Contribution of ion mobility for structural analysis and analytical chemistry: use of selective IMS shift reagents (SSR).

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Overview

Ion mobility is a gas phase separation technique based on the Collisional Cross Section (CCS) of ions. It discriminates isobaric and isomeric ions provided their CCS difference is larger than the instrumental resolution. This work proposes a new method to overcome this limitation while providing additional structural information. A Selective Shift Reagent (SSR) is a ligand specifically modifying the CCS of ions. Indeed specific non-covalent complexes can be form with a suitable SSR to reach the required selectivity and the CCS induced shift. A CID dissociation of the complex may be used after IMS separation to produce specific MS/MS spectra of the targeted analyte. This concept paves the way for new analytical strategies by ion mobility based on non-covalent complex formation.

Introduction

Ion mobility (IM) is a separation technique which allows temporal separation of ion depending of theirs three-dimensional structures[1]. To surpass the usual resolution of IM it is necessary to use some strategies in addition to a mobility settings optimization. The used of 18-crown-6 ethers, as shifting reagent[2] to enhance the separation in ion mobility between peptides has already been descripte.[3,4] Depending of the number of 18-crown-6 ethers fixed, peptides were more or less shifted which increase the peak capacity of ion mobility separation. Though, 18-crown-6 ethers are selective to a specific analyte, which lead to an uncontrolled coordination complexes formation. In order to discriminate two ions with similar CCS, it is necessary that shifting reagents are specific to one of them. In this case, the term *selective shifting reagent* (SSR) is introduced. SSR is a ligand which specifically bind with a target ion depending of their physicochemical properties.

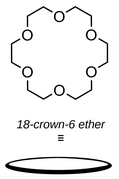
Methods

18-crown-6 ether and 24-crown-8 ether were purchased from Across Organic. β-cyclodextrin was purchased from Sigma Aldrich (Belgium). Mixtures of models and SSR compounds are diluted in 50% methanol, 49.5% water and 0.5% formic acid before the injection. Sample concentration was fixed to 5 or 10µM and SSR concentration was going from 100 to 500µM. Spectra and mobilograms present in this poster have all been acquired with the Waters Synapt G2 (Manchester, England). In the TWIMS cell, the drift gas is nitrogen at moderate pressure (around 2-3 mbar)
ChemDraw was used for designing structure. Chem3D pro v11.0 was used for structure optimization using MM2 force fields (Minimize calculations/MM2/Minimize Energy options). Gaussian09 was used for electronic studies by DFT with the pseudo potentieal B3LYP and the base 6-31G+(d,p) (Fig 1. and Fig 3.) and 6-31G(d) (Fig 2.) for all atoms.

Proof of concept

SSR depending of chemical group

SSR used:



18-crown-6 ether

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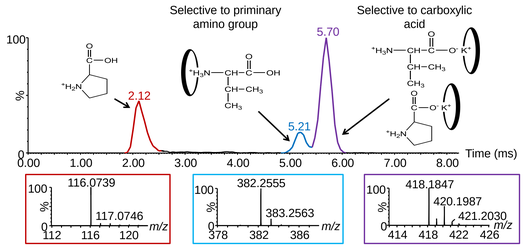


Fig1. : Mobilogram of extract current of ions with a m/z of 116.07 (proline); 118.08 (valine); 382.26 (valine coordinated with 18-crown-6 ether); 418.18 (proline coordinated with 18-crown-6 ether and K+) and 420.20 (valine coordinated with 18-crown-6 ether and K+) of an IM-MS infusion of a 5µM proline, 5µM valine and 250µM 18-crown-6 ether solution.

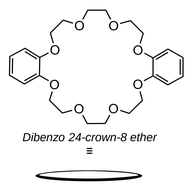
18-crown-6 ether:
- Probe of priminary amino group: Complex formed only with valine
- Specific IMS shifting of compound containing primary amino group

18-crown-6 ether and potassium:
- Probe of carboxylate groups: Complex formed with valine and proline
- Specific IMS shifting of compound containing carboxylate groups

SSR can be selective to specific chemical group (depending on interractions)

SSR depending of steric hindrance

SSR used:



Dibenzo 24-crown-8 ether

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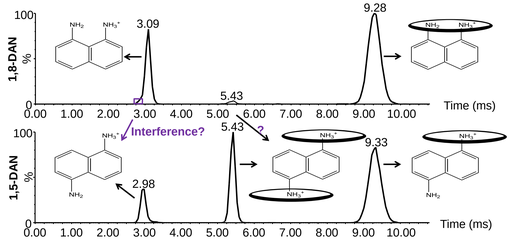


Fig 2.: Mobilograms of extract current of ions with a m/z of 159.10 (corresponding to DAN), 528.28 (corresponding to doubly charged DAN coordinated with two dibenzo 24-crown-8 ether), 607.33 (corresponding to monocharged DAN coordinated with dibenzo-24-crown-8 ether). 1,8-DAN: With 1,8-diaminonaphtalene. 1,5-DAN. With 1,5-diaminonaphtalene.

