Author's response

Interactive comment on "Addition of a fast GC to SIFT-MS for analyses of individual monoterpenes in mixtures" by Michal Lacko et al.

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Very nice work! I have a couple of questions:

1) What is the length of the second column that was used ("MXT-Volatiles")? Was it also 5 meters?

The length of both used columns (MXT-1 and MXT-Volatiles) was 5 meters. This was clarified in the revised manuscript.

2) Since the fast-GC part is so simple, would it be possible to enhance the time resolution of the measurement by using parallel fast-GC lines? Basically to inject gas pulses into parallel columns one after another and analyze them sequentially.

This is a very interesting suggestion. Sequential analysis, could be used to improve the time resolution of GC analysis down to several seconds. However, the hardware would have to be much more complicated and possibly artefacts due to the Nyquist Theorem could occur. Additionally, the idea of several parallel lines could simply be used to improve sensitivity by increasing the total flow of a sample whilest keeping the same quality of separation. This idea was added to the conclusions as indication for further work.

3) As I understand it, normally fast-GC is done using smaller inner diameter (0.15- 0.18 mm or even smaller) columns that what you have used here (0.25 mm). Can you comment why you used 0.28 mm columns and would it be possible to use a smaller I.D. column instead (to increase gas velocity)?

We have chosen the metallic column following the previous fast GC PTR-MS applications as it can be heated ohmically. Also the decreased I.D. would lead to a smaller flow rate limiting sensitivity. We have proposed in conclusions to use even wider column for increased flow rate.

Please note that there is a typo on page 2, line 15: "which can affects" -> "which can affect".

Thank you. We have corrected these.

Interactive comment on "Addition of a fast GC to SIFT-MS for analyses of individual monoterpenes in mixtures" by Michal Lacko et al.

Anonymous Referee #3

Received and published: 14 April 2019

The authors present the use of soft ionization mass spectrometry (SIFT-MS) combined with a fast-GC system in order to achieve separation and identification of different monoterpenes. The capabilities of two different columns are discussed. Furthermore, the potential use of different ionization modes when operating the SIFT-MS in order to better separate the monoterpene mixtures is suggested as a method to improve separation for this type of systems. After following the revisions suggested below, the publication should be suitable for AMT.

Specific comments

In the "abstract" and "summary and conclusions" sections of the manuscript, the achievement of quantitative analysis is suggested. This is not supported though by the main text and is even discussed that it's not the case by the authors on page 17, line 14. To my understanding, a quantitative analysis would provide ppb values of the individual monoterpenes together with their detection limits. On the contrary, only normalized intensity values are provided throughout the whole manuscript, for a mixture of monoterpenes that are not fully separated in the conditions used except in one case-study where the retention times are high (Fig. S3, 5V, retention time: 500s). It is therefore essential that the abstract and summary are re-written to avoid any misleading suggestion of quantification that overpromotes the presented work. The authors should work towards providing a more representative view of the manuscript that is related to the separation optimization of a monoterpene mixture using a low-resolution fast GC combined with the information obtained from differences in fragmentation patterns when using different ionization in the SIFT-MS.

This is an important comment we have thus extended the study by including quantitative method of calculation and by obtaining calibration curves from which actual LODs were calculated and these are given in the revised manuscript.

There is only one point in the manuscript where the authors discuss the detection limits of their technique that are as high as 100 ppb (page 17, line 15). How was that calculated? Did the authors perform calibrations for the individual monoterpenes? Where could this technique be applied with this high detection limits? I would expect that the values used in this study are not applicable to ambient field measurements since they are higher than any ambient observations. Comparison of this technique to other fast GC techniques shows differences in the limit of detection by orders of magnitude (page 17, line 15). As discussed in section 4.5, this technique is, therefore, inferior to others but could still be useful for identifying monoterpenes based on fragmentation. This should be the main part of the abstract and conclusions sections. This should be further discussed in the manuscript, especially since the authors attempt to publish in an atmospheric measurement technique journal.

Explicit measurements of LOD for our setup using MXT-Volatiles GC column were carried out and value the LOD is given in the revised text. The LODs were determined for α-pinene and R-limonene

from analysis of a calibration curve as three times the standard error of predicted intercept value divided by the slope of the calibration regression line. The LoD of fast GC with SIFT-MS is inferior about factor of 10 to fastGC PTR-MS. As it was demonstrated by analysis of coniferous samples, this can still be used for rapid analyses of stronger monoterpene sources. We have also proposed in conclusions that an additional solution for improving sensitivity is using multicolumn capable to achieve higher sample flow.

In order to obtain valuable information, the authors suggest that changing ionization in the SIFT-MS is recommended. This implies that in order to obtain valuable information relative to other techniques the GC-SIFT should run in both ionization modes. What would be the time needed to go through an H3O+ and a NO+ cycle? How much more is the time compared to other fast GC techniques that only run once and with better resolution (page 17, line 16)?

We have added this information to the revised manuscript.

Overall, I would recommend that the value of this work and the comparison of this technique to others should be further discussed and emphasized throughout the manuscript.

We have improved the discussion of the actual strengths and weaknesses in the conclusion.

Section 2 is hard to read and I would suggest restructuring. In the first sentence of the section the authors introduce Fig. 1 but this is not followed by a discussion of the figure, the instrument parts, and operation. On the contrary, they discuss the column options and operating details and then go through the temperature profiles. I would recommend the following structure: A. A discussion of the parts of the fast GC preseparation system and the modes of operation with their details that are discussed in section 2.1 and page 4, line 15 to page 5, line 4, B. Operating details together with columns of choice and temperature profiles.

We agree. We have changed the order so the article is more readable.

In section 3.1 a short discussion regarding the humidity dependences is presented that is not supported by any figure or graph. Was the humidity of the different samples measured? If so, shouldn't these values be provided in all figures, especially since the effects seem to be substantial? Furthermore, this paragraph and further discussion should be part of the results and discussions and not the section it is now.

Effect of humidity on ion chemistry is substantial, however water has much shorter elution time and thus reaches SIFT-MS before the monoterpenes. If the temperature of the column is not very high and water CG peak do not overlap with the monoterpene peaks, water influence on ion product ratio will be small. The edge of the water peak can be, however, still influential for fastest monotepenes as a-pinene or camphene. The issue was clarified and more details about influence of water on ion chemistry are now discussed in the revised manuscript.

Section 4.1 and 4.2 have an overlap of results and discussion that makes these sections hard to follow. I would recommend that the authors work towards restructuring these sections to a clearer presentation of the results that the table and figures promote followed by a detailed discussion, for each graph, for each column, and the comparison of the two columns. A characteristic example of the difficulty of the reader to follow the results and discussion is the title of section 4.2 that has little to do with what is discussed in it. Furthermore, please discuss why NO+ was not tested for the MTXVolatiles column.

Sections 4.1 and 4.2 were carefully reconstructed to be more logical and easy to follow.

Technical comments

Title and manuscript: change “analyses” to “analysis”

Corrected.

Page 1, line 19: change to “. . .to separate them in less than 180 s. . .”.

Corrected.

Page 2, line 15: change to “. . . which can affect human health. . .”.

Corrected.

Page 2, line 29: correct to “fast GC-PTR-ToF-MS” and in general correct throughout the manuscript “fastGC” to fast GC”.

We are deliberately distinguishing between the general „fast CG“ term describing the technique and „fastGC“ witch is a trademark name of the commercially available module for PTR-MS, produced by Ionicon (https://www.ionicon.com/product/accessories/fastgc) and used in the referenced studies as factGC-PTR-ToF-MS.

Page 2, line 33: change to “we report method development results aimed to. . .”.

Corrected.

Page 6, line 27: change to “are given in Table 1, and discussed in section 4”.

Corrected.

Page 6, line 28: This is hard to follow sentence. Rephrase.

The sentence was reformulated.

Page 6, line 30: Change to “reagent”

Corrected.

Page 8, line 2: change to “saturation vapor pressures”

Corrected.

Page 17, line 27: change citation style

Corrected.

Page 17, line 17: Which results? What are the authors comparing here?

We would like to compare limits of detection for SIFT-MS and PTR-MS setup. The sentence was clarified.

Page 18, line 22-23: “. . . allows analysis of mixtures of monoterpenes in the air in short time periods. . .” Is that the case for ambient measurements in the detection limits of the system? Isn’t this overpromoting the capabilities of the system?

For concentration above the detection limit (20 ppbv) is system able to analyse monoterpene mixtures. 60 s retention for MX-1 column as well as 180s for MXT-Vol column are considered as short time.

Table S1: It will be nice to add the m/z of detection.

We added the m/z information to the table.

Interactive comment on "Addition of a fast GC to SIFT-MS for analyses of individual monoterpenes in mixtures" by Michal Lacko et al.

Anonymous Referee #2

Received and published: 30 April 2019

The paper "Addition of a fast GC to SIFT-MS for analyses of individual monoterpenes in mixtures" by Lacko at all, submitted to AMT is work with potential. The addition of FastGC to the SIFT-MS is described in details, acknowledging most of the difficulties that method development brings. However, I find several major issues which prevent me from recommending its publication in AMT. First, the work is unnecessary long for the amount of information given. Two columns were compared, MXT-1 and MXT-Volatiles, but only the latter one gives acceptable separation. It is clear that MXT-1 is not suitable for this system (too fast separation, with not much control over the retention time). For any future version of the manuscript I suggest avoid the entire sections of MTX-1 column (perhaps it could be briefly mentioned in the supplementary data).

