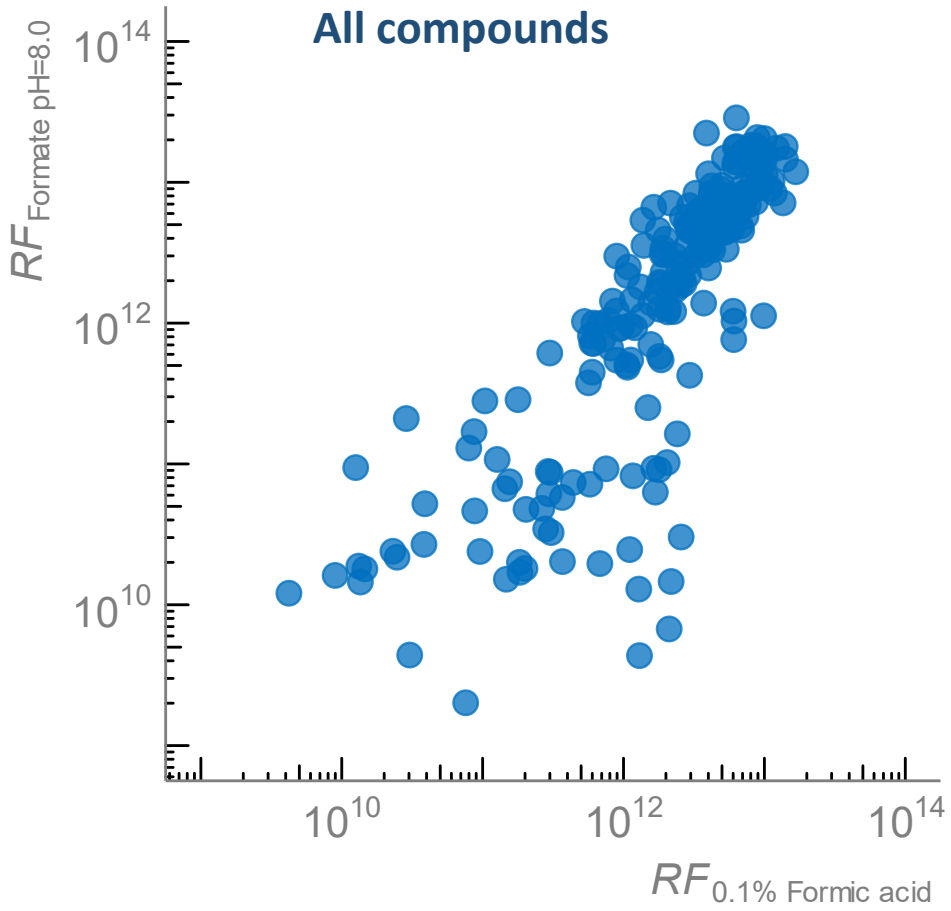
pH



pH has significant effect on low ionizable compounds :

RESULTS

pH

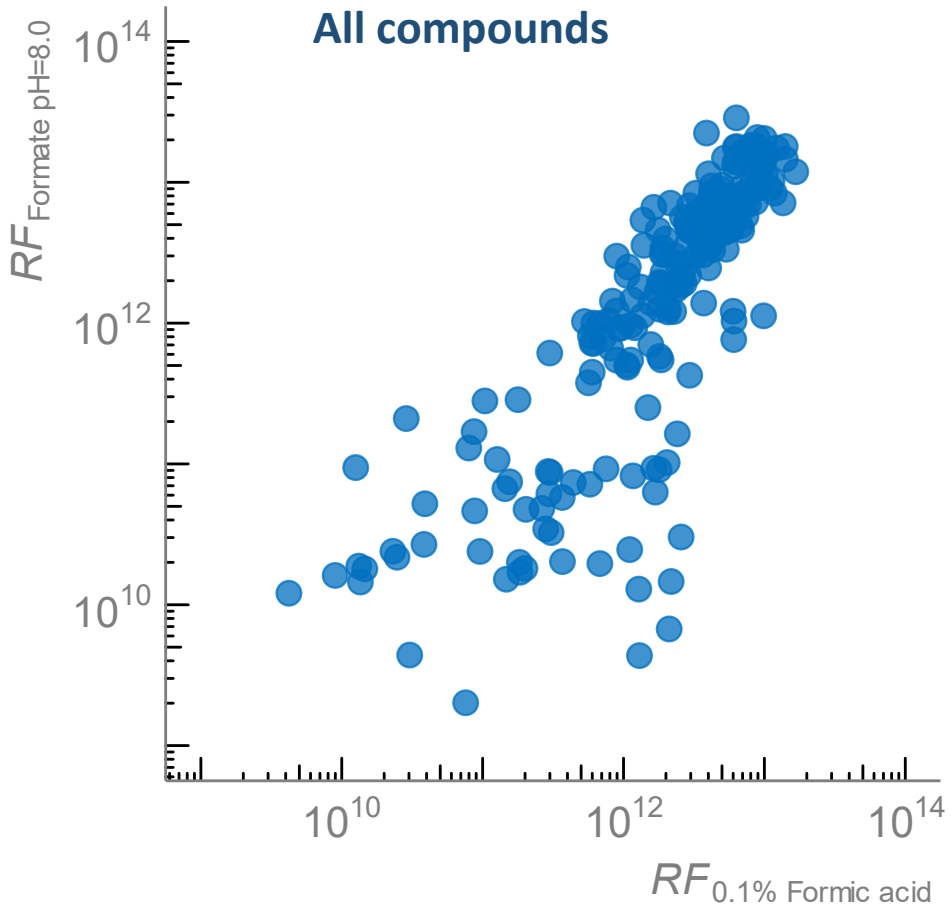


pH has significant effect on low ionizable compounds :

Weak acids

RESULTS

pH



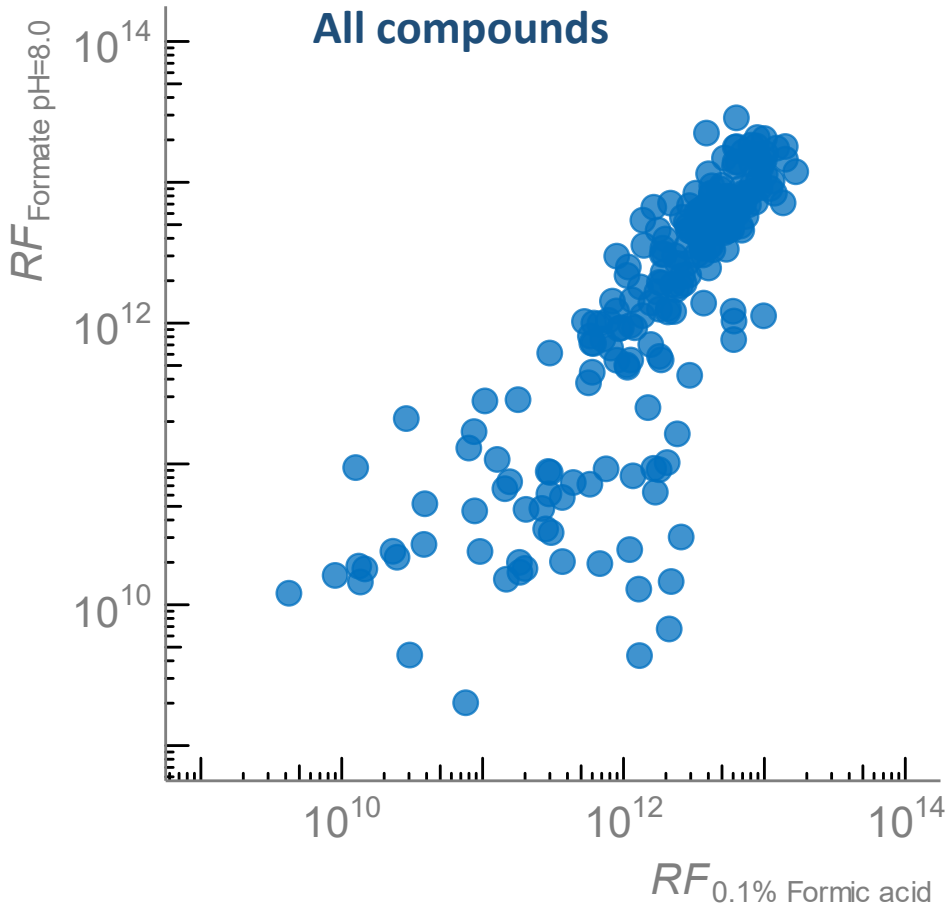
pH has significant effect on low ionizable compounds :

Weak acids

Weak bases

RESULTS

pH



pH has significant effect on low ionizable compounds :

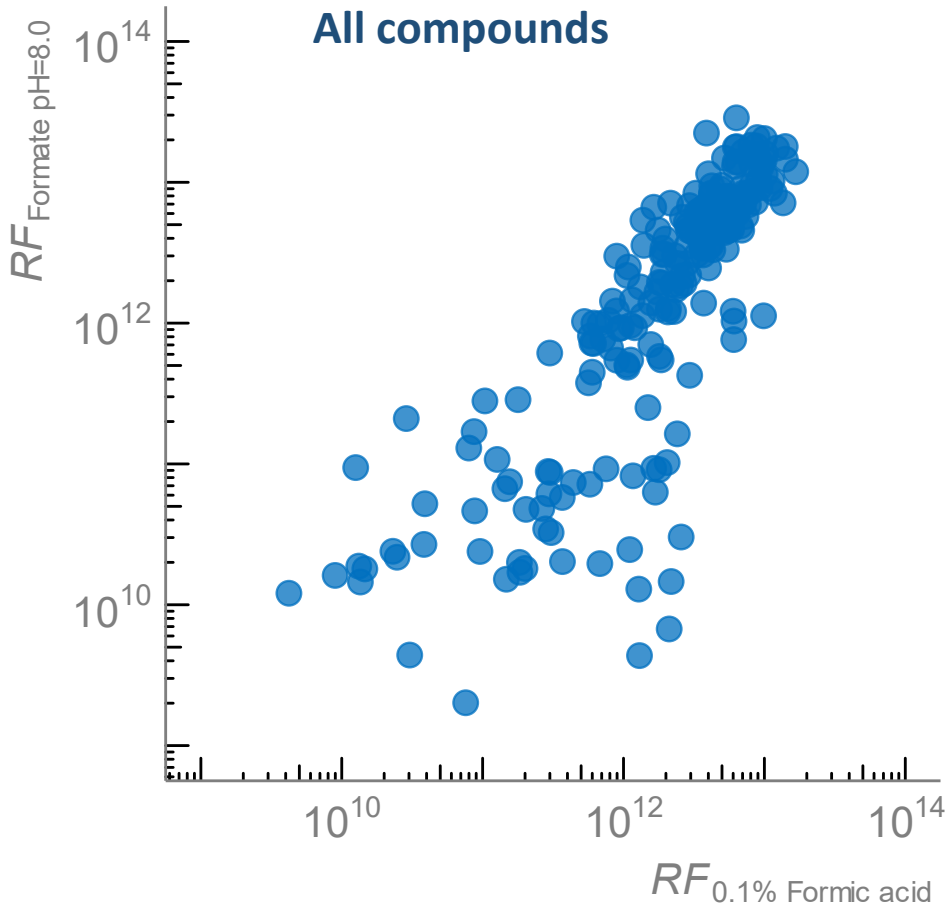
Weak acids

Weak bases

Neutral compounds

RESULTS

pH



pH has significant effect on low ionizable compounds :

