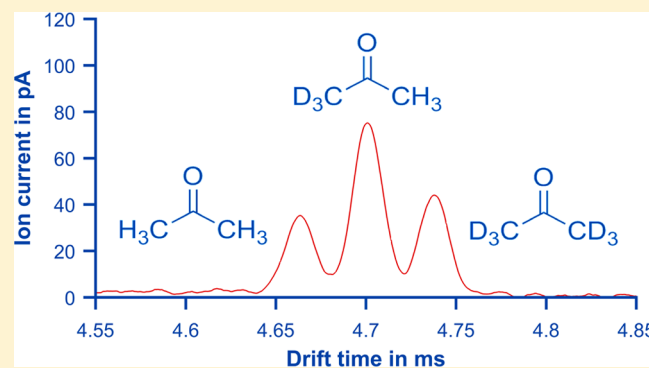Article

Separation of Isotopologues in Ultra-High-Resolution Ion Mobility Spectrometry

Ansgar T. Kirk,*[ORCID] Christian-Robert Raddatz, and Stefan Zimmermann

Leibniz Universität Hannover, Institute of Electrical Engineering and Measurement Technology, Appelstrasse 9A, 30167 Hannover, Germany

ABSTRACT: Ion mobility spectrometry provides ion separation in the gas phase mainly based on differing ion-neutral collision cross sections, enabling powerful analysis of many isomers. However, the separation also has a miniscule mass dependence due to the acceleration and collision properties. In this work, we show for the first time that using a compact ultra-high-resolution ion mobility spectrometer with a resolving power of 250 and an UV ionization source enables the separation of isotopologues with ion mobility spectrometry. This is demonstrated for regular and perdeuterated acetone, benzene, and toluene as well as toluene-$^{13}C_7$ in nitrogen and in purified air as drift gas. The observed peak shifts in the ion mobility spectrum agree with the basic ion mobility equation when using nitrogen as drift gas and also agree with a combination of this equation with Blanc's law when using purified air as drift gas. For benzene and toluene, a reduction in the ion-neutral collision cross section of the isotopically replaced species is observed. Furthermore, a third peak formed from regular and perdeuterated acetone is observed, which can most likely be attributed to the exchange of a methyl group.

Ion mobility spectrometers (IMS) separate ions based on their motion through a neutral drift gas under the influence of an electric field.[1] Generally, this separation capability has led to two groups of applications. On the one hand, being an atmospheric pressure ion separation, they are readily coupled to efficient atmospheric pressure chemical ionizations sources, allowing the design of extremely fast and sensitive trace gas detectors. This has led to a widespread use in military and security applications.[2,3] On the other hand, ion mobility spectrometry can be seen as a size-based chromatographic separation, allowing the measurement of ion-neutral collision cross sections[4] or simply adding an additional separation dimension to, for example, MS or GC-MS systems.[5]

The ion mobility K is defined as the proportionality factor between electric field strength E and ion drift velocity v_d and can for low ion energies be estimated by eq 1. It is often referred to as the Mason–Schamp equation, although it dates back much further to the work of Langevin.[6] A comprehensive overview on ion mobility theory can be found in the work of Mason and McDaniel,[7] while Siems, Viehland, and Hill[8] give a more recent derivation of eq 1 with strong focus on the necessary approximations.

$$K = \frac{v_d}{E} = \frac{3}{16}\frac{ze}{N}\sqrt{\frac{2\pi}{k_b T}\left(\frac{1}{m}+\frac{1}{M}\right)}\frac{1}{\Omega} \qquad (1)$$

Equation 1 parameters are the charge state of the ion z, the number of drift gas molecules per volume N, the absolute temperature T, the mass of the ion m, the mass of a drift gas molecule M, and the collision cross section between them, Ω. It is especially important to note that the mobility of an ion is always specific to a specific drift gas. As the ion mass and the neutral molecule mass are combined into a so-called reduced mass, which is dominated by the smaller of the two masses, the mass of the ion only plays a miniscule role in most applications, as it is typically heavier than the small drift gas molecules. Thus, ion mobility spectrometry is mostly a separation based on the ratio of ion-neutral collision cross section to charge state, Ω/z. When a dependence on the ion mass is observed, it is typically due to the obvious correlation between molecule size and weight. Consequently, no isotopic effects have been observed in ion mobility spectrometry so far. However, this topic has received strong interest from a theoretical point of view.[9–11] If a separation of isotopes in an ion mobility spectrometer could be achieved, it would be of high interest for several purposes. First, possessing data on the ion mobility of isotopic mixtures enables testing of the equations used to describe the properties of ion mobility, as these equations must be able to explain the observed peak shift. Thus, a large amount of understanding may be gained from such separations. Second, the mobility shift between isotopes can theoretically be used to calculate their mass.[9] This enables direct mass measurement at atmospheric pressure, which could provide a multitude of interesting applications. For example, the mass is needed to calculate the

ion-neutral collision cross section of an ion from its mobility. Knowing the mass under atmospheric pressure conditions as opposed to the one inside a mass spectrometer might provide important insights when clustering with neutral molecules plays a role. Third, isotopically substituted species may be used as markers in ion mobility experiments, as it is now standard procedure in other techniques.