Second, the detection limit for this system is 100 ppb. Unfortunately, this is not close to the ambient levels of monoterpene concentrations or any plant chamber experiment loads. So, the relevance of this method is not within the scope of ATM, but rather in the fields where the technique can be used (monoterpene concentrations >100 ppb). Thus, I suggest to the authors to consider submitting these findings to a more suitable journal dealing with mass spectrometry techniques in general.

Minor comments: The manuscript in general needs more clarity: E.g. In the Abstract "the headspace of three conifer needle samples was analysed" it is not clear what do you mean here. The abstract should be clear and stand-alone. I believe you mean "needle samples of three conniver species"?

Thank you for pointing this out. The statement was clarified.

P4 L20. "(1 to 8 s)" is it 1 or 8 s you used? Or this is a range you can set? Again, I had to search in the following text to understand this better.

We clarified this information in the text so now it is stated what values were used in the measurements.

P8 L3-10. Not entirely clear enough. How did you enclose the plant branches? You mention temperature stress! But, how long it passed from the cutting? Did you use any light during the measurement?

We agree that this information can be important for description of a terpene emission. Plant branches were enclosed by wrapping the parafilm around the cut. Samples were measured app 30 minutes after harvest. Only a scattered light in the laboratory was presented. This information was added to the revised manuscript.

Interactive comment on "Addition of a fast GC to SIFT-MS for analyses of individual monoterpenes in mixtures" by Michal Lacko et al.

Anonymous Referee #1

Received and published: 7 May 2019

This work describes a GC-CIMS measurement technique developed to improve understanding of the composition of monoterpenes in the atmosphere which is an active area of interest in the atmospheric chemistry community due to key their roles in processes leading to formation of ozone and secondary organic aerosol (SOA) and is therefore highly relevant to the scope of AMT.

A series of experiments on individual standards of monoterpene isomers, monoterpene standard mixtures and the headspace of conifer foliage samples using a bespoke fast GC system coupled with a SIFT-MS is presented to demonstrate the potential application of fast GC-SIFT-MS for the separation and analysis of monoterpenes and other isomers in atmospheric and laboratory studies that is not currently achievable with SIFT-MS alone. The performance of two different GC columns in the fast GC SIFT-MS system was assessed - a generic (MXT-1) GC column and an application specific GC column (MXT-Volatiles). In addition, two reagent ions (NO+, H3O+) were used in the SIFT-MS system to aid in compound identification.

This work represents one of the first, if not the first, reported trial of a fast GC coupled with an SIFT-MS system which has a considerable user group worldwide. As noted in the manuscript introduction, this is an area of active development with previous papers describing fast GC coupled with other chemical ionization mass spectrometry (CIMS) systems, in particular PTR-MS (Materic et al 2015, Pallozzi et al 2016). Given the similarities between SIFT-MS and PTR-MS it could be considered that this paper does not represent a substantially novel development.

The original contributions to atmospheric measurement practice are:

1) The comparison of two GC columns - a generic (MXT-1) GC column (as used in previous fast-GC and GC-PTR-MS studies) and an application specific GC column (MXT-Volatiles) – this has relevance to the wider fast GC applications (SIFT-MS, PTRMS, other CIMS, fast GC-FID. . .) in which MXT-1 column has been used.

2) The first reported use of NO+ reagent ions in a fast GC - CIMS set-up.

However, additional additions/revisions are required for substantial conclusions to be reached regarding the performance and potential applications of fast-GC-SIFT-MS for quantification of monoterpene isomers. Specifically, more quantitative information is required on the detection limits, sensitivity and procedures for the quantification of species concentrations- see specific comments below.

Specific comments

Detection limit - p 17 Line 15 states "The present experiments indicate that using the fast GC-SIFT-MS combination, it is possible to achieve only qualitative analysis of the monoterpene mixture with a limit of the detection of about 100 ppb." Detection limits of 100 ppb is a major limitation for the application of fastGC-SIFTMS to measurements of individual monoterpenes in ambient air where concentrations are typically orders of magnitude lower (1 -10 ppb). The manuscript must include descriptions of:

1) How the stated detection limit of ~100 ppb was determined?

2) Why is this detection limit so high?

3) Potential improvements to the instrumental set-up that would reduce the detection limit to a range that would allow its application to measurements of ambient air (< 1 ppb).

All the raised questions are relevant are now discussed in the revised manuscript. The estimated LoD of 100 ppbv was not obtained by a proper calibration but just guessed. We have carried out additional determination of the LoD, using the MXT-Volatiles column. As the main limitation we identified the limited flow of the column witch decrease the sensitivity of the technique down to below 20 ppb. For higher temperatures the chromatograms have narrower peaks and LoD is much better (below 6 ppb). The limitation of SIFT-MS sensitivity depends on the total sample flow through the GC setup. This can be improved by using multiple parallel columns. All this discussion has been added to the revised manuscript.

Without these additions the application of this measurement technique for atmospheric measurements is limited making the relevance of this work to AMT highly questionable.

Quantification - The abstract, p 1 Line 18 states "Thus, it is possible to quantify components of a monoterpene mixture in less than 45 s by the MXT-1 column and to separate them in less 180 s by the MXT Volatiles column." Concentrations of monoterpenes are not quantified in this work and this claim is contradicted in the text p 17 Line 15 (as shown above) "it is possible to achieve only qualitative analysis of the monoterpene mixture". There are other similar contradictory statements in the manuscript which must be addressed.

This is again an important comment. Considering this and the comments of the other referee we have extended the study by including quantitative method of calculation and by obtaining calibration curves from which actual LODs were calculated and these are given in the revised manuscript.

Calibration – Wht is the sensitivity of this method? Was the system calibrated with certified gas standards containing one or more monoterpenes, and an empirical calibration factor determined?

We have carried out additional experiments to determine LoD and added that information to the revised manuscript. The sensitivity in the terms of retention times can be defined by the temperature profile, the length of the column and column type. The calibration was done using the diffusion tube method (Thompson and Perry, 2009) and the concentrations were determined by direct sampling SIFT-MS. This is now explained in Section 3.3

Absolute quantification - In lieu of an empirical calibration factor, the well-defined conditions in the SIFT-MS permit calculation of the concentrations of monoterpenes based on the raw signals of reagent and analyte ions (ie [m/z 137] as defined in section 3.2 of the manuscript), known reaction rates, and branching ratiosand instrument parameters as described in the SIFT-MS literature (e.g. Smith and Spanel 2005, Mass Spectrom. Reviews, 24, 661 – 700).

Yes, calculation of the concentrations u of monoterpenes sing the SIFT-MS is based on the raw signals of reagent and analyte ions. We have clarified this by adding a dedicated section dealing with quantification.

Direct measurement via SIFT-MS - Was direct quantification via SIFT-MS (without GC column) performed? Few comparisons of NO+ and H3O+ measurements of monoterpenes are available in the published literature and would be a valuable contribution.

Measurement of MS using the SIFT-MS was carried out and the results are now included in Supplement.

Both the detection limit and the sensitivity of the method are critical to understanding the application of this method for measurements of monoterpenes in the atmosphere and in laboratory studies. Neither are adequately described here making the relevance of this work to AMT highly questionable.

Relative abundance - In lieu of quantitative determination of individual monoterpene isomers, can the peak areas be used to estimate the relative abundance of each monoterpene species in the samples (mixtures and leaf headspace samples) ?

The peak areas be used, if separated, to estimate absolute concentration of each monoterpene. In not well separated chromatograms (as observed for MXT-1 column), absolute concentration cannot be properly estimated. However, using the additional regent ion, we can analyse coalesced peaks and determined in they do contain one or more monoterpenes. The discussion was improved to clarify these points.

Understanding the rel. abundance of monoterpenes is key to determining accurate calibration factors (see deGouw et al. (2003) JGR-Atmospheres 108, D21), and more importantly understanding the OH reactivity of BVOC dominated atmospheres. Suggest including NO+ and H3O+ reaction rates in Table 1 to demonstrate the importance of understanding the monoterpene composition to the accuracy of CIMS monoterpene measurements based on a single m/z, and adding a table of OH and O3 reaction rates for each monoterpene isomer identified and their relative abundance in leaf samples as well as some discussion regarding the potential contribution of different monoterpenes in the oxidation budgets of atmospheres dominated by emissions from these plant species. Overall, the measurement system and its operation are sufficiently explained however, inadequate information of the performance of this method in terms of detection limit and sensitivity are provided and potential future developments to improve performance are not adequately covered. Without this additional information the manuscript does not provide a substantial enough contribution to development of atmospheric measurement techniques for publication in AMT.

We have added the required information and we changed the revised manuscript to clarify the importance of the isomeric analysis of monoterpenes.

A key issue with CIMS instruments such as SIFT-MS and PTR-MS is essentially we know how much there is but we don't know what it is? Adding pre-separation techniques attempts to overcome this however, the data presented in this paper essentially reverses the challenge- we know what there is but not how much ? The manuscript requires a clear procedure for the quantification of monoterpene concentrations and/or the relative abundance of monoterpene isomers from the raw data in order to demonstrate the usefulness of this method over direct measurements with SIFT-MS. Quantification has been demonstrated in related instruments (Jones et al 2014, Materic et al 2015, Pallozzi et al 2016).and it is unclear why it was not part of this work.

Quantification method and its results are now discussed in the revised manuscript.

If these additions/revisions can be made, the following technical comments should also be considered.