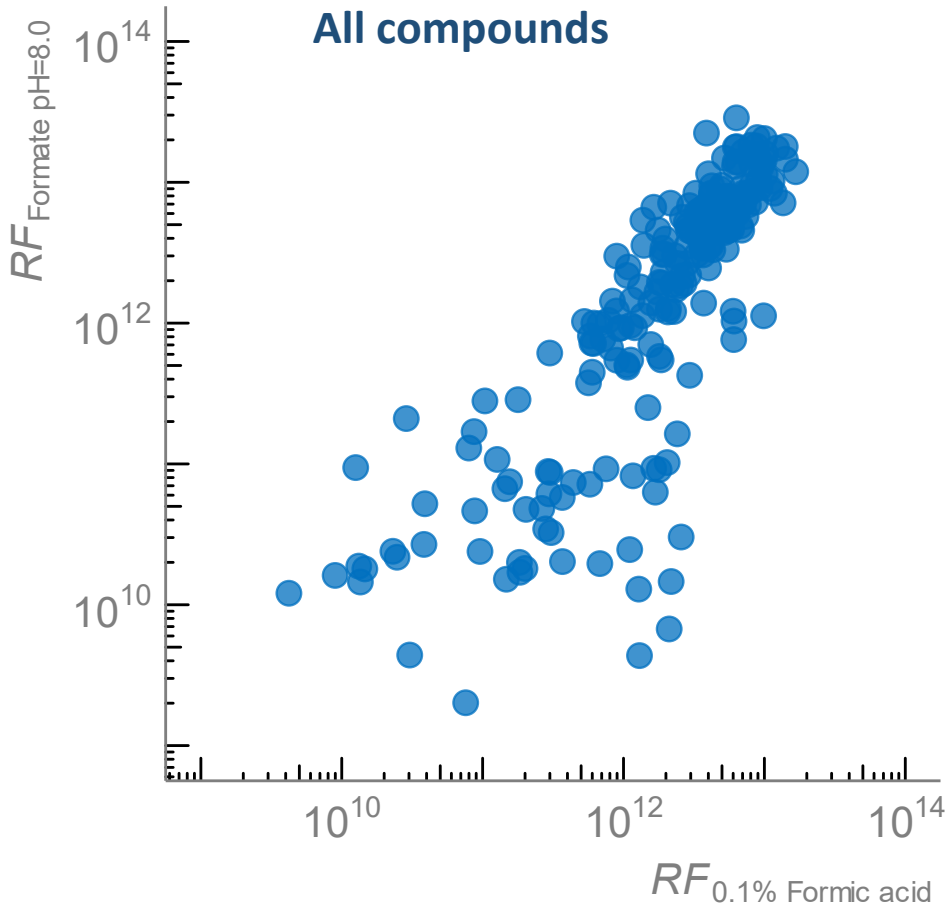
Weak acids

Weak bases

Neutral compounds

RESULTS

pH



pH has significant effect on low ionizable compounds :