With regard to the last point, it should be noted that isotopic effects such as the separation of isotopomers[12,13] or the two chlorine isotopes[14] have been observed in field asymmetric ion mobility spectrometry (FAIMS), even at lower resolving powers than those currently achieved by low field ion mobility spectrometers. Field asymmetric ion mobility spectrometry does not separate based on the ion mobility itself, but rather on its change at extremely high electric field strengths. Here the difference between the ion mobility at low and high electric field strengths is measured. Thus, several additional nonlinear effects come into play. As at least some of them are mass or mass-distribution dependent, this facilitates separation but has so far made full understanding or calculation impossible.

Furthermore, a shift in ion mobility has been observed when comparing β-adrenergic agonists with their deuterated counterparts.[15] However, these shifts were up to 2 orders of magnitude larger than the shifts expected from the mass difference, indicating that they were most likely caused by a different effect such as possibly a change in the folding of the molecules or alternate charge locations.

◼ EXPECTED MOBILITY SHIFT BETWEEN ISOTOPES

Several theoretical papers on the topic of isotopic effects in ion mobility spectrometry already exist, dealing with the possibility of mass measurement,[9] the expected peak shift from different ion isotopes,[10] or both different ion and drift gas isotopes.[11] However, they all agree on the peak shift between two isotopes or isotopologues at low electric field strengths. The expected relative drift time t_r, defined as the ratio between the two drift times in a drift tube ion mobility spectrometer, of two isotopes 1 and 2 in a monomolecular drift gas can be directly calculated from eq 1 and is given by eq 2.

$$t_r = \frac{t_{d,2}}{t_{d,1}} = \frac{K_1}{K_2} = \sqrt{\frac{m_1^{-1} + M^{-1}}{m_2^{-1} + M^{-1}}} \frac{\Omega_2}{\Omega_1} \tag{2}$$

Under the assumption that the two isotopes possess identical ion-neutral collision cross sections, the last term vanishes and the relative drift time can be calculated directly from the masses. While this is a sound assumption for monatomic ions, where the unchanged electron shell should result in an unchanged ion-neutral collision cross section, isotopic substitutions may theoretically have an effect on the ion-neutral collision cross section of polyatomic ions[16,10] or, more accurately, also on some correction factors which are usually (but wrongfully) absorbed into the ion-neutral collision cross section.[8] A second assumption is that only two different ion isotopes and a single neutral gas isotope are present, whose masses can be put into eq 2 to calculate the shift between the two visible peaks. In reality, however, there will be a large variety of different isotopic replacements in different abundances. As eq 2 is highly nonlinear regarding the masses, it would be theoretically necessary to calculate the shift for every possible ion isotope or isotopologue using a superposition of the different neutral gas isotopes and sum the single peaks to obtain the observed substance peak. For example,

including three oxygen, two carbon, and two hydrogen isotopes while ignoring the position of the replaced atom, acetone can exists in 84 different isotopic combinations. However, when using isotopically labeled substances for the experiment, e.g. regular and perdeuterated acetone, only two isotopologues will be present in high abundance. Furthermore, the low abundance of different drift gas isotopes will also only result in a miniscule difference in ion mobility. This is shown by Figure 1, which
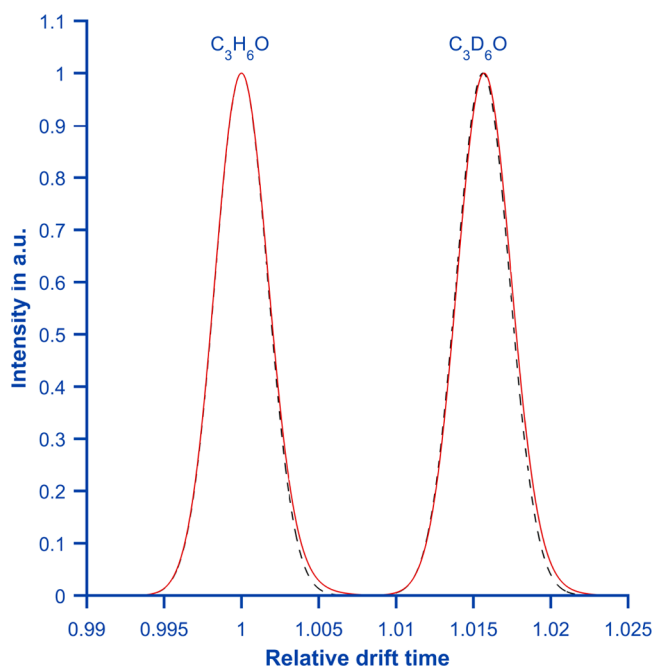
Figure 1. Comparison of two Gaussian peaks placed at the two expected mobilities of regular and perdeuterated acetone (dashed line) with a superposition of the 84 peaks resulting from the full isotope distribution (solid line). Each of the 84 isotopologue peaks is also weighted with the three possible nitrogen isotope combinations in the drift gas. The difference is negligible at a resolving power of $R_p = 250$.

compares the exact superposition of the 84 different isotope combinations in the 3 nitrogen molecule combinations with two Gaussian peaks placed according to eq 2. It can be seen that in such a case, the difference is negligible and eq 2 can still be used to predict the peak positions exactly.