Technical comments

Whole manuscript– replace SCI-MS with CIMS, the term chemical ionisation mass spectrometry (CIMS) is an established mass spectrometry term for analytical systems including SIFT-MS, PTR-MS etc

This project was funded from a EC project IMPACT involving 10 European institutions including those specialising in atmospheric research and the term Soft chemical-ionisation mass-spectrometry (SCIMS) is by consensus used to refer to SIFT-MS, PTR-MS and related techniques. Thus we prefer to keep SCIMS in this paper.

Abstract p1 line 18, change “quantify” to "qualitatively identify”

Corrected.

Abstract – add a couple of sentences at the end -what is the practical significance of this work? what is the theoretical significance?

P2 line 3, change “The analytical ion-molecule reactions” to “ The chemical ionisation reactions”

Corrected.

P2, line 13, suggest addition of a new paragraph discussing the fact that due to issues with stability of monoterpene mixtures in certified gas standards, CIMS instruments employed in ambient air studies are often calibrated with certified gas standards containing only one or two monoterpenes, (typically a-pinene). However the instrument response differs between isomers due to differences in their ionization reaction rates and branching ratios. To determine an accurate (weighted) instrument sensitivity value for monoterpenes, the relative abundance of monoterpene isomers must be known (see deGouw et al. (2003) JGR-Atmospheres 108, D21).

A short paragraph mentioning the calibration issues with monoterpene standards was added to main text.

P2 paragraph lines 13 – 21 – these concepts need to be re-visited in discussion and summary to demonstrate the usefulness of these techniques.

We have added the discussion to the conclusion.

P2 line 21, move these two sentences into subsequent paragraph “Gas chromatography mass spectrometry (GC-MS) coupled with pre-concentration techniques has been developed to successfully identify and quantify different atmospheric monoterpenes (Janson, 1993; Räisänen et al., 2009; Song et al., 2015). However, the requirements of pre-concentration and long cycle time (more than 1h) are obviously unsuitable for real-time measurements.”

Both sentences were moved into a subsequent paragraph

P4, “It is interesting to note that the flow of sampled air, established by the pressure difference between ambient atmosphere and the low pressure of the SIFT-MS flow tube, changes with the column temperature due to the variation of the dynamic viscosity of the air (see Fig. 2).” – Does this affect flow tube residence time (reaction time, t) important in SIFT-MS quantification calculations?

It is affecting the total sample flow through the system and thus the calculation of quantifications. This effect can be estimated and included to the quantification calculation.

P4, line 16, Can measurements by the SIFT-MS when the GC set-up is in “normal mode” be considered an instrument zero (SIFT-MS instrument background)? Can you use this data to calculate the detection limit and subtract from “sampling mode” measurements?

Yes, during the „normal mode“ we can measure the background signal for selected masses, which can be considered as instrument zero. This value is usually found to be negligible; therefore, we don’t

have to subtract it. Information about detection limit obtained from calibration curves by the 3 sigma method was added to the revised manuscript.

P5, line 16- "Sampling was repeated several times to improve sensitivity." No data for sensitivity is presented.

We agree, we changed the sentence.

P5 Section 3- insert details on the time it takes to switch between reagent ions and to achieve stable ion signals- this is crucial if NO+ and H3O+ are to be used for compound identification. What was the intensity and purity of the reagent ion signals?

We agree. Information was added to the revised manuscript. Switching between regent ions is very fast and require only tens of milliseconds. Purity of reagent ions is defend by the injection quadrupole, level of parasite ions is usually below 1%. Count rate of primary ions is usually in range of one million.

P7 insert section (after section 3.2) describing quantification procedure (as discussed in specific comments above) either using empirically derived calibration factors or via absolute quantification procedure based on [m/z 137] for H3O+ mode; and [m/z 136] for NO+ mode.

We added a new section (Section 3.3) discussing the quantification.

P8 Section 4.1 Comparison of columns: MXT-1 vs MXT-volatiles. The comparison of these two columns is valid given the use of the MXT-1 column in related instruments presented in the published literature (Jone et al 2014, Pallozzi et al 2016, Materic et al 2015 etc).

We have discussed this in the revised manuscript.

P8 paragraph line 12 -18 – Your approach needs to be more clearly articulated – for instance, firstly the instrument response to individual monoterpene species, in terms of retention time, and product ion ratios, was characterized via analysis of a series of prepared standards with both the MXT1 and MXT volatile columns and when H3O+ and NO+ were employed as the primary reagent ion in the SIFT-MS. Secondly, the separation of monoterpenes isomers using two columns, and the two reagent ions (NO+, H3O+) was demonstrated through analysis of prepared mixtures containing 8 monoterpenes. Lastly, the application of the GC-SIFT-MS for the separation (and quantification?) of monoterpene isomers in a real-world analysis is presented in a series of leaf headspace analyses.

The initial paragraph was updated and clarified according to proposed schema.

Section 3.3, Note it is unclear whether the same individual standards and mixtures of monoterpene were analysed by both NO+ and H3O+ in the same analysis runs?

The same mixture was used and this is now stated in the revised manuscript.

P8 line 22 – "Whilst the retention times for individual monoterpenes are different, they are not sufficiently stable (fluctuate by > 1 s, see Table 1) in the present fast GC device for analyses based on retention time only to be reliable." Suggested improvements to instrument design?

The fluctuation of retention may be caused by the fluctuation of the column temperature and therefore for longer column and lower temperature it may be reduced. (Effect will be less significant for longer retention times). This is now discussed in conclusion.

P8 line 28, the following statement is unclear "the peak shapes cannot be compared directly but the peak width (FWHM) increased only two times for the MXT-Volatiles column". Also define FWHM.

The sentence was removed from the text.

P9, Table 1 – add columns for reaction rates of monoterpenes with NO+ and H3O+ - consider landscape page layout (see comment above re Relative Abundance)

We have decided to keep the rate constants for the interaction of monoterpenes with regent ions in the Supplementary, together with the full list of potential products. The use of rate constant in the calculation of concentrations depends on all secondary ions produced from initial proton transfer, charge transfer of association interaction.

P11 Section 4.1 – Discussion of response to individual monoterpene standards. Insert Figure S2 and a corresponding plot for the MXT-volatiles column into section 4.1. These are very helpful when interpreting subsequent Figures 3 and 4. What conclusions can be reached from the tests of individual monoterpene standards – based on these tests what peaks are likely to co-elute, and what peaks are likely to be able to be separated in analysis of an unknown mixture? These tests provide the fundamental information for interpretation of the data from mixtures and leaf samples and should be included in the main text.

The plots of the response to individual monoterpene standards are quite busy and thus it is more clear to present them in the Table 1. Data are additionally directly shown in Fig 4 (bottom part) as horizontal lines, showing position of each monoterpene standard. For MXT-Vol column the identification is apparent. The discussion regarding identification of monoterpenes is clear in identification of monoterpene mixture, where for MXT-1: peak A is due to co-elution of α-pinene, camphene and myrcene. Peak B is due to the presence of β-pinene exclusively and peaks C and D are due to the remaining four monoterpenes, mainly 3-carene and R-Limonen. We hope that the revised manuscript makes all of this clear.

P10 line 10 “As observed for both columns, separation can be improved by decreasing the column temperature (see Fig. S3 in the Supplement), however this may increase the chromatogram width and thus decrease the sensitivity of the technique. Additional sensitivity can be achieved by increasing the injection time, which will, however, increase the peak width.” – this discussion is not quantitative, no explicit sensitivity data is presented.

Unfortunately, we do not have quantitative data to demonstrate effect of injection time on sensitivity. The statement was corrected in revised manuscript.

The discussion in Section 4.1 regarding analysis of mixtures needs to be restructured.:

1) provide a direct comparison between MXT-1 and MXT-volatiles at the same conditions. (~40- 45C). Figures 3 and 4 – Figure 3 is actually a comparison of H3O+ and NO+ and the data from the MXT1 and MXT-volatiles column are not compared side-byside. Format a page in landscape orientation, combine figures 3 and 4 (three panels) and present them in a compatible format (ie same formatting and labelling etc).

Direct side-by-side comparison of both columns is now given in the revised manuscript at room temperature and at 40C.

2)Discuss challenges and potential improvements ie stability in retention times, improved separation via decreasing column temp, improved sensitivity by increasing injection times.

Discussion regarding stability and separation is clarified in the revised manuscript.

3)Present MXT-volatiles column data under optimized conditions – ie "The MXTVolatiles column facilitates identification of all monoterpenes present in the mixture for temperatures close to room temperature (see Fig. S3 in the Supplement)." – the top panel in the S3 plot is key to demonstrating the achievable separation of the MXTvolatiles column - move it from the supplement to the main body. The additional species identifiable using this technique compared to the MXT-1 set-up need to be more clearly summarised.

The room temperature data are compared side-by-side fir both columns. The discussion of the comparison of the two columns is improved by reorganisation of the sub sections.

P12 Paragraph lines 8 – 17- needs to be moved to later in the discussion or into section 4.5 to show that aside from potentially better selectivity other co-benefits of employing the NO+ reagent ion in CIMS measurements of BVOCs, in particular in measurements of isoprene (See Karl et al 2012 ACP 12:11877 – 11884, and Karl et al 2014 Int J. Mass Spectrom. 365-366:15-19). There are many more species which interfere with quantification of isoprene in H3O+ reagent ion mode such as furan, 2,3,2-MBO, C5 aldehydes.

Discussion regarding benefits of NO+ reagent ion was extended to discuss potential benefit of identification of isoprene.

P12 line 19 " However, the ratios obtained for α-pinene and myrcene are somewhat variable between the FS and MIM data and they also differ somewhat from the literature values." – be quantitative ie state % variability. Is the variability a result of changes in the reagent ion intensity (consider using normalised intensity), or composition (eg % reagent ion impurities of H3O+(H2O), O2+, NO+)?