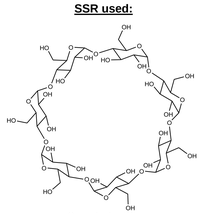
Complexes with one dibenzo 24-crown-8 ether complex:
- Probe of an accessible priminary amino group: Complex formed with both DAN
- Specific IMS shifting of compound containing accessible priminary amino group

Complexes with two (or more) dibenzo 24-crown-8 ether complex :
- Probe of two (or more) accessible priminary amino groups : Complex formed only with 1,5-DAN
- Specific IMS shifting of compound containing carboxylate groups

SSR can selective to the accessibility of specific chemical group (depending on the steric hindrance)

SSR depending of charge repartition

SSR used:



β-cyclodextrin

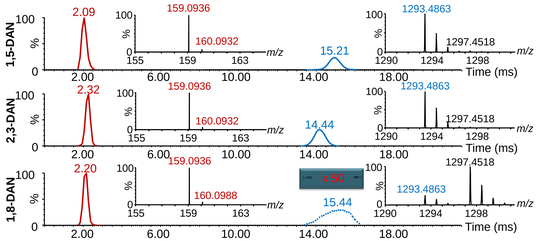


Fig 3.: Mobilograms of extract current of ions with a m/z of 159.10 (corresponding to DAN) and 1293.49 (corresponding to monocharged DAN coordinated with β-cyclodextrin). 1,5-DAN. With 1,5-diaminonaphtalene. 2,3-DAN: With 2,3-diaminonaphtalene. 1,8-DAN. With 1,8-diaminonaphtalene.

Specificity of SSR depend of the position of functional group (charge repartition)
- Complexes only formed with 1,5-DAN and 2-3-DAN

Structure of complexes depend of the position functional group (charge repartition)
- CCS of complex with 2,3-DAN < CCS of complex with 1,5-DAN

SSR can form a specific complex depending of the charge repartition. Use of IM-MS and SSR allow the discrimination between 1,5-DAN; 1,8-DAN and 2,3-DAN

Application to a biological sample

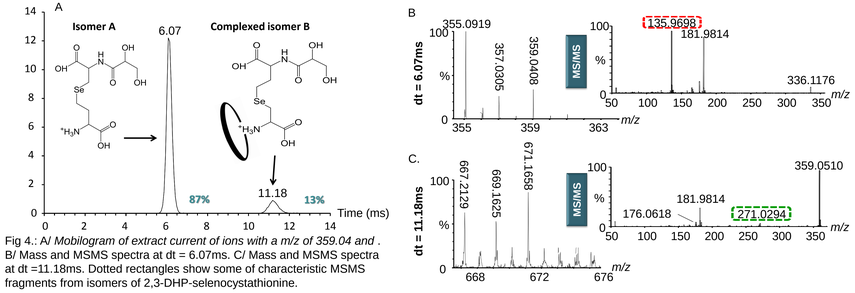
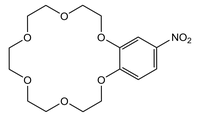


Fig 4.: A/ *Mobilogram of extract current of ions with a m/z of 359.04 and* . B/ Mass and MSMS spectra at dt = 6.07ms. C/ Mass and MSMS spectra at dt =11.18ms. Dotted rectangles show some of characteristic MSMS fragments from isomers of 2,3-DHP-selenocystathionine.

SSR used:



Nitrobenzo 15-crown-5 ether

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Travelling wave ion mobility did not successfully separate the native isomers of 2,3-DHP-selenocystathionine, most likely because of a small difference in collision cross sections of the isomers. The use of a nitrobenzo 15-crown-5 ether as SSR allowed to perform the separation and quantification of the isomer ratio (13% - 87%).

The values presented in this study are in good agreement with theoretical values obtained by computational chemistry and isomer ratio determined from raw data by Dernovics and coworkers after slight separation of this selenocompound extracted from a similar batch of Se-rich yeast by HILIC chromatography coupled to high resolution mass spectrometer.[5]

Conclusions

Proofs of concept were approached in order to artificially improve the resolution of gas phase ion mobility, and also for chemical probing in terms of functional group screening or relative position determination. As the specificity of SSR depends, at least, of the possibility to form coordination complexes, SSR can specifically change the collisional cross section of a target compound. The experimental results observed after ion mobility separation of target ion after coordination complex formation is a shift of its arrival drift time to a superior value than without the addition of SSR. SSR present an interesting analytical purpose to virtually increase the resolution of ion mobility spectrometry. Results suggest that any kind of (weak or strong) interactions between SSR and target compound depend of a lot of physicochemical properties. Consequently, any kind of host-guest systems can be potentially used and optimized for the required selectivity. It could be performed empirically or guided by computational chemistry calculations. This concept paves the way of new possibilities of separation by ion mobility depending of the affinity in gas phase of target ions from a real sample (e.g. biological origin sample) with its respective SSR. The use of SSR enhance the separation of gas phase ion mobility for the identification and relative quantification determination of isomer compounds such as oligopeptides (see Far and coworkers, "359 isomer relative quantification by 3D LC-IMS, to be submitted, Anal. Chem.; Far and coworker, "SeMet isobar contaminant identification by IMS) when the resolution of ion mobility spectrometry is limited.

References

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