For drift gas mixtures, which are quite common due to ubiquitous use of purified air as a drift gas, the situation is, however, even more complicated. We will use the notation of i as the index for counting different isotopes and j as the index for counting different neutral gases. Thus, the mobility of ion i in neutral gas j shall be denoted $K_{i,j}$. The mobility K_i is then an average of the mobilities $K_{i,j}$ weighted according to their mole fraction f_j, as given by Blanc's law, eq 3.[17]

$$K_i = \left(\sum_j \frac{f_j}{K_{i,j}} \right)^{-1} \tag{3}$$

When calculating the relative drift time, now not only the reduced masses remain, but both the fraction of the different gases and, more importantly, the different ion-neutral collision cross section of the ions in them become weighting factors. This is quite intuitive, as ions will hit larger and more common molecules more often, increasing the influence on the relative

drift time. Thus, the relative drift time becomes eq 4, which simplifies to eq 2 for a single drift gas.

$$t_r = \left(\sum_j \frac{f_j \Omega_{2,j}}{\sqrt{m_2^{-1} + M_j^{-1}}} \right) \Big/ \left(\sum_j \frac{f_j \Omega_{1,j}}{\sqrt{m_1^{-1} + M_j^{-1}}} \right) \qquad (4)$$

It should be noted that eq 4 can be solved without knowing the ion-neutral collision cross section in every drift gas, as the equation can be normalized by dividing through one of the ion-neutral collision cross sections, thus requiring only knowledge about the change between different drift gases. We estimate this change using the polarization limit theory of Langevin,[6,7] assuming that the collision cross section between ion and neutral only changes due to the different polarizabilities of the different neutrals, represented by their relative permittivity κ' as given by eq 5. Better results should be obtainable by using molecular modeling in combination with an accordingly modified version of a mobility simulation tool such as MOBCAL[18,19] or PC/GC,[20,21] which does, however, exceed the scope of this work.

$$\frac{\Omega_{1,1}}{\Omega_{1,2}} = \sqrt{\frac{\kappa_1' - 1}{\kappa_2' - 1}} \qquad (5)$$

Thus, for purified air which contains mostly nitrogen, oxygen, and argon, we will assume the form given by eq 6. It has the advantage that apart from the last term, representing the change of ion-neutral collision cross section between different isotopes, all variables in it are known material properties. Thus, it can be readily solved using only a calculator.

$$t_r = \frac{\dfrac{f_{N2}}{\sqrt{m_2^{-1} + M_{N2}^{-1}}} + \dfrac{f_{O2}}{\sqrt{m_2^{-1} + M_{O2}^{-1}}} \sqrt{\dfrac{\kappa_{O2}' - 1}{\kappa_{N2}' - 1}} + \dfrac{f_{Ar}}{\sqrt{m_2^{-1} + M_{Ar}^{-1}}} \sqrt{\dfrac{\kappa_{Ar}' - 1}{\kappa_{N2}' - 1}}}{\dfrac{f_{N2}}{\sqrt{m_1^{-1} + M_{N2}^{-1}}} + \dfrac{f_{O2}}{\sqrt{m_1^{-1} + M_{O2}^{-1}}} \sqrt{\dfrac{\kappa_{O2}' - 1}{\kappa_{N2}' - 1}} + \dfrac{f_{Ar}}{\sqrt{m_1^{-1} + M_{Ar}^{-1}}} \sqrt{\dfrac{\kappa_{Ar}' - 1}{\kappa_{N2}' - 1}}} \frac{\Omega_{2,N2}}{\Omega_{1,N2}} \qquad (6)$$

Table 1 summarizes the calculated relative drift times between acetone and perdeuterated acetone in pure nitrogen using eq 2 and in purified air using eq 6, while including different amount of detail for eq 6. It can be seen that the difference between the various values for purified air is about an order of magnitude smaller than the difference between nitrogen and air, indicating that a clear difference between nitrogen and purified air should be observable in the experiment. Furthermore, for the first experiments it should therefore not matter which approximation is used for the calculations in air.

Table 1. Calculated Relative Drift Times between Acetone and Perdeuterated Acetone[a]

calculation method	expected relative drift time
eq 2 with N_2 only	1.015620
eq 6 with N_2, O_2	1.015943
eq 6 with N_2 and O_2, and polarizability	1.015930
eq 6 with N_2, O_2, Ar	1.015982
eq 6 with N_2, O_2, Ar, and polarizability	1.015969

[a]The values for the relative permittivity were taken from the overview given by Dunn.[22]

Finally, it is noteworthy that all these calculations assume that the ion mobility spectrometer is still operated within the so-called low-field regime, where the electrical field does not add significant energy to the ions compared to their existing thermal energy. Unfortunately, while estimations do exist,[23] there is no exact rule at which field strengths this will begin to gain a significant influence, and it is therefore always prudent to check if a field dependence of an observed phenomenon exists.