The ion ratio is not dependent of the ion intensity or ion impurities, however, it can be affected by secondary processes with neutral water molecules or hydronium hydrates. The issues of water influence were clarified in the text. Changes of the ion ratio are now discussed in the revised manuscript.

P14 Section 4.3 –For this method to be useful in atmospheric research the concentrations of monoterpene isomers or an estimate of their relative abundance must be quantified from the data and presented here and section 4.4(see specific comments above re quantification).

Re-analysis of data was carried out to obtain concentrations of detected monoterpenes and thus demonstrate usefulness of the technique in atmospheric research.

P14 Section 4.4- be consistent – use dot point format as for previous section. Why is the data from non-optimized conditions (40C) presented? Was the analysis done at the optimal temperature (5V) for separation? If so, should be presented.

Dot point format is now used for this section in the revised manuscript. Analysis was carried out using 40C conditions only as we would like to compare MXT-1 and MXT-Vol columns at same conditions. Conditions optimal for separation are not applicable for SCI-MS techniques, as the separation need 700s to be provided. At temperature 40C is sufficient separations provided under 180 s.

P14 line 14, " The signal increase in the third region may indicates trace presence of (R)-(+)-limonene." – the m/z81 signal or the ion intensity?- not clear.

P15 Section 4.4- need to state that similar experiments but on a different series of conifer samples were also conducted using the MXT-volatiles column.

The information was added in the revised manuscript.

P15 Figure 5- consistent units (normalised intensity) should be used for all figures (3-6), label peaks in both H3O+ and NO+ chromatograms (both Fig 5&6). Query the signal to noise ratio of some of the identified peaks e.g. H3O+ spruce 3-carene / limonene peak. Re-iterates importance of quantifying method LoD.

Labelling in figures was modified. The observed fluctuation/variation in Fig 5-6 is caused by real signal representing presence of monoterpenes. The background intensity is close to 0. The signal to noise ratio is more than 300.

P17 Section 4.4 – This section should conclude with a table of the relative abundance of each monoterpene isomer in the leaf samples and their reaction rates with OH and O3 with associated discussion.

The calculated absolute concentrations of detected monoterpenes were added to the text. Table shoving the OH and O3 reaction rates with monoterpenes was created and include in the introduction. Discussion regarding importance of monoterpene separation and their different OH and O3 reactivity was added to conclusions.

P17 Section 4.5 – "The present experiments indicate that using the fast GC-SIFT-MS combination, it is possible to achieve only qualitative analysis of the monoterpene mixture with a limit of the detection of about 100 ppb. This is inferior to the previously described fastGC-PTR-MS systems (Materic et al., 2015; Pallozzi et al., 2016), which ´ achieved full separation with limit of the detection up to 1-2 ppt." – list the reasons for the difference in performance and potential future developments of the GC-SIFT-MS method to improve performance. This statement must be addressed in more detail as these significant limitations preclude the application of this method to ambient studies and make the inclusion of this work in AMT questionable.

We have carried out new experiments and obtained LOD from calibration curves. Ppt was a mistake, in Materic et al., 2015; Pallozzi et al., 2016 1-2 ppb was achieved.

P17 line 17 – start new paragraph at "However, one advantage of SIFT-MS is the facility to use two reagent ions, and the analysis of product ion ratios provides additional information. Thus, the combination of the data from the two reagent ions together with the analyses of the product ion signal ratios ri can be shown to improve the identification of monoterpenes." – be more specific, what additional compounds were identified using the reagent ion chemistry. Suggest insert discussion from 4.2, on usefulness of NO+ reagent ion for identification of other BVOCs here. As a side note, switchable reagent capability has been developed for PTR-MS and other CIMS and is not unique to SIFTMS.

Myrcene and camphene were stated as examples of monoterpenes that benefit from use of NO+ regent ion. The discussion was moved from section 4.2.

P17 line 20 – "The results obtained from the present study agree well with the literature reports." Be more specific, suggest – the results obtained from the analysis of leaf headspace samples agree well other studies in the published literature. Suggest authors present comparisons by tree species as a table with following columns plant species name; monoterpenes identified; rel. abundance where available; measurement method; time resolution; and where available: LoD & sensitivity; and literature reference. Focus discussion on number and rel. abundance of monoterpenes identified and the methods used, not on geographical variability or variability between species beyond the scope of this work. What is the potential advantage of this method over others? Time resolution?

The detail analysis and detail comparison between plants and their monoterpene concentration is behind the scope of this publication. We do not focus on geographical origin of samples. It is well known that concentration and composition of monoterpenes is very sensitive to the sampling technique. We have edited this section to discuss percentages rather than absolute concentrations.

P18 Section 5- "A new method has been developed that allows quantitative analyses of individual monoterpenes in mixtures using SIFT-MS enhanced by chromatographic preseparation." As previously stated this is not correct and contradicts the first line of the previous section (4.5) "The present experiments indicate that using the fast GC-SIFTMS combination, it is possible to achieve only qualitative analysis of the monoterpene mixture with a limit of the detection of about 100 ppb."

P18 line 16 start new paragraph at "A weakness of the current fast GC setup is the relatively poor temperature stability caused by a strong dependence on the laboratory ambient temperature. . .."

Corrected.

P18 line 18 "It has been shown that a clear advantage of SIFT-MS is the facility to use different reagent ions and to utilize the ratios of the specific product ions of their reactions with the various monoterpene isomers at the same retention time to improve the identification of the monoterpenes." Belongs in previous paragraph (P18, line 10).

We have rearranged the order.

P18 line 23 – "This novel idea of a fast GC-SIFT-MS combination could broaden the application of SIFT-MS to in situ trace gas analyses of complex mixtures such as ambient air and exhaled breath.". There are several issues with this statement: 1) SIFT-MS is already used for in situ ambient air and breath analysis- this technique GC-SIFTMS does not broaden its application. The practical significance of this work is that it aims to address the challenge of quantifying isomers in CIMS measurements of complex mixtures. 2) Also, need to preface this statement "With improved limits of detection and sensitivity, this novel fastGC-SIFT-MS could. . .. ." currently its application in ambient air analysis is limited due to high LoD and lack of data about its sensitivity.

We have rewritten the conclusion with these points in mind so it truly represents the outcome of this work including the additional LOD determinations.

What is the theoretical significance of this work- what will an improved understanding of the complex mixture of monoterpenes contribute to our understanding of atmospheric chemistry? Ie estimates of total OH reactivity etc.

The detailed answer to this is outside of the scope of this paper, nevertheless we hope that the revised manuscript indicates the further direction in fast GC SIFT-MS development.

Addition of a fast GC to SIFT-MS for ~~analyses~~analysis of individual monoterpenes in mixtures

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Abstract. Soft chemical ionization mass spectrometry (SCI-MS) techniques can be used to accurately quantify volatile organic compounds (VOCs) in air in real time; however, differentiation of isomers still represents a challenge. A suitable pre-separation technique is thus needed, ideally capable of analyses in a few tens of seconds. To this end, a bespoke fast GC with an electrically heated 5 m long metallic capillary column was coupled to selected ion flow tube mass spectrometry (SIFT-MS). To assess the performance of this combination a case study of monoterpene isomer ($C_{10}H_{16}$) analyses was carried out. The monoterpenes were quantified by SIFT-MS using H_3O^+ reagent ions (analyte ions $C_{10}H_{17}^+$, *m/z* 137, and $C_6H_9^+$, *m/z* 81) and NO^+ reagent ions (analyte ions $C_{10}H_{16}^+$, *m/z* 136, and $C_7H_9^+$, *m/z* 93). The combinations of the fragment ion relative intensities obtained using H_3O^+ and NO^+ were shown to be characteristic for the individual ~~monoterpens~~monoterpenes. Two non-polar GC columns (Restek Inc.) were tested: the advantage of MXT-1 was shorter retention whilst the advantage of MXT-Volatiles was better separation. Thus it is possible to ~~quantify~~identify components of a monoterpene mixture in less than 45 s by the MXT-1 column and to separate them in less than 180 s by the MXT-Volatiles column. Quality of separation and sensitivity of present technique (LOD ~16 ppbv) was found to be inferior compared to commercially available fast-GC solutions coupled with proton transfer reaction mass spectrometry (PTR-MS, LOD ~1 ppbv) due to the limited sample flow through the column. However, using combinations of two reagent ions improved identification of monoterpenes not well resolved in the chromatograms. As an illustrative example, headspace of ~~three conifer~~ needle samples of three conifer species was analysed by both reagent ions and with both columns showing that mainly α-pinene, β-pinene and 3-carene were present. The system can thus be used for direct rapid monitoring of monoterpenes above 20 ppbv. Limitation of the sensitivity due to the total sample flow can be improved using a multicolumn pre-separation.