Weak acids

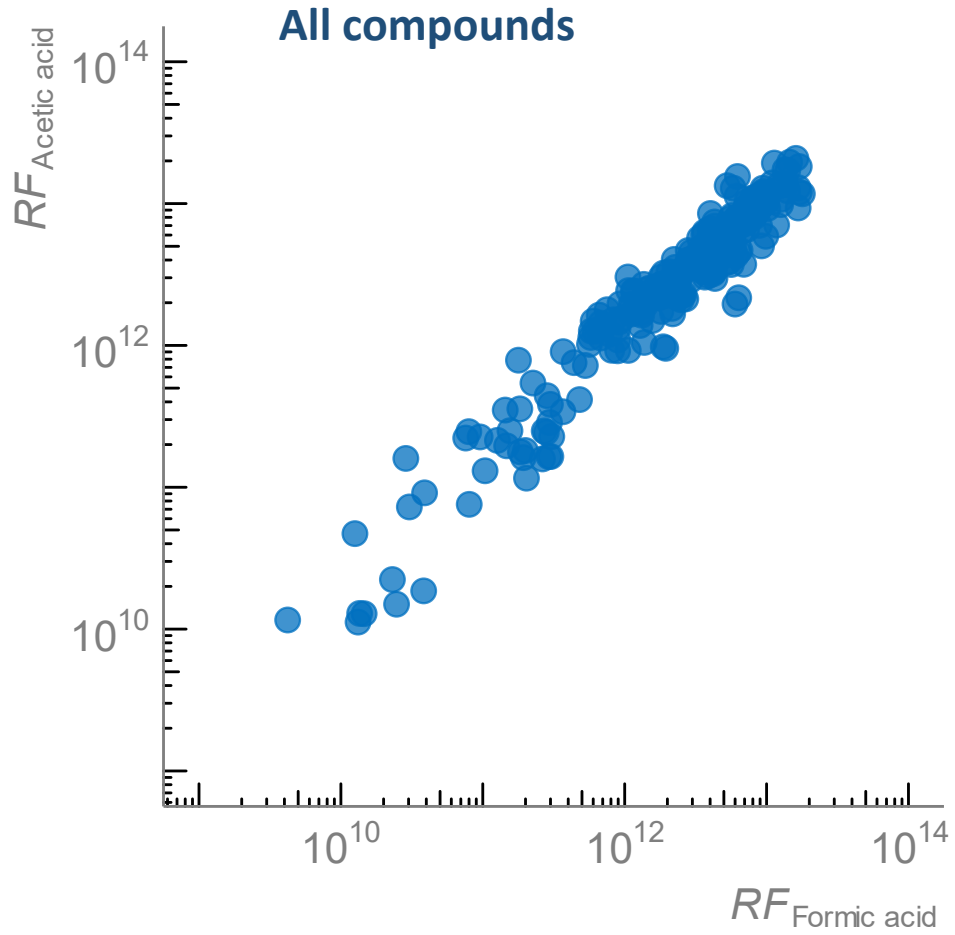
Weak bases

Neutral compounds

The effect of pH is statistically significant

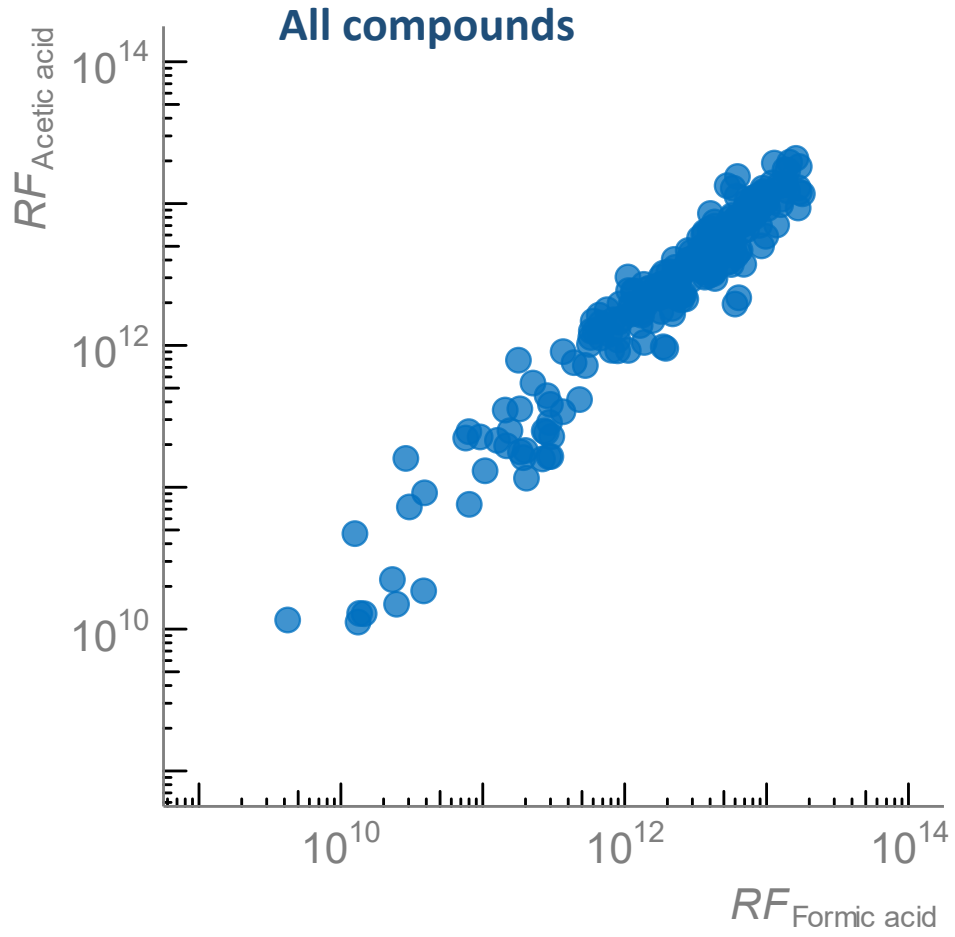
RESULTS

Additive



RESULTS

Additive



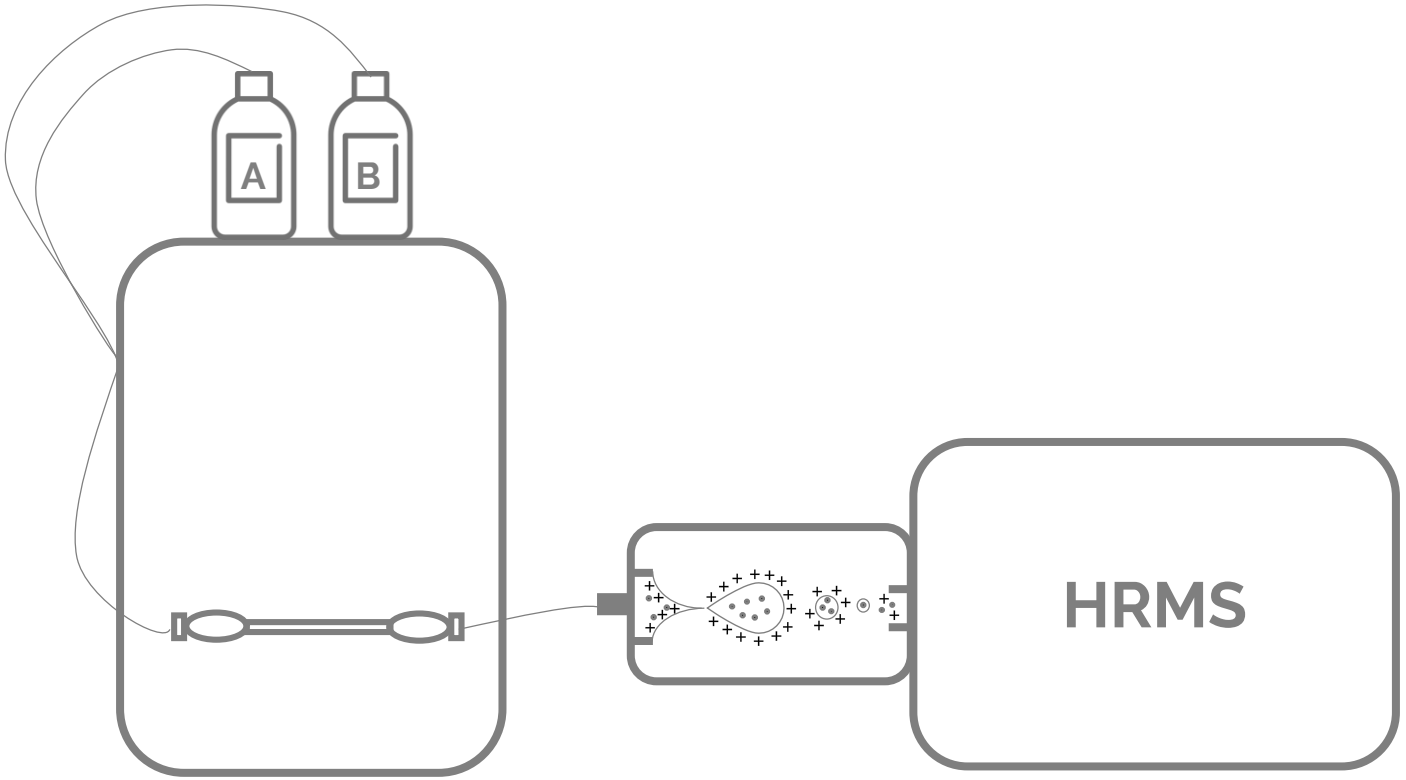
Effect of additive is not significant in the same pH

RESULTS

Experimental: Use three different mobile phases in a non-targeted analysis

RESULTS

Experimental: Use three different mobile phases in a non-targeted analysis



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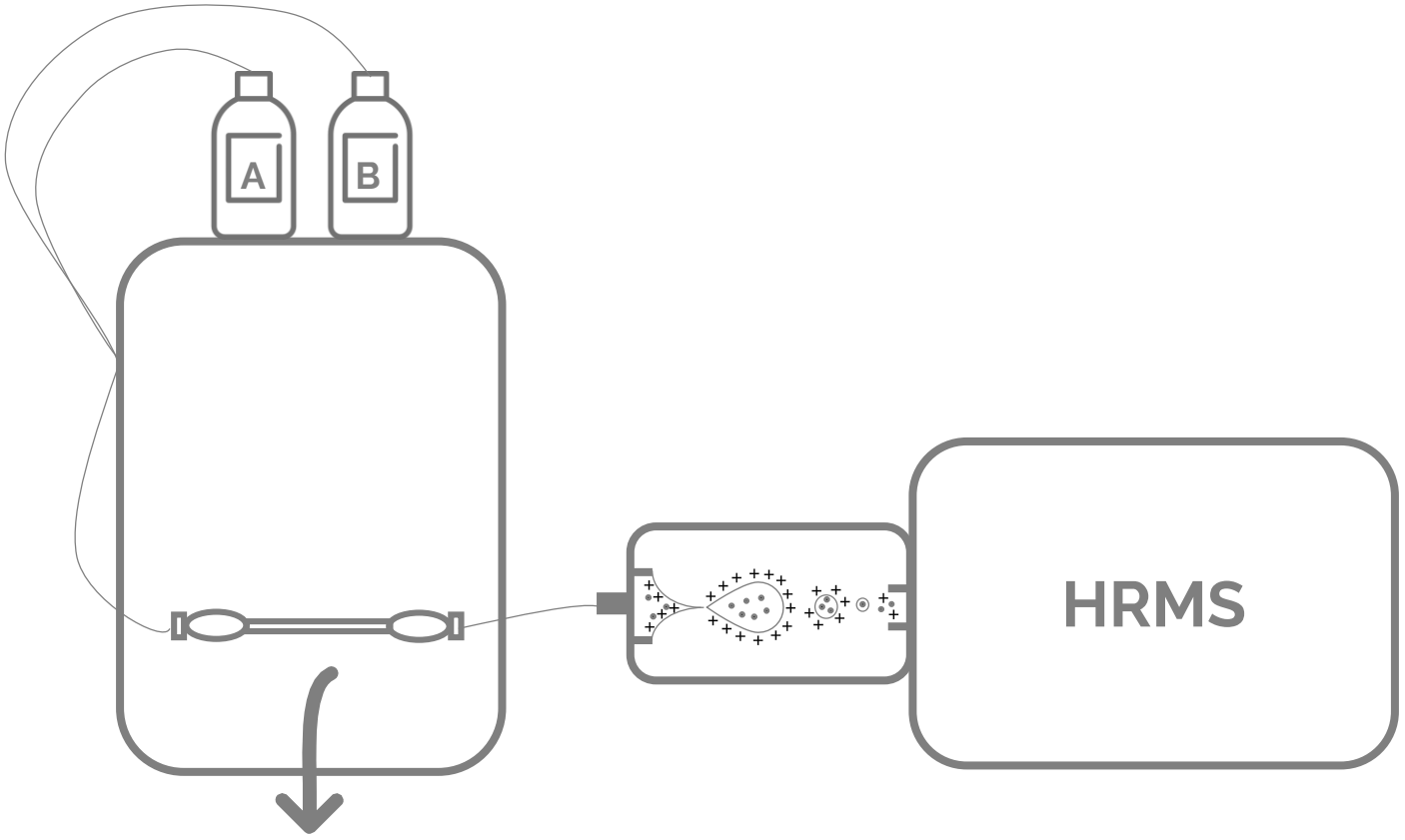
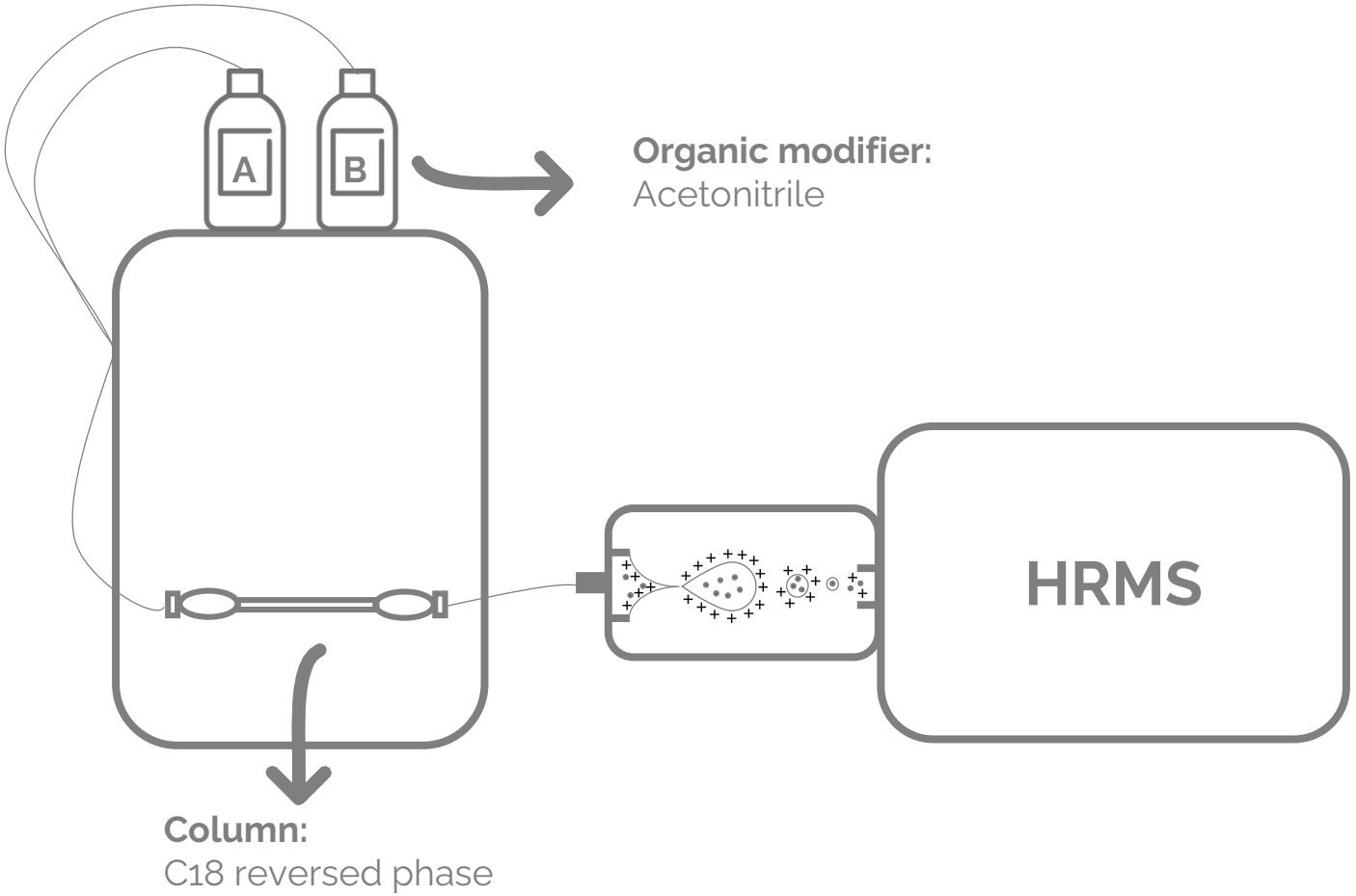