■ REQUIRED RESOLVING POWER

As proposed by Valentine and Clemmer,[9] it is possible to calculate the mass of the ion from the observed peak shift when assuming that both isotopes possess the same ion-neutral collision cross section. By setting $m_1 = m$ and $m_2 = m + n$ in eq 2, it is possible to solve for the mass m of the two isotopes with the mass difference n. Equation 7 is found by ignoring the unphysical solution of a negative mass. Compared to the equation used by Valentine and Clemmer,[9] the use of the relative drift time allows for a much more concise formulation; the results are, however, identical.

$$m = -\frac{M + n}{2} + \sqrt{\left(\frac{M + n}{2}\right)^2 - \frac{Mn}{(1 - t_r^2)}} \qquad (7)$$

As the mobility difference between different isotopes is rather miniscule, it is important to estimate how much resolving power R_p, defined as the ratio of the drift time of a peak to its full width at half-maximum (fwhm), is required to separate them. It can be shown that a resolving power R_p still allows the separation of two peaks possessing the relative drift time $t_{r,min}$ with a valley of 12.5% between them.

$$t_{r,min} = \frac{R_p + 1}{R_p - 1} \qquad (8)$$

Inserting $t_{r,min}$ calculated from eq 8 into eq 7, it is possible to find the highest ion mass that is still separable given a certain resolving power, isotope shift, and drift gas mass. The results from this calculation are plotted in Figure 2, showing the regions accessible using different devices together with the m/n pairs of a variety of test substances. For every line, the shaded region above it represents the separable m/n combinations. Due to their broad and affordable availability as both regular and perdeuterated substances, typical solvents were selected as possible test substances, such as acetone, benzene, toluene, acetonitrile, and acetic acid. However, it should be noted that one possible disadvantage of these substances is that the mass of a deuterium atom is twice the mass of a hydrogen atom. Thus, this substitution is rather extreme and might lead to additional effects.

A resolving power of 50 can be expected from good commercial devices, while a resolving power of up to 100 can only achieved by very few high-end commercial devices but also by several research instruments. However, these are quite insufficient to obtain isotopologue separation. A resolving power of 250 has been obtained by our group using a highly optimized, relatively compact 15 cm drift tube ion mobility
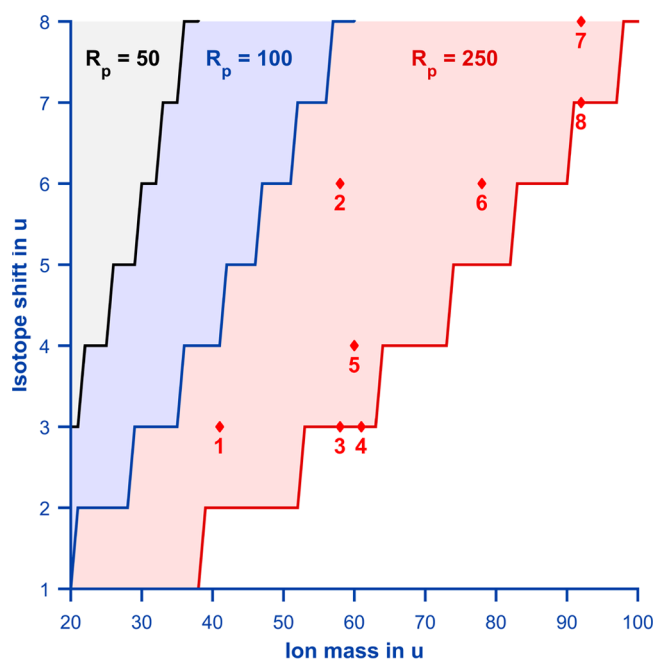
Figure 2. Combinations of different ion masses m and isotope shifts n measurable with different resolving power. The shaded areas are accessible with the respective resolving powers, while the markers denote the chosen test substances. 1: C_2D_3N/C_2H_3N, 2: C_3D_6O/C_3H_6O, 3: $C_3H_3D_3O/C_3H_6O$, 4: $C_3D_6O/C_3H_3D_3O$, 5: $C_2D_4O_2/C_2H_4O_2$, 6: C_6D_6/C_6H_6, 7: C_7D_8/C_7H_8, 8: $^{13}C_7H_8/C_7H_8$.

Table 2. Operational Parameters of the Drift Tube

parameter	value
drift length	153 mm
drift region diameter	21 mm
UV source energy	10.6 eV
injection voltage	1800 V
injection time	5 μs
repetition rate	44 Hz
drift voltage	25 kV
drift field	163 V/mm
aperture voltage	160 V
drift gas flow	150 mL/min
sample gas flow	10 mL/min
dew point of drift gas and sample gas	−90 °C (90 ppbv water vapor concentration)
operating pressure	1018 mbar
operating temperature	25 °C

spectrometer.[24] To date, this has remained the highest resolving power reported for a single charged ion and is sufficient to obtain separation of the test substances marked in Figure 2. It should be noted that even higher resolving powers been reported for large, multiple charged ions using extended separation time techniques at reduced pressure, such as ion cyclotron ion mobility spectrometry[25] or trapped ion mobility spectrometry (TIMS).[26,27] However, neither of these techniques has been demonstrated yet for ions that fall into the region shown in Figure 2, and it is therefore unknown if they would be able to maintain their resolving power for such small ions.

It is also important to note that, because only relative drift times are measured, no high absolute precision is required, just extremely high resolving power and a stable drift gas composition. For example, even if the length of the drift region or the temperature is not known exactly, which would prohibit precision ion mobility measurements,[28] this does not shift the relative drift times.

■ EXPERIMENTAL SETUP

For all experiments in this paper, the ultra-high-resolution ion mobility spectrometer previously described[29,24] was used, whose operational parameters are summarized again in Table 2. For further information, these publications should be consulted.