1 Introduction

Standard analytical methods used to identify and quantify volatile organic compounds (VOCs) in air, such as thermal desorption gas chromatography mass spectrometry (TD-GC-MS), are often time consuming and cannot be used to investigate temporal changes in chemically evolving systems. In contrast, soft chemical ionization mass spectrometry (SCI-MS)

techniques, such as selected ion flow tube mass spectrometry (SIFT-MS) (Smith and Španěl, 2011a; Španěl et al., 2006) and proton transfer reaction mass spectrometry (PTR-MS) (Lindinger et al., 1998; Ellis and Mayhew, 2013; Smith and Španěl, 2011b) represent well-established real time tools to analyse a wide variety of VOCs in ambient air (Amelynck et al., 2013; de Gouw and Warneke, 2007; Malásková et al., 2019; Rinne et al., 2005; Schoon et al., 2003) and in headspace of biological samples (Shestivska et al., 2015; Shestivska et al., 2011; Shestivska et al., 2012). The advantage of SIFT-MS and PTR-MS lies in the possibility of online, real-time analysis obviating sample collection and pre-concentration of VOCs. In these techniques, defined reagent ions (usually H_3O^+, NO^+ or $O_2^{+\bullet}$) interact with trace VOCs present in gas samples introduced into a flow tube or a flow/drift tube. The ~~analytical ion molecule~~chemical ionisation reactions that produced analyte ions are variously proton transfer, adduct ion formation, charge transfer and hydride ion transfer, principally depending on the type of reagent ions used. This ion chemistry has been thoroughly reviewed in a number of publications (Smith and Španěl, 2005). These ion-molecule reactions are not greatly exothermic and so few product (analyte) ions are produced in each reaction, often just one or two, that can be readily identified. However, chemically similar molecules with the same atomic composition (structural isomers) usually produce identical analyte ions with similar branching ratios and therefore the neutral analyte molecules cannot be easily differentiated using SCI-MS alone (Smith et al., 2012). However, the reactions of the isomeric molecules may have different rate coefficients with the different reagent ions and lead to product ions at recognisably different branching ratios depending on their molecular geometry (Jordan et al., 2009; Pysanenko et al., 2009; Španěl and Smith, 1998; Wang et al., 2003). So the concurrent use of the available reagent ions in SIFT-MS analysis can sometimes be used to analyse and identify particular isomers.

Monoterpenes, mostly emitted from plants, are very important biogenic volatile organic compounds (BVOCs) in the atmosphere. Due to their high reactivity with atmospheric oxidants such hydroxyl radicals ($OH^\bullet$), monoterpenes reactions can lead to tropospheric ozone (O_3) accumulation as well as to secondary organic aerosol formation, which can ~~affects~~affect human health and contribute to global climate change (Chameides et al. (1992); Fehsenfeld et al. (1992); Kulmala et al. (2004)). Although all monoterpenes comprise two isoprene units and have the same molecular formula, $C_{10}H_{16}$, their reactivity (or lifetime) for reaction with $OH^\bullet$ and O_3 widely varies from minutes to days (Atkinson and Arey, 2003~~).~~) (See Table 1). The values of the net BVOC/$OH^\bullet$ reactivity measured in rainforests have been found to be higher than expected, which could be attributed to undetected monoterpenes or sesquiterpenes (Nolscher et al., 2016). Therefore, it is important to identify and individually quantify these BVOCs at their ambient trace levels.

Quantitative measurement of monoterpenes is often problematic due to the problems with stability of monoterpene mixtures in certified gas standards (Rhoderick and Lin, 2013). Therefore, fresh individual monoterpene standards or monoterpene mixtures are prepared from liquid standards. To determine an accurate instrument sensitivity to individual monoterpenes, the relative abundance of monoterpene isomers must be known (de Gouw et al., 2003).

Table 1. Monoterpenes included in the present study listed together with their atmospheric lifetimes and reactivities.

Compound	Lifetime for reaction with a [a]	Chemical lifetime[d]		Rate constant of O_3[e]	Rate constant of OH[f]
	OH[b] / O_3[c]	*Day*	*Night*		
α-pinene	2.6 hrs 4.6 hrs	2-3 hrs	5-30 min	8.7	5.45 ± 0.32
β -pinene	1.8 hrs 1.1 day	2-3 hrs	5-30 min	1.5	7.95 ± 0.52
Camphene	2.6 hrs 18 day	nd	nd	9.0[g]	5.33[g]
Myrcene	39 min 50 min	40-80 min	5-20 min	49	21.3 ± 1.6
3-carene	1.6 hrs 11 hrs	nd	nd	3.8	8.70 ± 0.43
R-limonene	49 min 2.0 hrs	40-80 min	5-20 min	21	16.9 ± 0.5
α-terpinene	23 min 1 min	< 5 min	< 2 min	870	36.0 ± 4.0
γ-terpinene	47 min 2.8 hrs	nd	nd	14	17.6 ± 1.8

[a] taken from Atkinson (Atkinson and Arey, 2003) unless noted otherwise.
[b] Assumed OH radical concentration: 2.0×10^6 molecule cm^{-3}, 12-h daytime average.
[c] Assumed O_3 concentration: 7×10^{11} molecule cm^{-3}, 24-h average.
[d] Lifetimes are estimated in relation to [NO_3] = 10 ppt, [O_3] = 20 ppb for night; and [OH] = 10^6 molecules per cm^3, [O_3] = 20 ppb for day light conditions. (Kesselmeier and Staudt, 1999) (unless noted otherwise)
[e] Rate constants (in units of 10^{-17} cm^3 $molecule^{-1}$ s^{-1}) for the gas-phase reactions of O_3 with a monoterpenes have been determined at 296 ± 2 K and 740 torr total pressure of air or O_2 using a combination of absolute and relative rate techniques. (Atkinson et al., 1990) (unless noted otherwise)
[f] Rate constants (in units of 10^{-11} cm^3 $molecule^{-1}$ sec^{-1}) for the gas-phase reactions of the OH radical with monoterpenes have been determined in one atmosphere of air at 294 ± 1 K. (Atkinson et al., 1986) (unless noted otherwise)
[g] Rate constants of k(OH + isoprene) = 1.01×10^{-10} cm^3 $molecule^{-1}$ s^{-1}. O_3 reaction rate constants determined in 10^{-19} cm^3 $molecule^{-1}$ s^{-1} units. OH radical reaction rate constants determined in 10^{-11} cm^3 $molecule^{-1}$ s^{-1} units. (Atkinson et al., 1990)
nd – no data

Gas chromatography mass spectrometry (GC-MS) coupled with pre-concentration techniques has been developed to successfully identify and quantify different atmospheric monoterpenes (Janson, 1993; Räisänen et al., 2009; Song et al., 2015). However, the requirements of pre-concentration and long cycle time (more than 1h) are obviously unsuitable for real-time measurements.

A promising approach to the near real time analysis of isomeric molecules is to combine both SCI-MS and fast GC methods. Pre-separation provided by fast GC involves short columns with thin active layers, fast temperature ramps, fast injection systems and time resolutions below 5 min (Matisová and Dömötörová, 2003). Materic et al. (Materić et al., 2015) established a system using PTR-MS coupled with a fast GC to detect individual monoterpenes and achieved the separation of six most common monoterpenes at a limit of detection down to 1~~.2~~ ppbv. Pallozzi et al. then compared a fastCG-PTR-ToF-MS system

with traditional GC-MS methods, discussing the limitations of the fast GC setup on some BVOCs emitted from plants, including monoterpenes (Pallozzi et al., 2016). SIFT-MS is also widely used in VOCs analyses (Allardyce et al., 2006; Smith and Španěl, 2011b, 2005b). It has well-defined analytical reaction conditions and the H_3O^+, NO^+ and $O_2^{+\bullet}$ reagent ions can be switched rapidly to analyse time-varying trace gases in air samples. In the present article, we report ~~the results of~~ method development results aimed ~~at~~to selective analyses of individual monoterpenes in mixtures in air using a bespoke fast GC/SIFT-MS combination with H_3O^+ and NO^+ reagent ions. This involved the analysis of both prepared laboratory monoterpene/air mixtures and headspace of the foliage of different pine trees.