Column:
C18 reversed phase

RESULTS

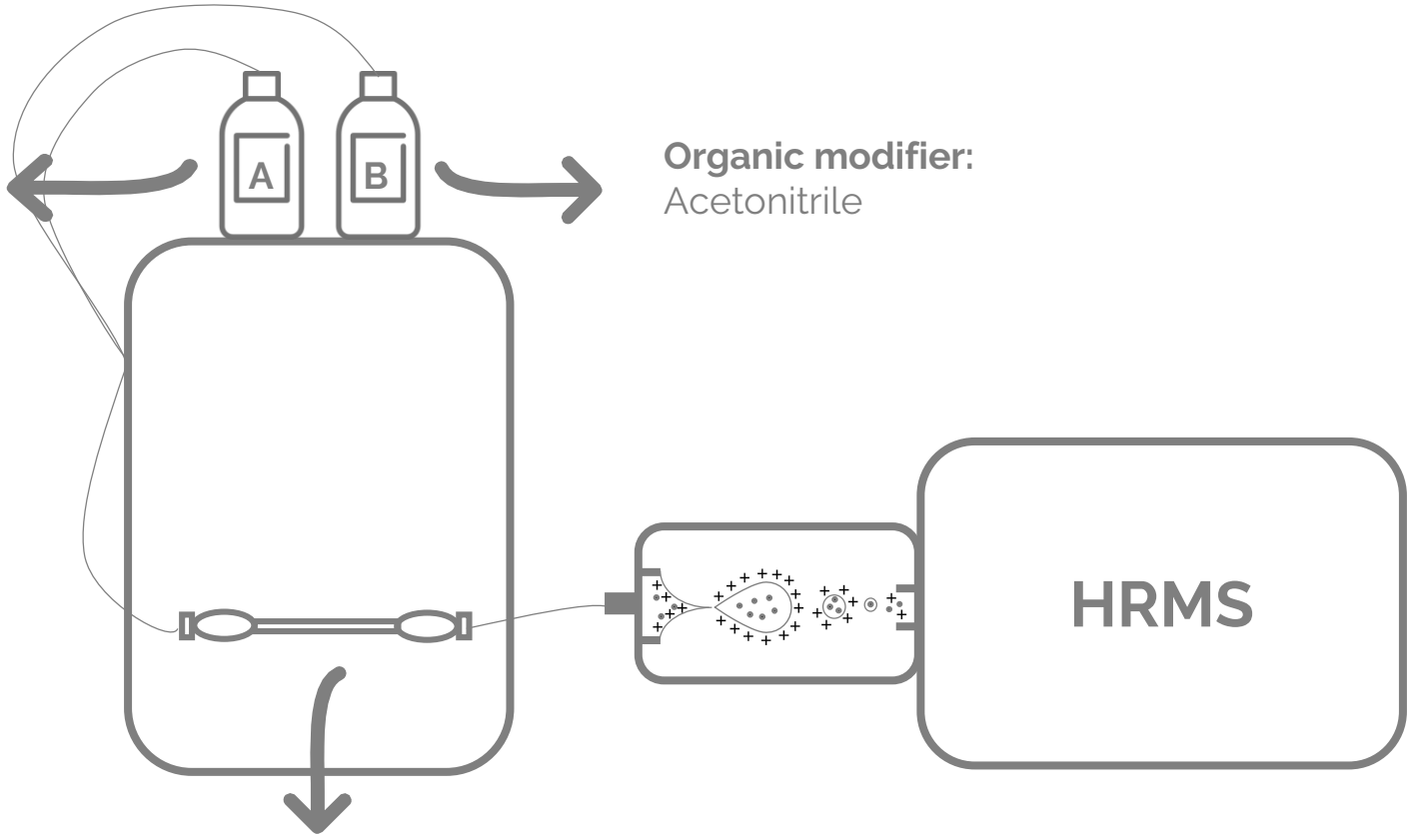
Experimental: Use three different mobile phases in a non-targeted analysis



RESULTS

Experimental: Use three different mobile phases in a non-targeted analysis

Water phase with:
0.1% Formic acid (pH~2.7)
0.1% Ammonia (pH=10.0)
5 mM Ammonium fluoride



Organic modifier:
Acetonitrile

Column:
C18 reversed phase

RESULTS

Experimental: Use three different mobile phases in a non-targeted analysis

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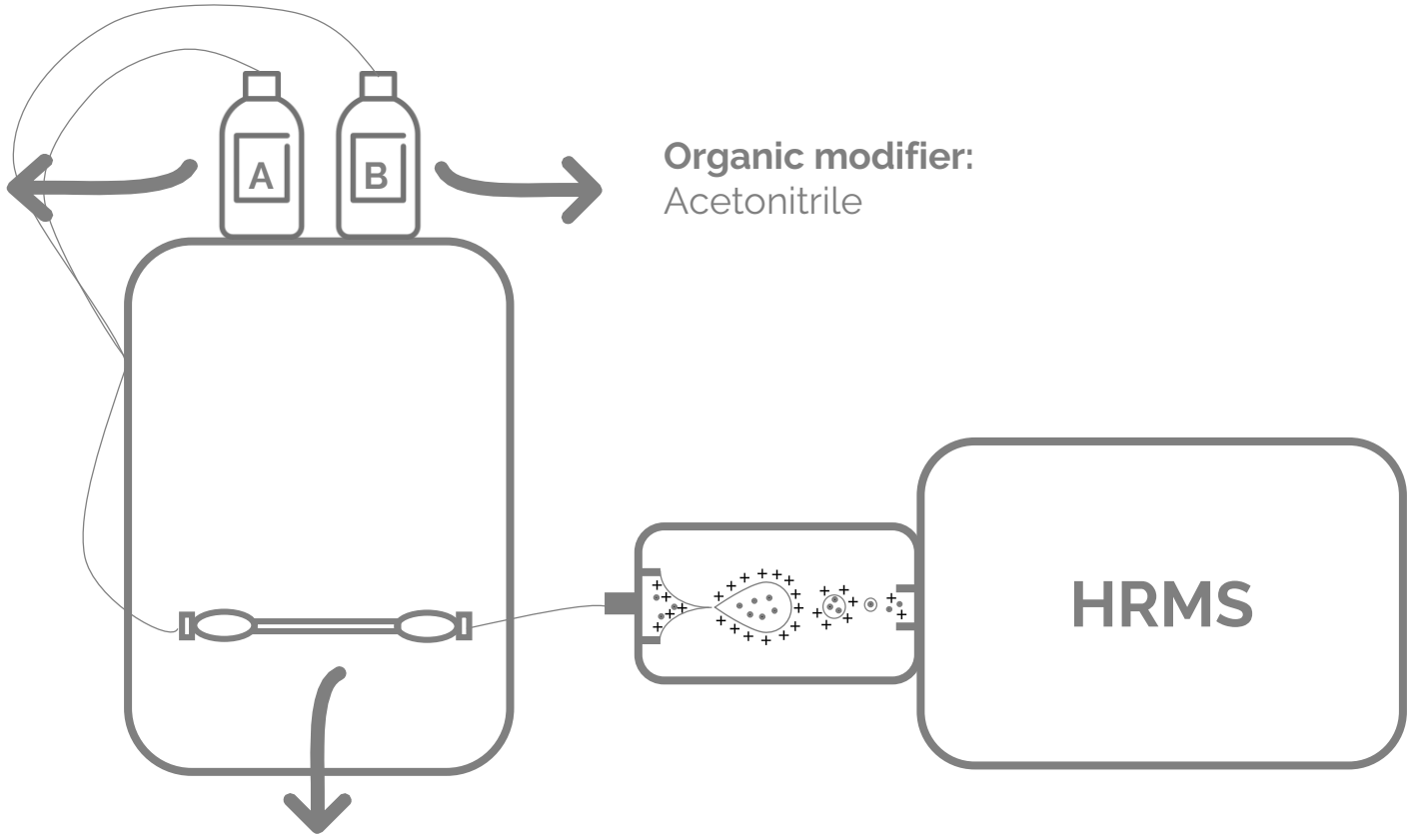