Special care was taken to maintain an extremely low dew point to ensure that no clustering with water molecules takes places, using a pressure swing absorber and two moisture traps in series. Compared to our earlier publication,[24] the radioactive tritium source was replaced with a krypton discharge UV lamp (Haraeus) to ensure a direct ionization of the target molecule, again to ensure that the measured molecule is indeed the species of interest and not protonated by a water cluster.

Furthermore, the setup now includes the possibility to switch both the drift gas and all gas flows inside the gas mixing apparatus between purified air and nitrogen in order to evaluate the different peak shifts in the different drift gases.

All chemicals were purchased from Sigma-Aldrich Germany with a purity >99%. The perdeuterated substances possess a deuterium purity of >99.6%. All samples were introduced using a Vici Dynacalibrator Model 150 permeation oven and homemade permeation tubes.

It should be noted that voltages as high as 25 kV entail a high risk of hazardous electrical shocks and arcing, and thus experiments including them should only be set up and operated by experienced personal. Especially, all air gaps should be sufficiently large or small according to Paschen's law in order to avoid electrical breakdown.

■ RESULTS AND DISCUSSION

Figure 3 shows the measured spectra of acetone, benzene, and toluene using pure nitrogen as drift and carrier gas. Acetonitrile and acetic acid possess relatively high ionization energies (12.2 and 10.65 eV) and could thus not be ionized with the current UV source. In all four acquired spectra, clearly separated isotopologue peaks are visible. Their nature was verified by adding only one of the two isotopically pure species to the sample gas and observing which of the two peaks remains in the spectrum. The slightly different peak heights are caused by slight differences between the homemade permeation tubes, but are not of concern for the separation. The multiple measurements shown in the first and third panel were aligned with respect to the drift time for better comparability. It is interesting to note that in the acetone spectrum, a third peak appears, which is, as shown by the black traces, present in neither of the two single substance spectra of acetone and perdeuterated acetone. This peak is most likely caused by the exchange of a deuterated methyl group against a nondeuterated one, as it requires both acetone species to form and possesses a mobility between theirs. Identifying a possible mechanism for this is still part of our current research.

All experiments were repeated 15 times in order to evaluate the reproducibility of the measurement and obtain a confidence interval for the shift. Table 3 lists the calculated relative drift time from eq 2 under the assumption of constant ion-neutral collision cross sections, the measured relative drift time, and the relative 95% confidence interval of the measurement as well as
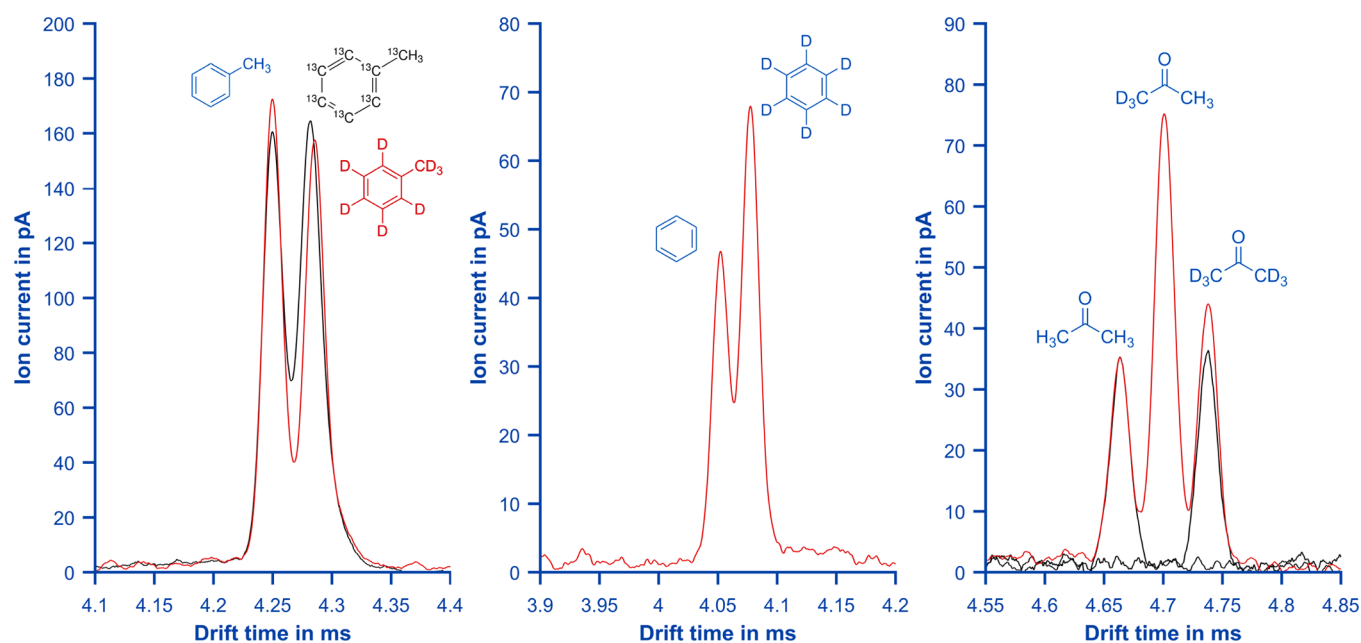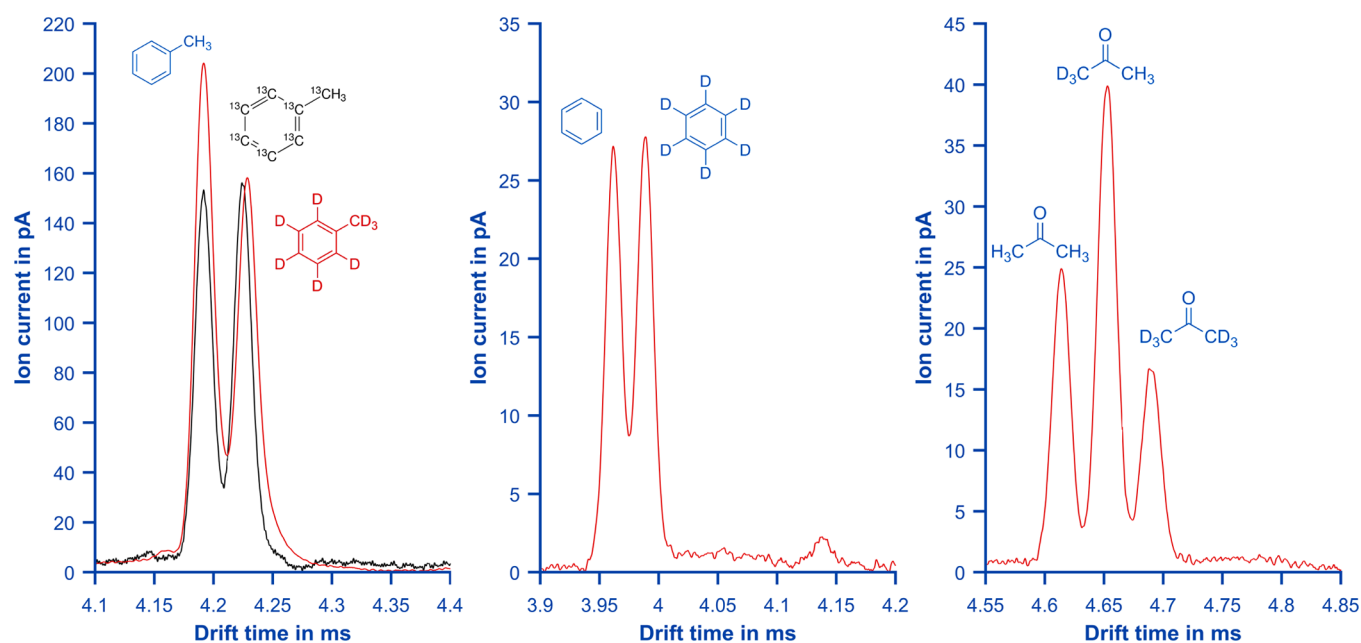
Figure 3. From left to right spectra in nitrogen of toluene and perdeuterated toluene as well as toluene and toluene-$^{13}C_7$, benzene and perdeuterated benzene, and acetone, acetone with one deuterated methyl group, and acetone with two deuterated methyl groups. The black lines in the third panel are the single substance spectra of acetone and perdeuterated acetone.