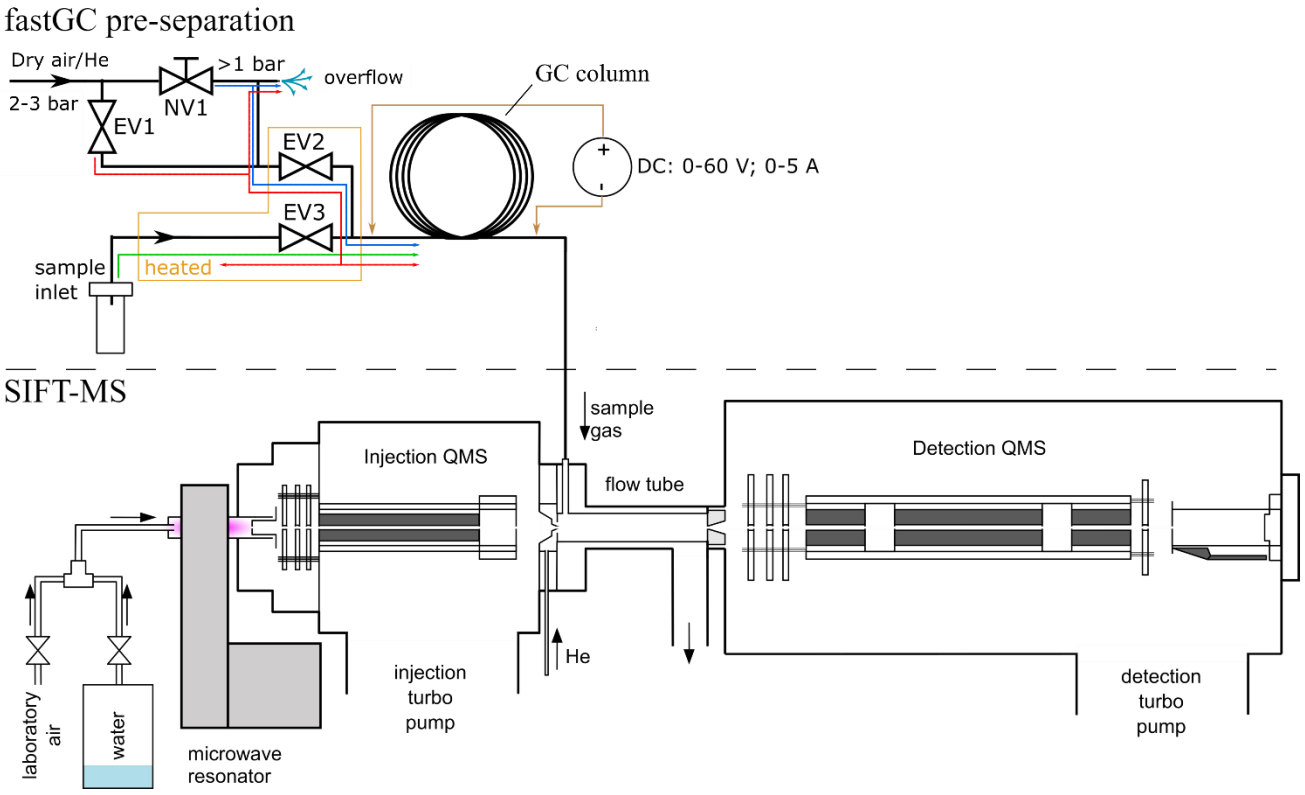
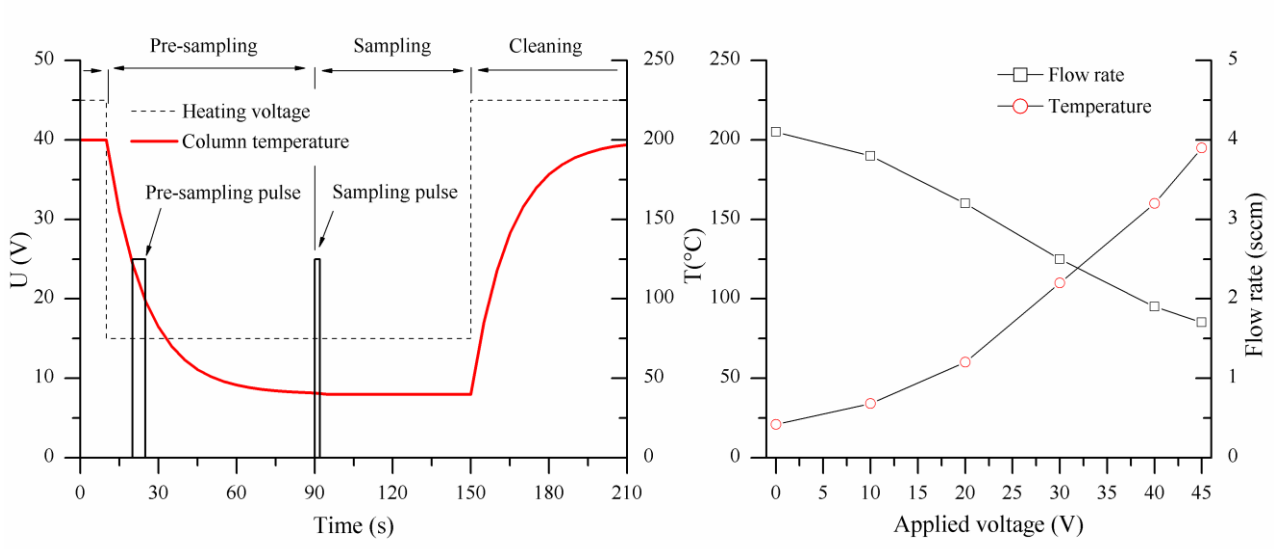


Figure 1: Schematic visualization of the fast GC-SIFT-MS experiment. Coloured dashed lines in the inlet part of the ~~fastGC~~fast GC represent gas flow through the system of the valves EV1-3. The blue line traces the "normal mode" regime, the green line represents the "sampling mode" and the red line represents the "cleaning mode".

2 Construction of a fast GC device for pre-separation

The experimental setup of the bespoke fast GC setup constructed as an addition to SIFT-MS is shown in Fig. 1. ~~In the experiments, two different GC columns were tested. First, a 5 m long nonpolar general purpose chromatography metallic column MXT-1 (0.28 mm × 0.1 um active phase, Restek Inc.) using dry air as the carrier gas, which was chosen according to the previous PTR-MS fastGC analyses (Romano et al., 2014). Additionally, a second, application-specific column for volatile~~

~~organic pollutants, MXT-Volatiles (0.28 mm × 1.25 um active phase, Restek Inc.), was used with helium carrier gas. In order to facilitate direct resistive heating, the coil-shaped stainless steel columns (resistivity ~4.2 Ω/m) were electrically isolated and connected to a regulated 60 V, 5 A DC power supply. Appearance of cold spots was suppressed by ensuring that the electrical current runs through the entire length of the columns. The temperatures of the columns were monitored by a K-type probe connected to their centres (see the right part of Figure 2 for the temperature variation with applied voltage). It is interesting to note that the flow of sampled air established by the pressure difference between ambient atmosphere and the low pressure of the SIFT-MS flow tube changes with the column temperature due to the variation of the dynamic viscosity of the air (see Fig. 2).~~



~~**Figure 2: Left: the applied heating voltage (dashed) and the temperature profile of the column (red) during the fast GC cycle. The pulses indicate the opening of the valve EV3 during the pre-sampling and the sampling periods. Right: The increase of the column temperature and the related decrease of the carrier gas flow rate with the heating voltage.**~~

The routing of the sample and the carrier gases was controlled by solenoid valves (Parker VSONC-2S25-VD-F, < 30ms response), labelled in Fig. 1 as EV1, EV2 and EV3. The needle valve NV1 was used in combination with an overflow relieve tube to fine-adjust the flow rate of the carrier gas (20-50 sccm from a gas cylinder regulator set to about 2 bar) so that the air pressure at the column entrance is held just above ambient. The region of the sampling input line, EV2, EV3 and their connection with the column are permanently heated to ~60 °C to prevent adsorption of sample gas/vapour and to reduce memory effects.

Three modes of gas flow are possible as illustrated in **Fig. 1**:

- The **"normal mode":** EV2 is open and both EV1 and EV3 are closed. Carrier gas flows through NV1, partly vented via the overflow relieve but mostly into the column. The pressure at the column entrance is just above the ambient atmosphere and a constant flow rate of clean carrier gas (synthetic air or helium) is thus achieved.

- The **"sampling mode"**: EV1 and EV2 are closed and EV3 is open. Sample air is introduced into the column in a short time (1 to 8 s) after which the "normal mode" is resumed.
- The **"cleaning mode":** All valves are open and the carrier gas taken directly from the cylinder regulator is introduced into the column (higher than normal flow) and purges the sample line via EV3. The overflow relieve flow rate is not sufficient to diminish the pressure.

The modes can be switched either manually or controlled from the SIFT-MS software.

~~2.1 The fast GC operation~~

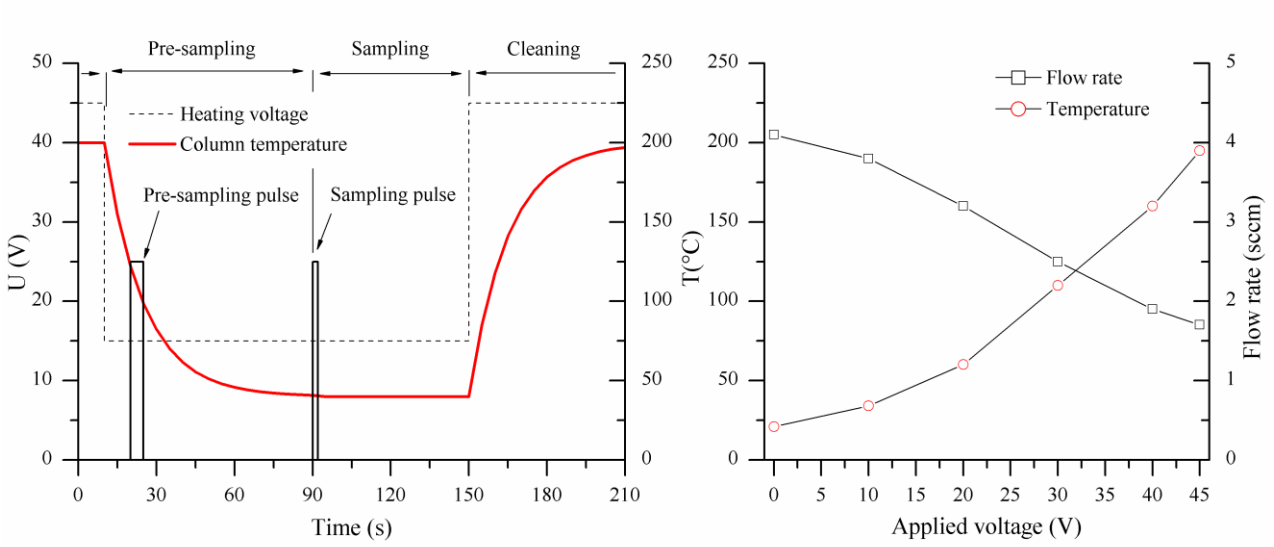


Figure 2: Left: the applied heating voltage (dashed) and the temperature profile of the column (red) during the fast GC cycle. The pulses indicate the opening of the valve EV3 during the pre-sampling and the sampling periods. Right: The increase of the column temperature and the related decrease of the carrier gas flow rate with the heating voltage.