Acquisition: ESI+

Column:
C18 reversed phase

EXPERIMENTAL

Wastewater samples from Henriksdal wastewater treatment plant

EXPERIMENTAL

Wastewater samples from Henriksdal wastewater treatment plant

Influent

Effluent

EXPERIMENTAL

Wastewater samples from Henriksdal wastewater treatment plant

Influent

Effluent

Spiked Influent

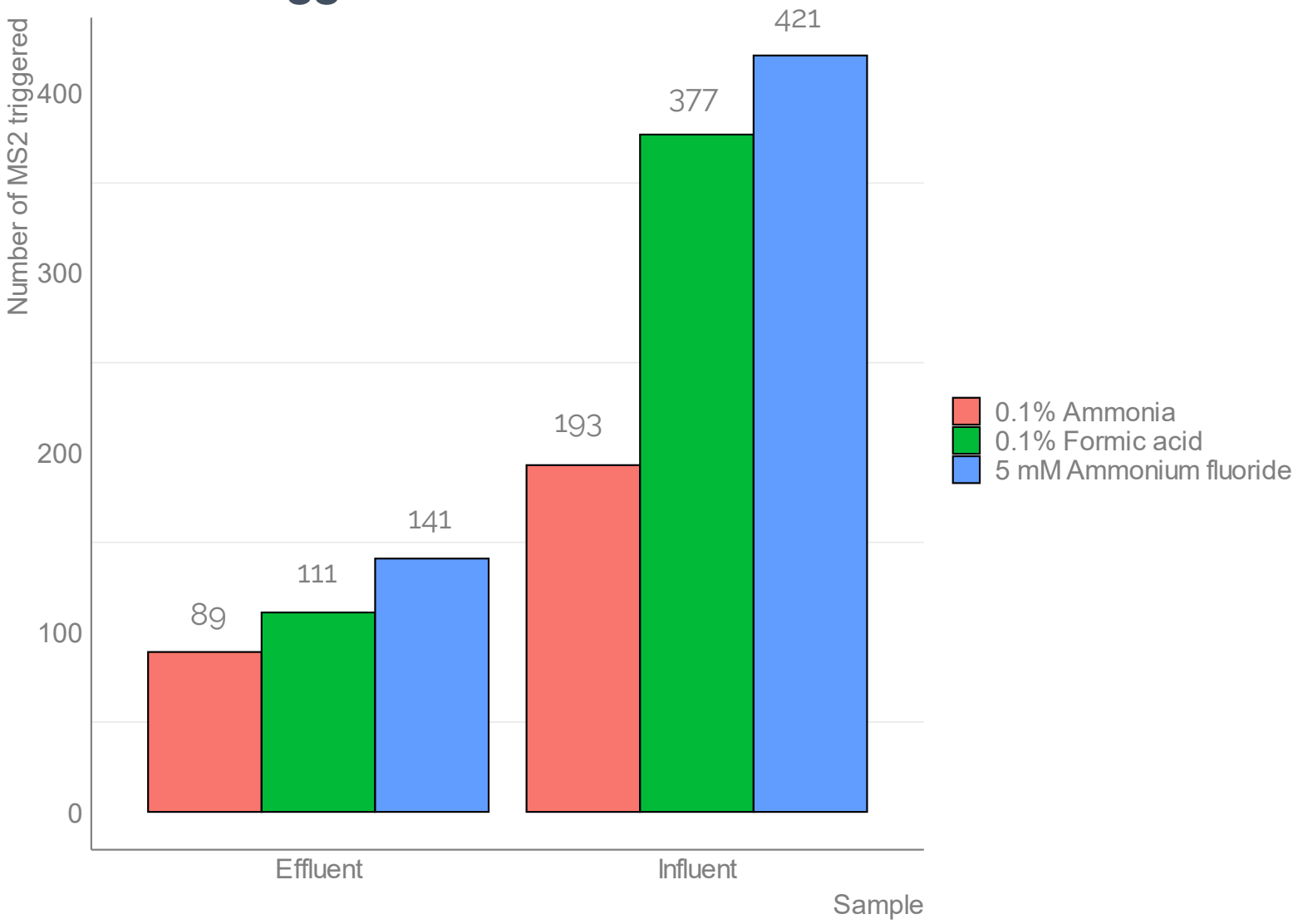
Spiked Effluent

Mix of 79 compounds

Amina Souihi

RESULTS

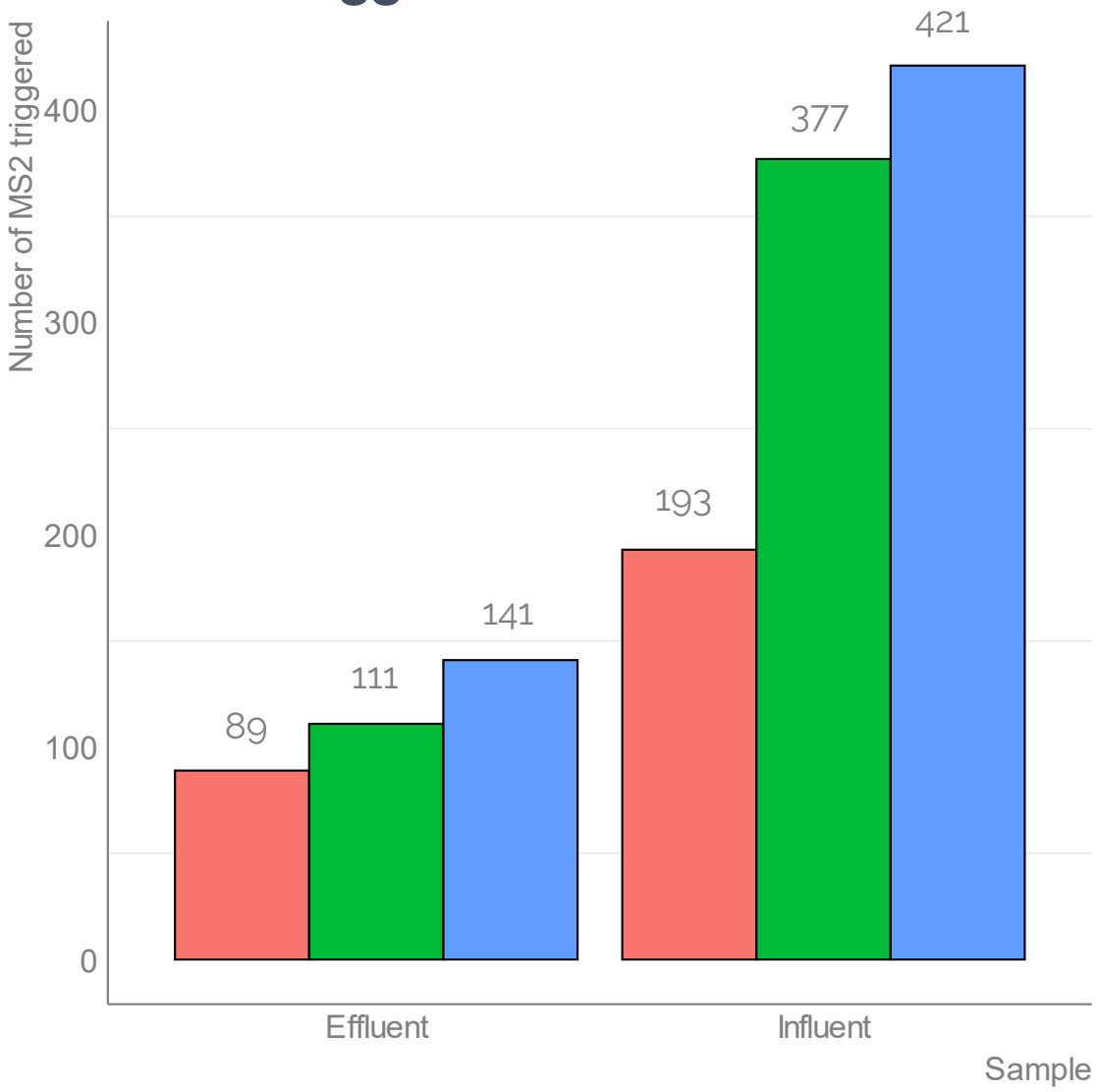
Number of triggered MS^2



Amina Souihi

RESULTS

Number of triggered MS^2

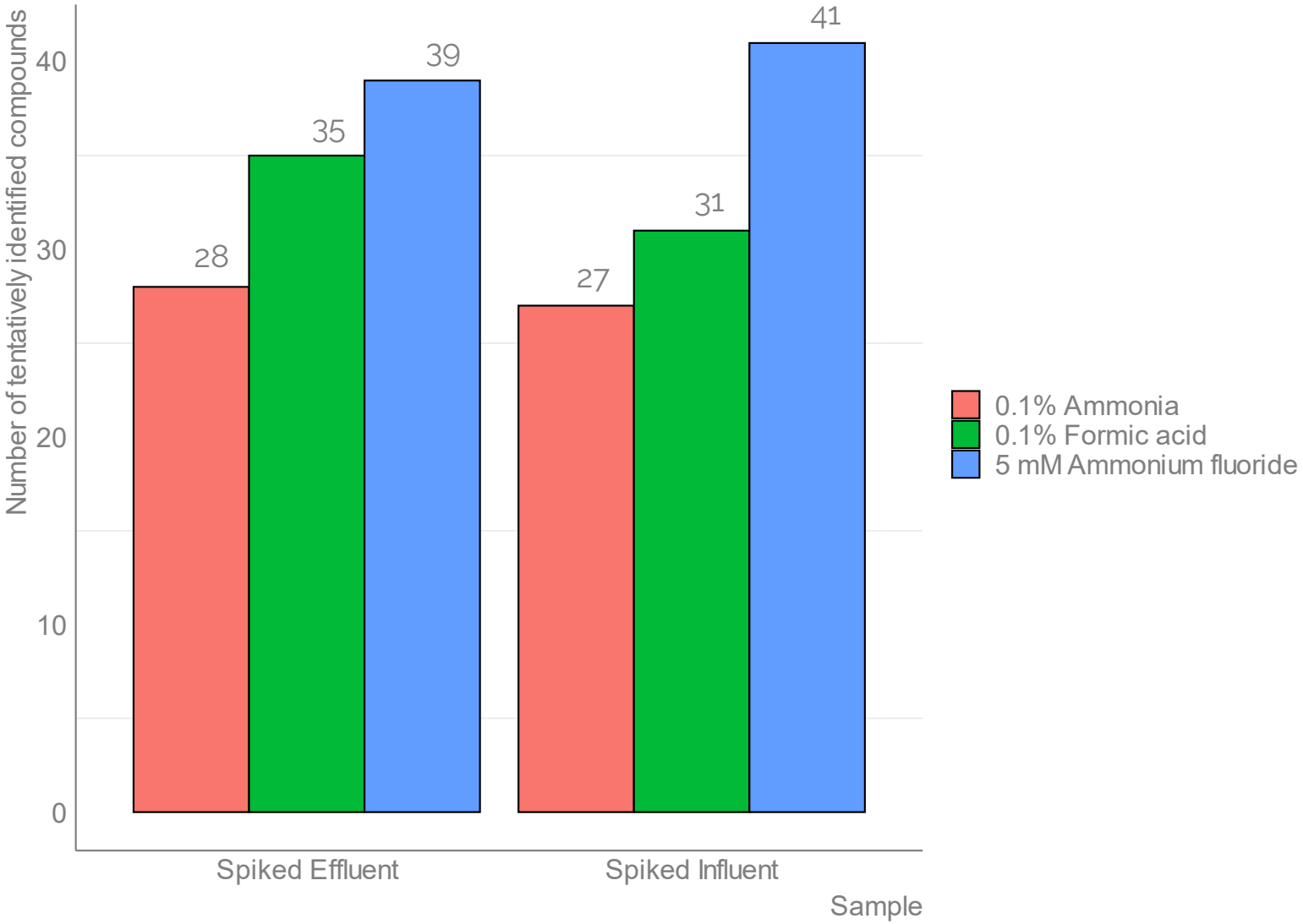