Table 3. Calculated and Measured Peak Shifts in Nitrogen

peak 1	peak 2	calculated relative drift time	measured relative drift time	95% confidence interval in %	calculation difference in %
C_3H_6O	C_3D_6O	1.015620	1.015948	±1.69	−2.05
C_3H_6O	$C_3H_3D_3O$	1.008104	1.008077	±2.76	0.33
$C_3H_3D_3O$	C_3D_6O	1.007456	1.007808	±2.39	−4.50
C_6H_6	C_6D_6	1.009570	1.006040	±3.40	58.45
C_7H_8	C_7D_8	1.009466	1.008269	±0.43	14.47
C_7H_8	$^{13}C_7H_8$	1.008353	1.007609	±0.73	8.90

Figure 4. From left to right spectra in purified air of toluene and perdeuterated toluene as well as toluene and toluene-$^{13}C_7$, benzene and perdeuterated benzene, and acetone, acetone with one deuterated methyl group, and acetone with two deuterated methyl groups.

the difference between calculated and measured values. The confidence interval and calculation difference do not refer to the relative drift time, but only to the shift, that is the relative drift time minus one. We chose this notation as otherwise all

Table 4. Calculated and Measured Peak Shifts in Purified Air

peak 1	peak 2	calculated relative drift time	measured relative drift time	95% confidence interval in %	calculation difference in %
C_3H_6O	C_3D_6O	1.015969	1.016408	±0.42	−2.68
C_3H_6O	$C_3H_3D_3O$	1.008282	1.008337	±0.37	−0.66
$C_3H_3D_3O$	C_3D_6O	1.007623	1.008004	±0.63	−4.76
C_6H_6	C_6D_6	1.009804	1.006693	±1.02	46.46
C_7H_8	C_7D_8	1.009709	1.008670	±1.72	11.98
C_7H_8	$^{13}C_7H_8$	1.008567	1.007850	±0.48	8.37

errors would appear to be extremely small due to the relative drift time always being about one.