The operation sequence for air analysis is as follows: A column is first heated up to 200 °C in the "cleaning mode" for three minutes prior to commencing the "normal mode" with an appropriate heating voltage setting (e.g. ~~10~~15 V as shown in Fig. 2). Whilst the column cools down, a pre-sampling interval (8-10 s "sampling mode", see ~~Figure~~Fig. 2) is applied in order to refill the "dead volume" comprising the EV3 valve and the sampling inlet by air at its entrance. After the column reaches working temperature and a steady flow of clean carrier gas is established, the sample for actual analysis is introduced by enabling the "sampling mode" for ~~1 to 8 s~~selected amount of time. ~~. The GC separation then takes place over typically 60 – 300 s whilst the eluent is continuously analysed by SIFT-MS. It is possible to apply a heating ramp during this period.~~

In the experiments, two different GC columns were tested. First, a 5 m long nonpolar general-purpose chromatography metallic column MXT-1 (0.28 mm × 0.1 um active phase, Restek Inc.) using dry air as the carrier gas, which was chosen according to the previous PTR-MS fastGC analyses (Romano et al., 2014). Additionally, a second, application-specific column for volatile

organic pollutants, MXT-Volatiles (0.28 mm × 1.25 um active phase, Restek Inc.), was used with helium carrier gas. In order to facilitate direct resistive heating, the coil-shaped stainless steel columns (resistivity ~4.2 Ω/m) were electrically isolated and connected to a regulated 60 V, 5 A DC power supply. Appearance of cold spots was suppressed by ensuring that the electrical current runs through the entire length of the columns. The temperatures of the columns were monitored by a K-type probe connected to their centres (see the right part of Figure 2 for the temperature variation with applied voltage). It is interesting to note that the flow of sampled air established by the pressure difference between ambient atmosphere and the low pressure of the SIFT-MS flow tube changes with the column temperature due to the variation of the dynamic viscosity of the air (see Fig. 2). This effect can to be estimated and have to be included to a quantification calculation.

In the initial tests with the first generic MXT-1 column, the "sampling mode" duration was fixed at 1.8 s due to SIFT-MS software limitations. For the later tests with the second MXT-Volatiles column, the SIFT-MS operational software was upgraded to provide an arbitrary timing of the "sampling mode" duration~~.~~, where we used 6 or 12 s sampling intervals. Sampling was repeated several times to improve signal quality. The GC separation then takes place over typically 60 – 300 s whilst the eluent is continuously analysed by SIFT-MS. It is possible to apply a heating ramp during this period~~sensitivity~~.

Several heating ramp profiles were tested (see data for MXT-1 column in Fig. S1 in the Supplement); however, due to the short GC column and relatively long injection time, the monoterpene chromatogram peaks coalesced when the column temperature exceeded 60 °C and it was found that optimal chromatograms were obtained isothermally at 40 °C (15 V heating voltage). Effects of the heating voltage on the retention time and the chromatogram profile is illustrated in Fig. ~~S3~~S4 in the Supplement (data for MXT-Volatiles column).

3 SIFT-MS analyses of the eluent

In the present study, the *Profile 3* SIFT-MS instrument (Instrument Science, Crewe, UK) was used (Smith et al., 1999). Reagent ions are formed in a microwave discharge through a mixture of water vapour and atmospheric air (see Fig. 1). A mixture of ions is extracted from the discharge and focused into a quadrupole mass filter where they can be analysed according to their mass-to-charge ratio, *m/z*. Thus, the reagent ions H_3O^+, NO^+ or $O_2^{+\bullet}$ can be selected ($O_2^{+\bullet}$ was not used in the present experiment) and separately injected into flowing helium carrier gas (pressure p = 1.4 mbar, temperature T = 24 °C). Any internal energy possessed by the reagent ions is rapidly quenched in collisions with helium atoms leaving a thermalized ion swarm that is convected down the flow tube. Sample gas is introduced into the helium/thermalized swarm at a known flow rate that changes with the GC column temperature. The reagent ions react with the VOC molecules in the sample gas during a time period defined by the known flow speed of the ion swarm and the length of the flow tube. At the end of the flow tube, the ionic products (analyte ions) generated by ion-molecule reactions are sampled by a pinhole orifice into the analytical quadrupole mass spectrometer. The count rates of the reagent and analyte ions are obtained using a ~~channeltron~~single channel electron multiplier. Thus, full scan (FS) spectra can be obtained over a chosen *m/z* range to identify the analyte ions or rapidly switched between selected *m/z* values using the multiple-ion monitoring mode (MIM) (Španěl and Smith, 2013; Smith and

Španěl, 2011a). For the monoterpene study, FS mode was used for SIFT-MS analyses, whilst the MIM mode was used for fast GC-SIFT-MS setup. Typical count rate the reagent ions is one million cps, while amount of other ion lays below 1. Switching between two reagent ions requires milliseconds of time, as it depends mainly on the velocity of the carrier gas (12 000 cm.s^{-1}) and the length of the flow tube (5 cm). Therefore, the only limiting factor is a software sampling frequency, which depends on the amount of monitored ions, and is usually below one second.

3.1 Reactions of the H_3O^+ and NO^+ reagent ions with monoterpenes

In the present study, SIFT-MS analyses of monoterpenes were carried out using the previously investigated reactions of monoterpenes with H_3O^+ and $NO^{+\cdot}$ ions (Schoon et al., 2003; Wang et al., 2003). The H_3O^+ reactions are known to proceed via proton transfer forming $C_{10}H_{17}^+$ (*m/z* 137) that partially fragments to $C_6H_9^+$ (*m/z* 81) due to elimination of a C_4H_8 moiety from the nascent $(C_{10}H_{17})^*$ excited ion:

$$H_3O^+ + C_{10}H_{16} \rightarrow C_{10}H_{17}^+ + H_2O \quad (1a)$$

$$\rightarrow C_6H_9^+ + C_4H_8 + H_2O \quad (1b)$$

NO^+ reacts with monoterpenes by charge transfer forming the parent cation $C_{10}H_{16}^{+\bullet}$ (*m/z* 136) and a number of fragment ions, including $C_7H_9^+$:

$$NO^+ + C_{10}H_{16} \rightarrow C_{10}H_{16}^+ + NO \quad (2a)$$

$$\rightarrow C_7H_9^+ + NOC_3H_7 \quad (2b)$$

The exothermicity of charge transfer (2a) is represented by the difference between the ionization energies of the neutral NO (9.26 eV) and of the particular monoterpene (ranging from 8.07 eV for α-pinene to 8.4 eV for limonene) (Garcia et al., 2003; NIST). Other fragments, including $C_7H_8^+$, $C_7H_{10}^+$, $C_9H_{13}^+$ and $C_{10}H_{15}^+$, are also formed and the branching ratios between the channels (2a) to (2b) and other fragments depend on the isomeric structure of the monoterpene (Schoon et al., 2003; Wang et al., 2003) and are given in Table S1 in the Supplement. Based on this known ion chemistry, for the present study it was decided to analyse monoterpenes using both the H_3O^+ reagent ions by recording the $C_{10}H_{17}^+$ (*m/z* 137) and $C_6H_9^+$ (*m/z* 81) analyte ions and the NO^+ reagent ion by using the $C_{10}H_{16}^+$ (*m/z* 136) and $C_7H_9^+$ (*m/z* 93) analyte ions. To facilitate the identification of monoterpenes on the basis of the branching ratios of reactions (1) and (2), the product ion signal ratios [*m/z* 81]/[*m/z* 137] and [*m/z* 93]/[*m/z* 136] were determined under the conditions of the *Profile 3* SIFT-MS instrument using standard monoterpene mixtures, and these ratios (*r*) are given in Table ~~1.~~2, and discussed in Section 4.2.

The interaction of the primary ions with monoterpenes may be affected by presence of neutral water molecules and thus by different humidity of the sample ~~as~~. This was reported ~~previously~~ by Wang et al. (Wang et al., 2003) when decreased fragmentation of monoterpene product ions was observed in humid air samples~~.~~, what result in decrease of our product ion signal ratio *r* (see Section 3.2). For H_3O^+ regent ions, this change was significant for β-pinene (*r* reducing from 0.75 to 0.51), R-Limonene (*r* from 0.45 to 0.34) or 3-carene (*r* from 0.33 to 0.23). For the NO^+ ~~regent~~reagent ion, a significant effect was observed only for α-pinene (*r* from 0.32 to 0.08) and β-pinene (*r* from 0.25 to 0.05).

3.2 Analysis of the product ion intensity ratios

To facilitate assignment of the fast GC elution peaks to specific monoterpenes, mean fragment ion fractions $r_i = f_i/g_i$ = [*m/z* 81]/[*m/z* 137] (or for NO^+, $r_i = f_i/g_i$ = [*m/z* 93]/[*m/z* 136]) were calculated for each interval of retention times t_1 to t_2, as the weighted mean of the product ion signal ratios $\overline{r_w}$:

$$\overline{r_w} = \sum_{i=t_1}^{t_2} w_i \frac{f_i}{g_i};\ w_i = \frac{f_i+g_i}{\sum_{i=t_1}^{t_2} f_i+g_i}, \quad (3)$$

The weights (w_i) applied to each of several discreet measurements were based on the total signal of both ions f_i and g_i in order to emphasise the area within the peak. Time intervals t_1 to t_2 were chosen for each isomer as the area of the chromatographic peak where the total ion signal was >10% of the peak value.