5 mM ammonium fluoride yielded the highest number of triggered MS^2

Amina Souihi

RESULTS

Number of spiked compounds within the SIRIUS+CSI:FingerID tentative structures

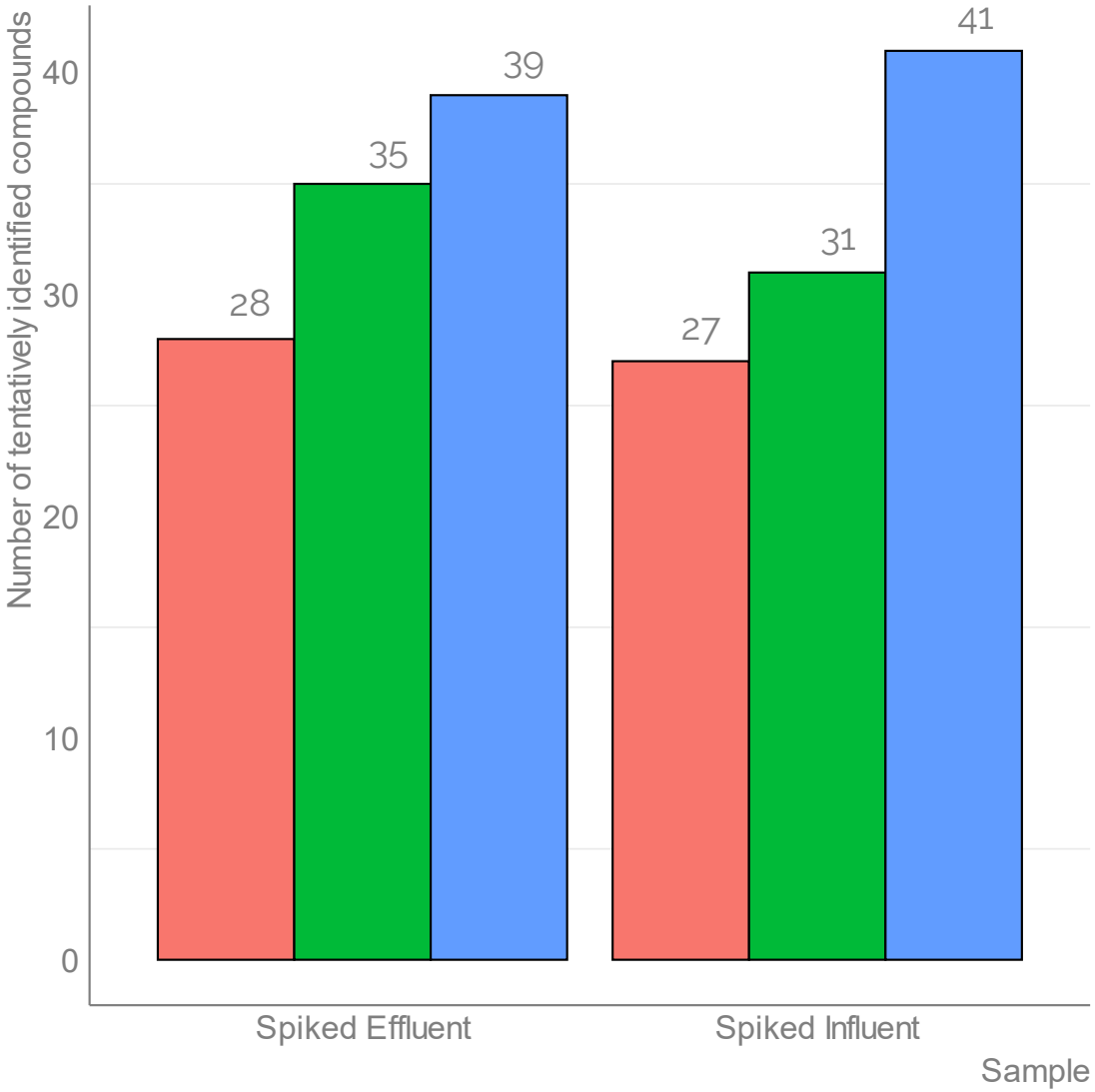


Dührkop et al. (2019)

Amina Souihi

RESULTS

Number of spiked compounds within the SIRIUS+CSI:FingerID tentative structures



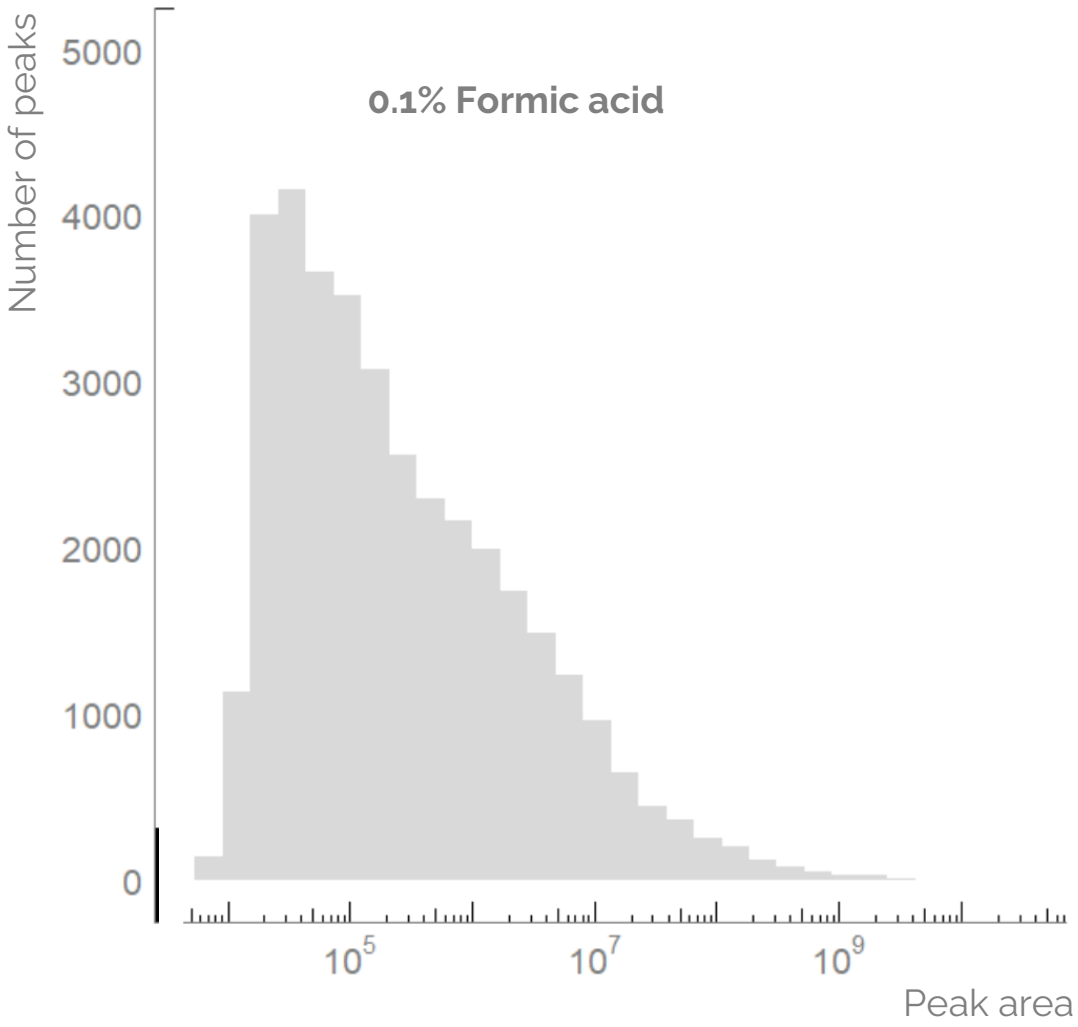
5 mM ammonium fluoride yielded the highest number of spiked compounds within SIRIUS tentative list

Dührkop et al. (2019)

Amina Souihi

RESULTS

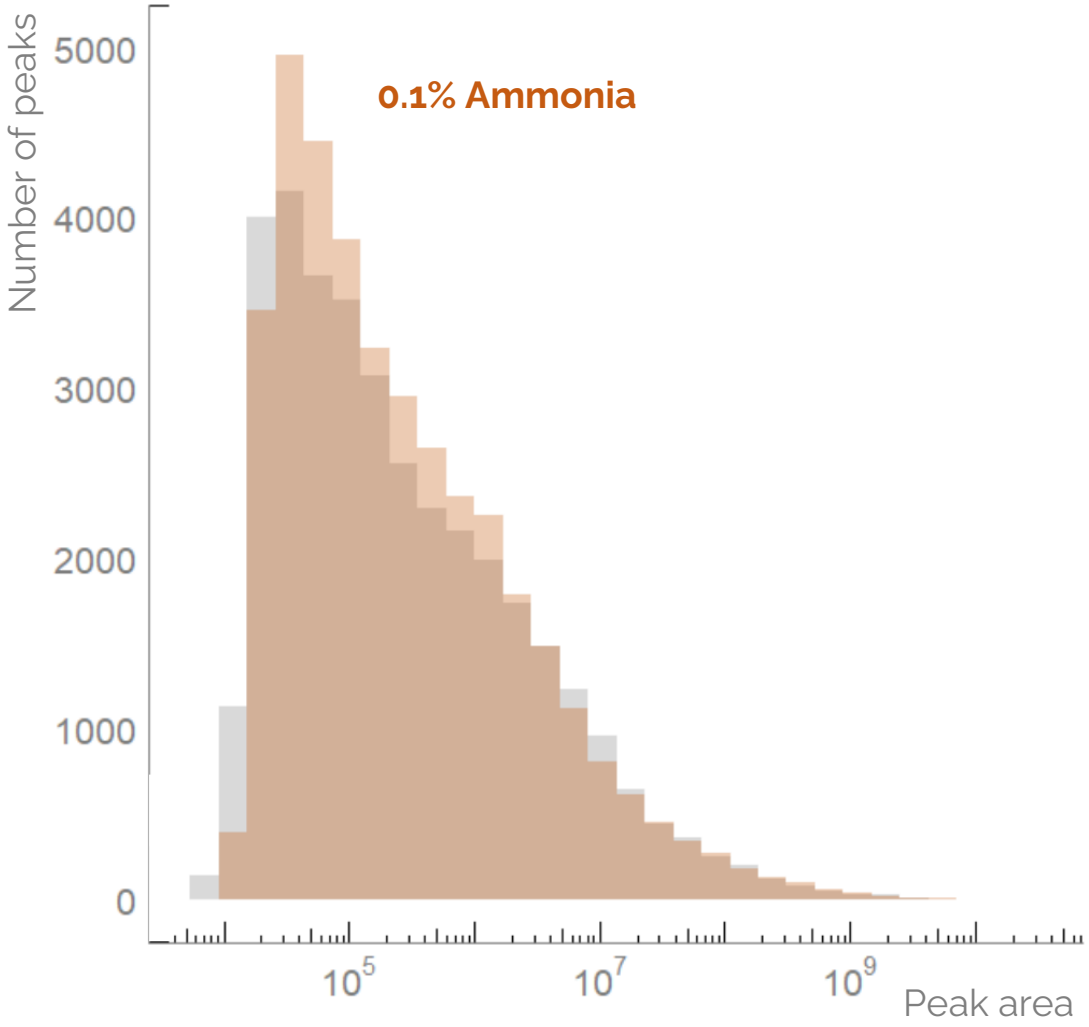
Influent wastewater – Intensity from full scan