It can be seen that in the case of the three acetone peaks, the shifts lie within or close to the reproducibility of the measurement. Although the peak with two deuterated methyl groups appears to be slightly too slow compared to the two other peaks, this difference is rather small. In the measurements of perdeuterated toluene, toluene-$^{13}C_7$, and especially perdeuterated benzene, however, a significant difference between the calculated and measured relative drift times can be observed. Especially the perdeuterated benzene peak should be 58% further away from its nondeuterated counterpart than it is in the measurement. To elaborate these deviations further, all four measurements were repeated using purified air as drift gas instead of nitrogen. The results of these measurements are shown in Figure 4. Again, the same peaks as in Figure 3 are visible, although they are all shifted to the left due to the change in drift gas. The measurement results are summarized in Table 4, which uses the same notation as Table 3. As expected from theoretical results due to the heavier oxygen molecules and argon atoms, an increase in all peak shifts can be observed. Again, the perdeuterated acetone peak is slightly too slow when compared to the other two acetone peaks, while the perdeuterated benzene and toluene as well as the toluene-$^{13}C_7$ are significantly too fast compared to their regular counterparts.

A possible explanation for this phenomenon can be seen from eq 2 and eq 6. When calculating the expected relative drift time, a constant ion-neutral collision cross section between isotopologues was assumed. However, both equations contain the same factor representing this very change in ion-neutral collision cross section in nitrogen. Thus, by dividing the measured relative drift time by the calculated one, it should be possible to obtain the change in ion-neutral collision cross section. The results of this calculation are summarized in Table 5.

Both measurements yield consistent results, indicating an approximately constant ion-neutral collision cross section between acetone with and without a deuterated methyl group and consequently a slightly larger ion-neutral collision cross

Table 5. Comparison between the Ion-Neutral Collision Cross Section Changes Estimated from Nitrogen and Air Measurements

peak 1	peak 2	$\Omega_{2,N2}/\Omega_{1,N2}$ in %	
		eq 2 nitrogen	eq 6 air
C_3H_6O	C_3D_6O	100.032	100.043
C_3H_6O	$C_3H_3D_3O$	99.997	100.005
$C_3H_3D_3O$	C_3D_6O	100.035	100.038
C_6H_6	C_6D_6	99.650	99.692
C_7H_8	C_7D_8	99.881	99.897
C_7H_8	$^{13}C_7H_8$	99.926	99.929

section for acetone with two deuterated methyl group when compared to either of the two. The ion-neutral collision cross section of benzene and also toluene to a lesser extent seems to be significantly decreased due to deuteration. Knowing that the carbon−deuterium bond is typically shorter than the carbon−hydrogen bond[30] and since these molecules are quite symmetrical, this seems likely a plausible result. However, the ion-neutral collision cross section of toluene-$^{13}C_7$ is, albeit less, also decreased, indicating that this cannot be the sole explanation. Furthermore, it can be seen that all values derived from the air measurement yield slightly larger ion-neutral collision cross section ratios, indicating a systematic error in the calculation, most likely caused by the polarization limit approximation.

As a final sanity check for the presented results, the perdeuterated toluene measurements in nitrogen were repeated at lower drift voltages until no sufficient peak separation and signal intensities could be obtained any more. As shown in Figure 5, there is no shift in the observed relative drift time
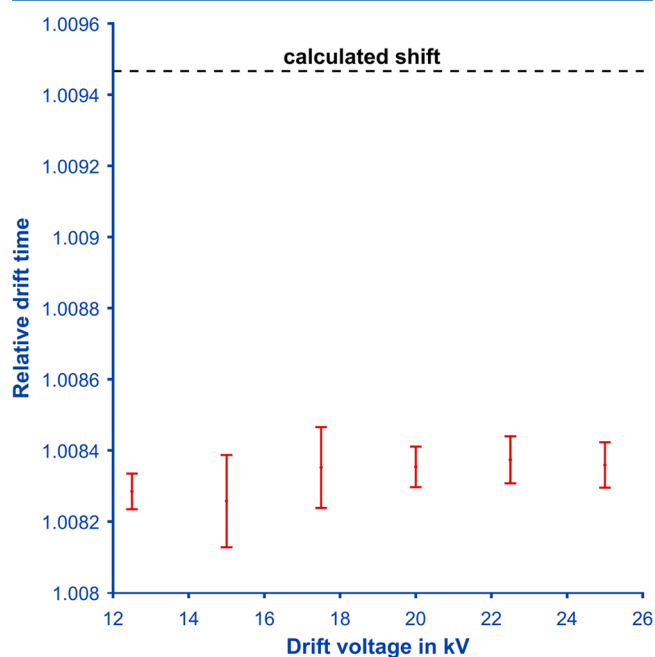

Figure 5. Relative drift time between toluene and perdeuterated toluene in nitrogen at different drift voltages. The error bars indicate 95% confidence intervals. No significant trend is observable.

even when the drift voltage is halved. This indicates that neither high-field effects due to the added energy from the electric field nor so-called start or end effects from the ion injection, which would add a flat time to the drift times, play a measurable role in this experiment. Combined with the already mentioned advantages of being a ratiometric measurement, which

eliminates all multiplicative errors, this gives us good confidence in the measured results.

CONCLUSION

In this work, it was shown that separation of isotopologues is possible using ultra-high-resolution ion mobility spectrometry and that the measured peak shifts are coherent with theoretical calculations. This enables the use of isotopically labeled substances both as tools for testing ion mobility equations and as markers in experiments. However, as a change in ion-neutral collision cross section was observed for some substances, mass measurement by ion mobility spectrometry is not yet possible for them, as this is based on the assumption of constant ion-neutral collision cross sections. Based on the current results, two routes should be followed to gain further understanding. On the one hand, measuring more substances with a possibly even higher resolving ion mobility spectrometer would significantly increase the so far scarce database. On the other hand, modifying existing software to perform structural and ion-neutral collision cross section calculations would allow both to gain better theoretical models (through knowing the change of the ion-neutral collision cross section between drift gases) and provide a theoretical estimate for the results (through estimating the ion-neutral collision cross section change between labeled and nonlabeled substances).

AUTHOR INFORMATION

Corresponding Author

*Fax: +495117623917. E-mail: kirk@geml.uni-hannover.de.

ORCID Ⓘ

Ansgar T. Kirk: 0000-0001-7152-3077

Author Contributions

All authors have given approval to the final version of the manuscript.

Notes

The authors declare no competing financial interest.

ACKNOWLEDGMENTS

REFERENCES

(1) Eiceman, G. A.; Karpas, Z.; Hill, H. H. *Ion mobility spectrometry*; CRC Press: Boca Raton, 2013.

(2) Eiceman, G. A.; Stone, J. A. *Anal. Chem.* **2004**, *76*, 390A−397A.

(3) Puton, J.; Namieśnik, J. *TrAC, Trends Anal. Chem.* **2016**, *85*, 10.

(4) Clemmer, D. E.; Jarrold, M. F. *J. Mass Spectrom.* **1997**, *32*, 577−592.

(5) Cohen, M. J.; Karasek, F. W. *J. Chromatogr. Sci.* **1970**, *8*, 330−337.

(6) Langevin, M. P. *J. Chim. Phys. Phys.-Chim. Biol.* **1905**, *5*, 245−288.

(7) Mason, E. A.; McDaniel, E. W. *Transport Properties of Ions in Gases*; Wiley-VCH Verlag GmbH & Co. KGaA: Weinheim, FRG, 1988.

(8) Siems, W. F.; Viehland, L. A.; Hill, H. H. *Anal. Chem.* **2012**, *84*, 9782−9791.

(9) Valentine, S. J.; Clemmer, D. E. *Anal. Chem.* **2009**, *81*, 5876−5880.

(10) Viehland, L. A. *Int. J. Ion Mobility Spectrom.* **2016**, *19*, 11−14.

(11) Viehland, L. A. *Int. J. Ion Mobility Spectrom.* **2016**, *19*, 1−10.

(12) Kaszycki, J. L.; Bowman, A. P.; Shvartsburg, A. A. *J. Am. Soc. Mass Spectrom.* **2016**, *27*, 795−799.

(13) Shvartsburg, A. A.; Clemmer, D. E.; Smith, R. D. *Anal. Chem.* **2010**, *82*, 8047−8051.

(14) Barnett, D. A.; Purves, R. W.; Guevremont, R. *Nucl. Instrum. Methods Phys. Res., Sect. A* **2000**, *450*, 179−185.

(15) Beucher, L.; Dervilly-Pinel, G.; Prevost, S.; Monteau, F.; Le Bizec, B. *Anal. Chem.* **2015**, *87*, 9234−9242.

(16) Viehland, L. A.; Lin, S. L.; Mason, E. A. *Chem. Phys.* **1981**, *54*, 341−364.

(17) Blanc, A. *J. Phys. Theor. Appl.* **1908**, *7*, 825−839.

(18) Mesleh, M. F.; Hunter, J. M.; Shvartsburg, A. A.; Schatz, G. C.; Jarrold, M. F. *J. Phys. Chem.* **1996**, *100*, 16082−16086.

(19) Shvartsburg, A. A.; Jarrold, M. F. *Chem. Phys. Lett.* **1996**, *261*, 86−91.

(20) Viehland, L. A. *Comput. Phys. Commun.* **2001**, *142*, 7−13.

(21) Viehland, L. A.; Chang, Y. *Comput. Phys. Commun.* **2010**, *181*, 1687−1696.

(22) Dunn, A. F. *Can. J. Phys.* **1964**, *42*, 1489−1498.

(23) Yousef, A.; Shrestha, S.; Viehland, L. A.; Lee, E. P. F.; Gray, B. R.; Ayles, V. L.; Wright, T. G.; Breckenridge, W. H. *J. Chem. Phys.* **2007**, *127*, 154309.

(24) Kirk, A. T.; Zimmermann, S. *Int. J. Ion Mobility Spectrom.* **2015**, *18*, 17−22.

(25) Glaskin, R. S.; Ewing, M. A.; Clemmer, D. E. *Anal. Chem.* **2013**, *85*, 7003−7008.

(26) Michelmann, K.; Silveira, J. A.; Ridgeway, M. E.; Park, M. A. *J. Am. Soc. Mass Spectrom.* **2015**, *26*, 14−24.

(27) Silveira, J. A.; Ridgeway, M. E.; Park, M. A. *Anal. Chem.* **2014**, *86*, 5624−5627.

(28) Hauck, B. C.; Siems, W. F.; Harden, C. S.; McHugh, V. M.; Hill, H. H., JR. *Rev. Sci. Instrum.* **2016**, *87*, 75104.

(29) Kirk, A. T.; Allers, M.; Cochems, P.; Langejuergen, J.; Zimmermann, S. *Analyst* **2013**, *138*, 5200−5207.

(30) Legon, A. C.; Millen, D. J. *Chem. Phys. Lett.* **1988**, *147*, 484−489.