Amina Souihi

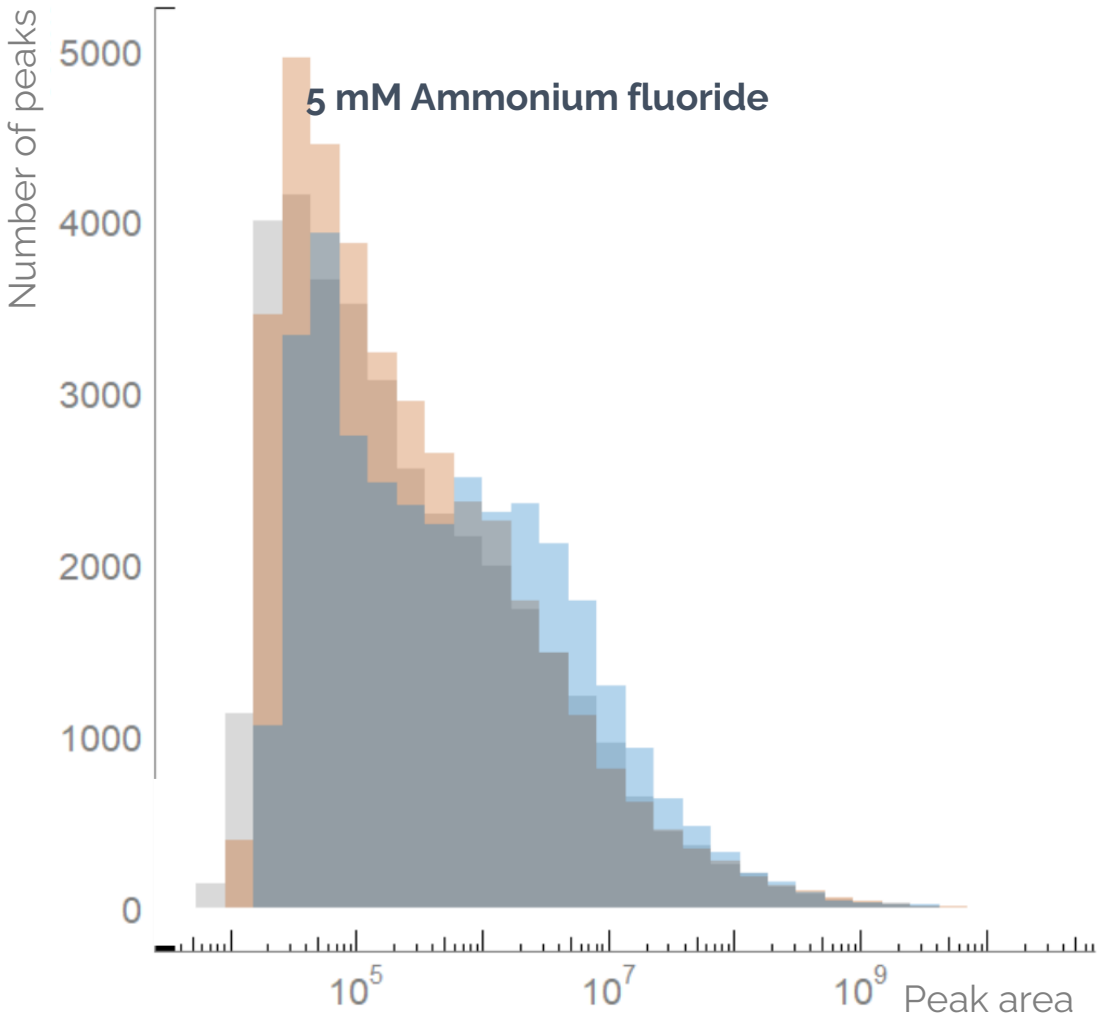
RESULTS

Influent wastewater – Intensity



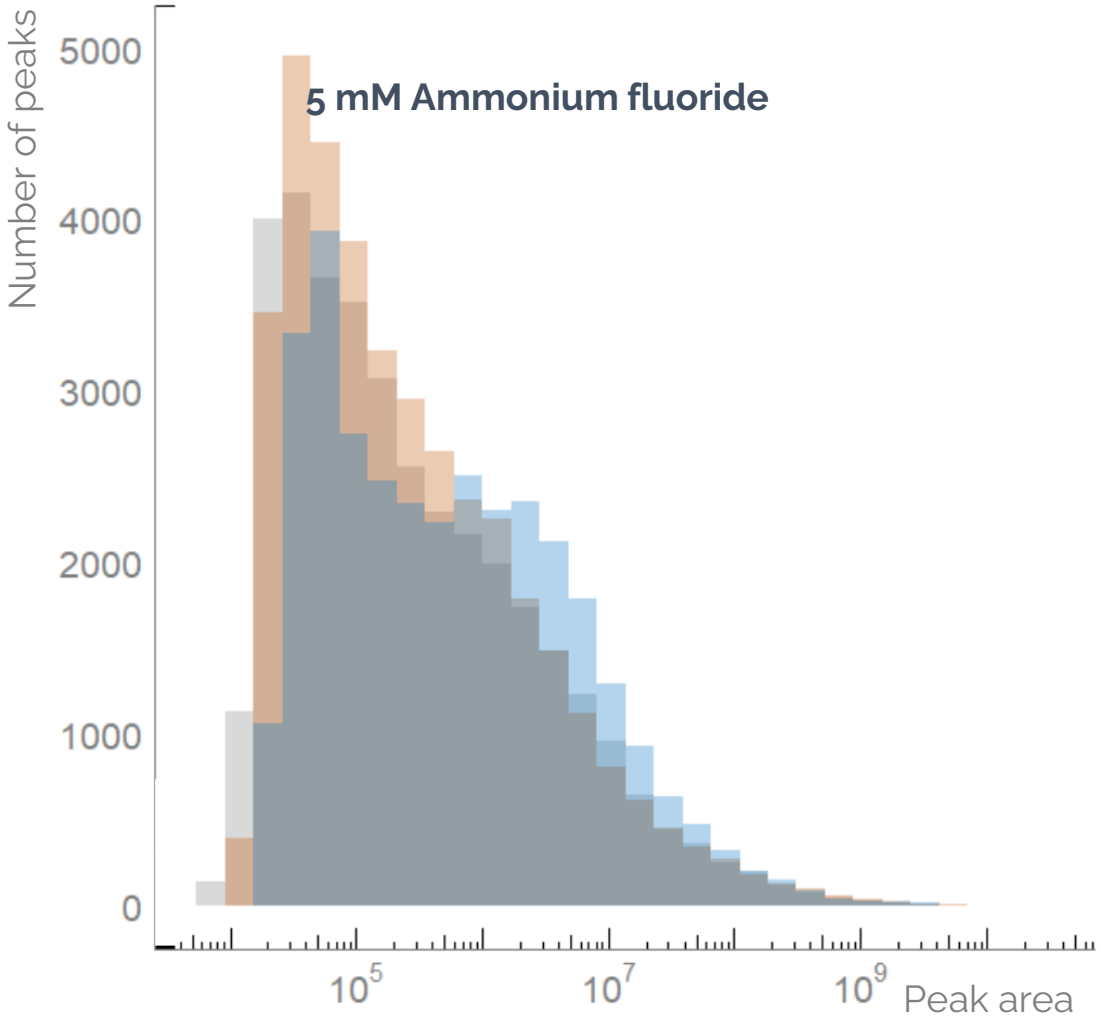
RESULTS

Influent wastewater – Intensity



RESULTS

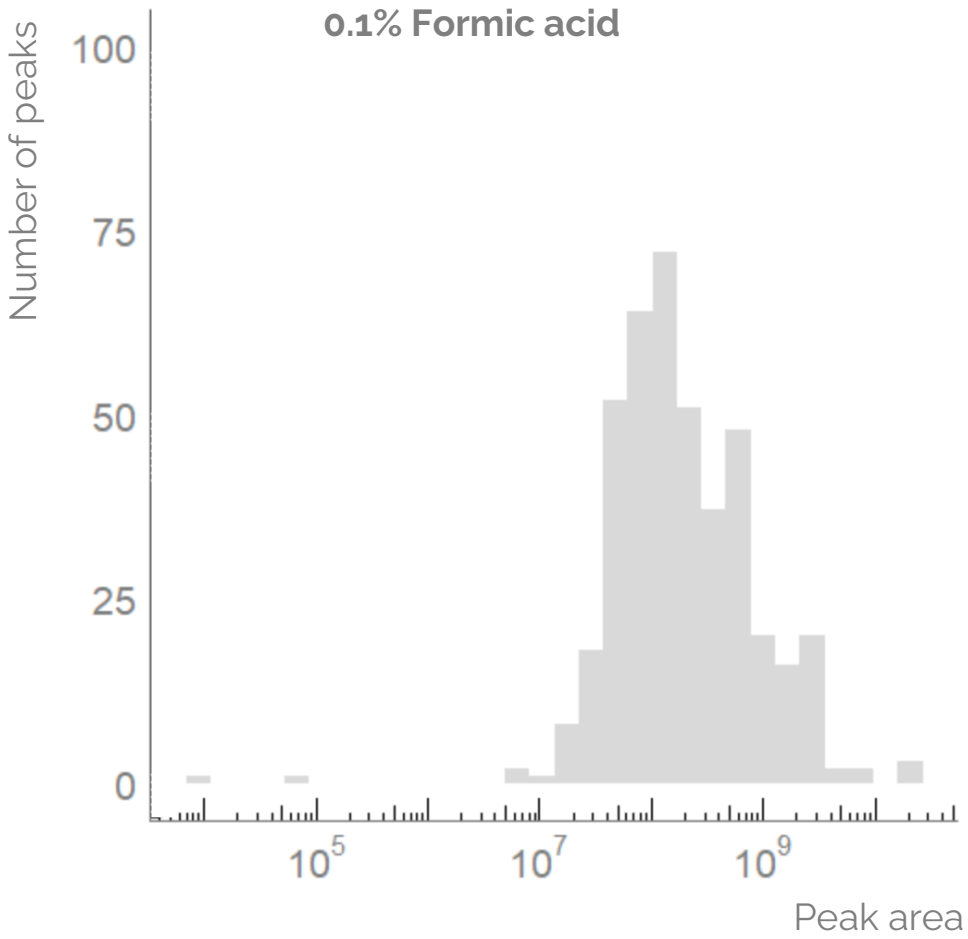
Influent wastewater – Intensity



5 mM ammonium fluoride yielded higher peak areas

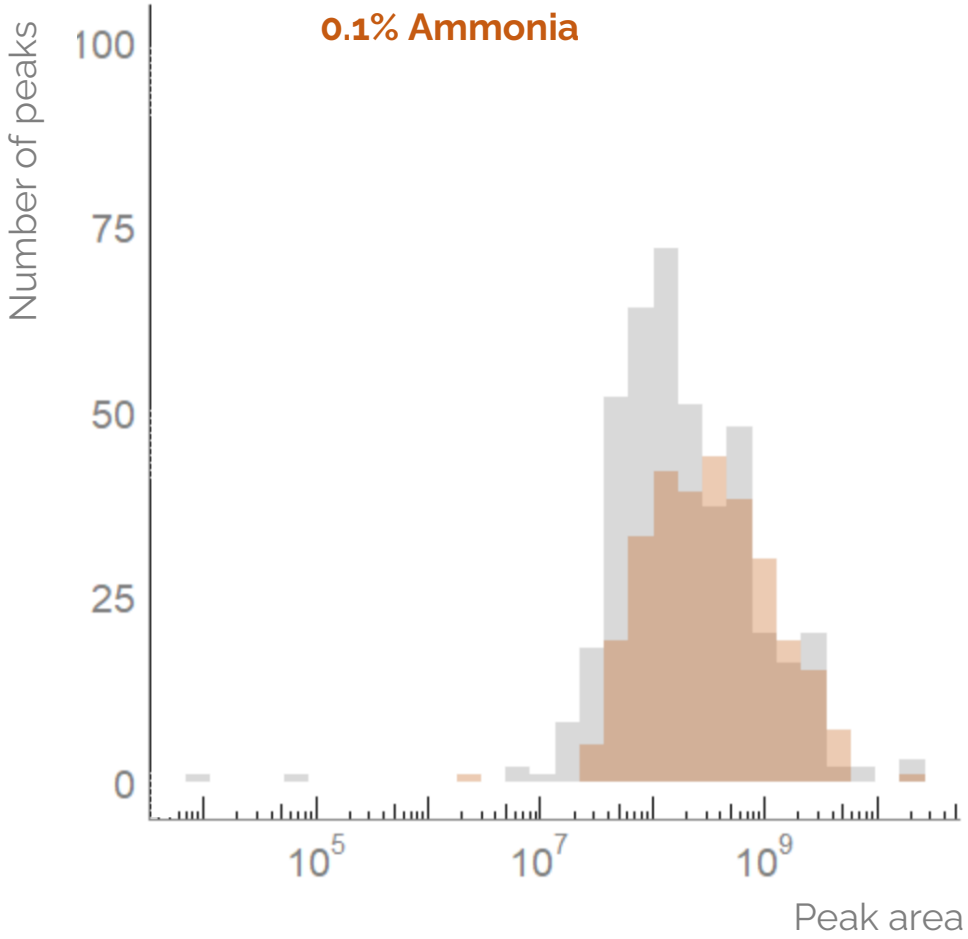
RESULTS

Influent wastewater - Intensity of MS^2 triggered



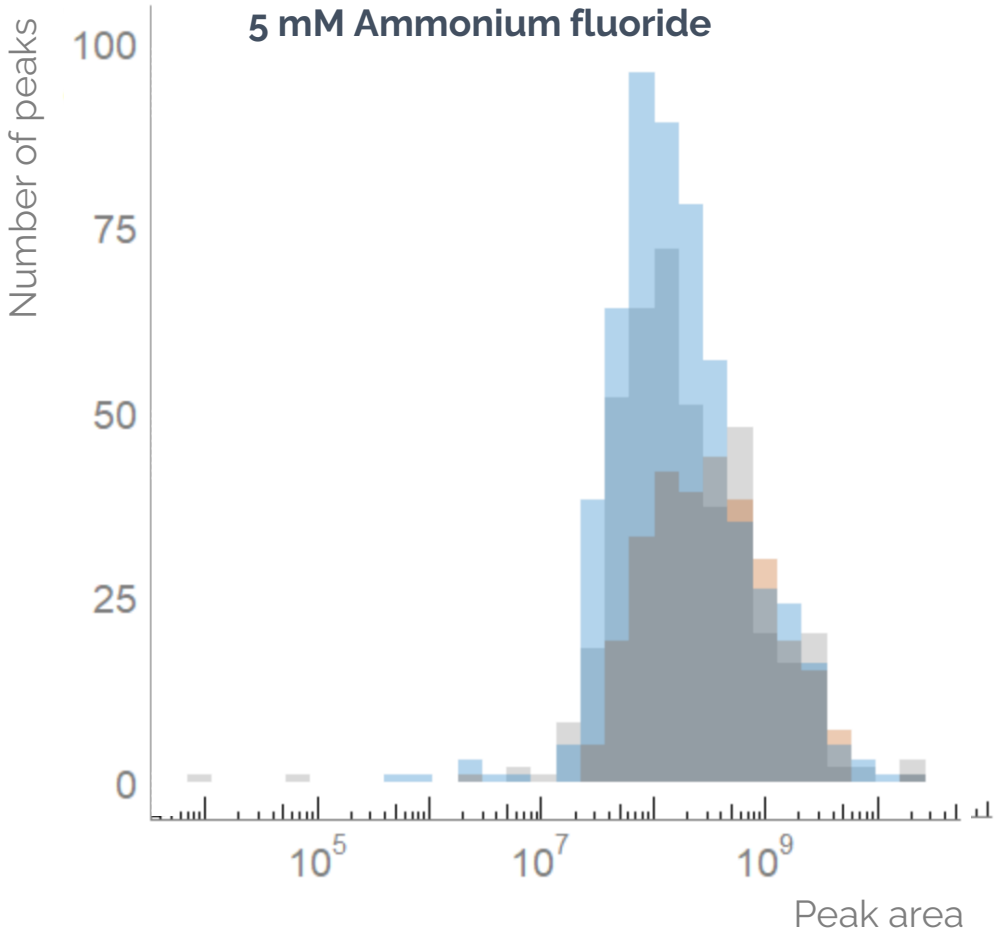
RESULTS

Influent wastewater - Intensity of MS^2 triggered



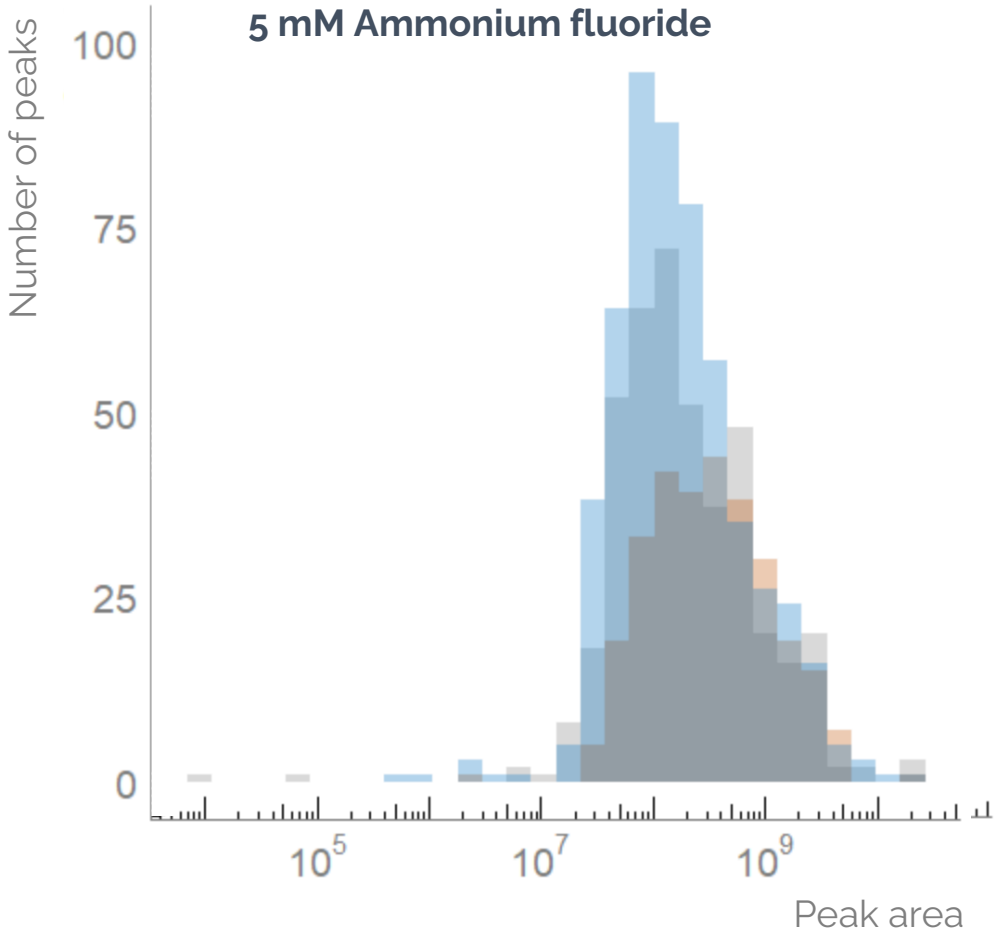
RESULTS

Influent wastewater - Intensity of MS^2 triggered



RESULTS

Influent wastewater - Intensity of MS^2 triggered



5 mM ammonium fluoride yielded the highest number of peaks with MS2 triggered

CONCLUSIONS

pH has a significant effect on the response factors of low ionizable compounds

Acidic conditions improved the response factors of weak bases and acids

Ammonium fluoride showed the highest number of MS^2 triggered and higher median peak areas

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Kruvelab group

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