The quality of the ratio estimation was assessed from the variation of the f_i/g_i ratio estimated as

$$\sigma_i^2 = Var(f/g) \approx \frac{\mu_f^2}{\mu_g^2}\left(\frac{\sigma_f^2}{\mu_f^2} + \frac{\sigma_g^2}{\mu_g^2}\right) = \frac{\mu_f^2}{\mu_g^2}\left(\frac{\lambda_f+\sigma_{bg_f}^2}{\mu_f^2} + \frac{\lambda_g+\sigma_{bg_g}^2}{\mu_g^2}\right), \quad (4)$$

where μ_f and μ_g represent intensities of the selected fragments and σ_f^2 and σ_g^2 are the variances of the μ_f and μ_g intensities estimated according to the Poisson distribution as the sum of distribution variance equal to the expected value $\lambda = \mu$ and background variance σ_{bg}^2 (Van Kempen and Van Vliet, 2000).

From this variation, the standard error of the weighted mean was calculated as:

$$\sigma_{\overline{r_w}} = \sqrt{\sum_{i=t_1}^{t_2} {w_i}^2 \sigma_i^2} \quad (5)$$

The weighted standard deviation of the f_i/g_i ratios was also routinely calculated as:

$$s = \sqrt{\frac{\sum_{i=t_1}^{t_2} w_i\left(\frac{f_i}{g_i}-\overline{r_w}\right)^2}{1-\sum_{i=t_1}^{t_2} {w_i}^2}} \quad (6)$$

~~3.3~~3.3 Fast GC SIFT-MS quantification

The total amount of eluting analyte, *C*, in each GC peak is determined by SIFT-MS from the area under the curve from the number density of the analyte molecules [M] (Španěl et al., 2006) in the flow tube recorded as a function of time, *t*, according to the equation:

$$C = \frac{1}{N_A}\int_0^{t_{max}} [M] S\, dt, \quad (7)$$

where N_A is the Avogadro constant and *S* is the constant volume flow rate of the sample and carrier gas mixture flowing into the SIFT-MS carrier gas as determined by the pumping speed of the SIFT-MS primary vacuum pump. Note that the flow rate of GC eluent gas does not enter this calculation and does not directly affect the determined amount of analyte expressed in nanomoles, nmol. [M] is calculated by the *Profile 3* software according to the SIFT-MS general method for the calculation of absolute trace gas concentrations from the reagent and product ion count rates, the reaction rate constants (see Table S1 in the Supplement) and the reaction time considering differential diffusion losses (see equation 15 in reference (Španěl et al., 2006).

The amount of analyte is proportional to its concertation [A] in sampled air and the sampled volume, V, given by the sampling flow rate (usually 3 sccm) and time (1.8 to 12 s) as:

$$C = [A]\frac{V}{V_m}, \quad (8)$$

where $[V_m]$ = 24.0 L/mol is the molar volume of air at 293 K.

3.4 Reference chemicals and plant samples

All monoterpenes used in the experiments, viz. ((+)-α-pinene (98%), (+)-β-pinene (≥98.5% analytical standard), camphene (95%), myrcene (≥90% analytical standard), -3-carene (≥98.5% analytical standard), R-limonene (≥99.0% analytical standard), α-terpinene (≥95%) and γ-terpinene (97 %), were purchased from Sigma-Aldrich Co. Individual monoterpene vapour standards and monoterpene vapour mixtures were prepared by the diffusion tube method (Thompson and Perry, 2009). Thus, for individual standards, about 5 µl of each monoterpene was placed in a 2 ml vial closed by PTFE septum caps. Each vial was then penetrated with a diffusion tube (1/16" OD x 0.25 mm ID x 5 cm length PEEK capillary) and placed into a 15 ml glass vial closed by a PTFE septum. The headspace of the 15 ml vial was sampled after stabilization (>–30 minutes) of the concentration. Humidity of the headspace was typically 1.5% water vapour by volume as determined by SIFT-MS. For the α-pinene, the intensities were too high and thus they had to be reduced by placing only trace amount of sample into the 2ml vial. For the mixture preparations, a similar approach was used; several vials containing different monoterpene, penetrated by PEEK capillaries, were placed together into a 500 ml bottle. Note that the concentrations of the individual isomers in the mixture are different due to the variations in their saturated vapour pressures. The same mixture was used for H_3O^+ and NO^+ experiments with the MXT-1 column.

To demonstrate the applicability of the fast GC/SIFT-MS analyses to real samples, three different types of coniferous tree needles were prepared: Spruce (*Pincea punges*), Fir (*Abies concolor*) and Pine (*Pinus nigra*) (see Fig. ~~S4 – S6~~S5 – S7 in the Supplement). For the first study using the MXT-1 column, the needle samples (0.26 g Spruce, 0.42 g Fir and 0.32 g Pine) were collected in the urban area of Prague in June 2017 and stored in 10 ml vials from which the headspace was sampled~~.~~ 30 min after harvesting. For the later study using the MXT-Volatiles column, pine tree twigs were collected in June 2018 from the same trees (21.8 g Spruce, 21.4 g Fir and 20.6 g Pine). The exposed cuts of the twigs were sealed by wrapping the parafilm around the cut. The samples were placed into a Nalophan bag of volume approximately one litre. During the analyses, the laboratory was thermalized to the outdoor temperature (about 30 °C) to reduce thermal shock to the samples. In the laboratory, only a scattered natural light was present.

4 Results and discussion

To investigate if the various monoterpenes in a mixture could be effectively distinguished using SIFT-MS enhanced by the fast GC pre-separation, eight common biogenic monoterpenes were investigated. ~~The mixture of~~ Individual monoterpene

standards ~~was~~were analysed first with both MXT-1 and MXT-Volatiles column to obtain the instrument response in terms of retention times and product ion ratios using two reagent ions H_3O^+ and NO^+. The separation of monoterpenes was demonstrated through analysis of prepared monoterpene mixture. Separation of both GC columns was compared using isothermal GC at ~~a column~~ temperature ~~of~~ 40 ~~°~~to 45 °C. The elution times of all studied monoterpenes were within 45 s of total retention time for MXT-1 column and within 180 s for MXT-Volatiles column. Using the information on the ratios of ion products for the H_3O^+ and NO^+ reactions together with the GC retention times, it was possible to identify the composition of a reference standard mixture. Finally, the same procedure was used to analyse the leaf headspace of three ~~fresh pine tree needle~~coniferous samples~~.~~ to demonstrate the analysis of real samples.

4.1 Comparison of columns: MXT-1 vs. MXT-Volatiles

In the present experiment we used heated columns isothermally to the temperature app. 40 °C due to the behaviour of the MXT-1 column. For higher temperatures, the monoterpene chromatogram peaks coalesced while for lower temperatures a significant influence of the lab air temperature fluctuations was apparent. At these conditions for MXT-1 column, monoterpenes are not fully separated and thus, fast GC with MXT-1 column alone (at 40 °C) provides only qualitative analysis. The retention times determined from the chromatograms obtained for individual monoterpenes at 40 °C are given in Table 2. For MXT-1 column, the apparent difference in retention times observed between the two reagent ions was probably caused by the temperature fluctuations of the column. Whilst the retention times for individual monoterpenes are different, they are not sufficiently stable (fluctuate by > 1 s, see Table ~~1 together with their $\overline{r_w}$ values (see equation 3). For MXT-1 column, the apparent difference in retention times observed between the two reagent ions was probably caused by the temperature fluctuations of the column. Whilst the retention times for individual monoterpenes are different, they are not sufficiently stable (fluctuate by > 1 s, see Table 1~~2) in the present fast GC device for analyses based on retention time only to be reliable. A noticeable effect of ambient temperature on the rate of passive column cooling was observed resulting in changes of the column temperature profile and thus in variations of the monoterpene retention times. Therefore, for longer column and higher temperature it may be reduced. Use of the MXT-Volatiles column resulted in about five times longer retention times and better GC peaks separation at the same operational conditions (flow rate, temperature and pressure) due to the higher efficiency of the 1.25 um active phase (compared to 0.1 um for MXT-1 column). ~~Use of the MXT-Volatiles column resulted in about five times longer retention times and better GC peaks separation at the same operational conditions (flow rate, temperature and pressure) due to the higher efficiency of the 1.25 um active phase (compared to 0.1 um for MXT-1 column). Due to the different sampling times used with each column (1.8 s for MXT-1 and 5 to 12 s for MXT-Volatiles) the peak shapes cannot be compared directly but the peak width (FWHM)~~

The quality of the separation could be increased by using hydrogen as a carrier gas and by a faster sample injection, as demonstrated by Materic et al. (Materić et al., 2015) with fastGC PTR-MS by where complete separation of monoterpenes was achieved~~only two times.~~

~~The performance of both MXT-1 and MXT-Volatiles columns were compared by analyses of a gas mixture of eight monoterpenes. For the MXT-1 column, four characteristic GC peaks were identified for both reagent ions, marked as A, B, C and D with retention time of 17.6 s, 20.8 s, 26.3 s and ~ 30 s for H_3O^+, and 17.5 s, 20.7 s, 26.3 s and ~ 30 s for NO^+ (see Fig. 3). Based on the retention times obtained for individual monoterpenes (see **Fig.** S2 in the Supplement), peak A is due to co-elution of α-pinene, camphene and myrcene. Peak B is due to the presence of β-pinene exclusively and peaks C and D are due to the remaining four monoterpenes. Note that the individual peak heights are influenced by the monoterpene saturated vapour pressures (see **Table 1**). Using the MXT-1 column at these conditions it is not possible to achieve separate GC peaks for individual monoterpenes, however qualitative analysis is possible.~~

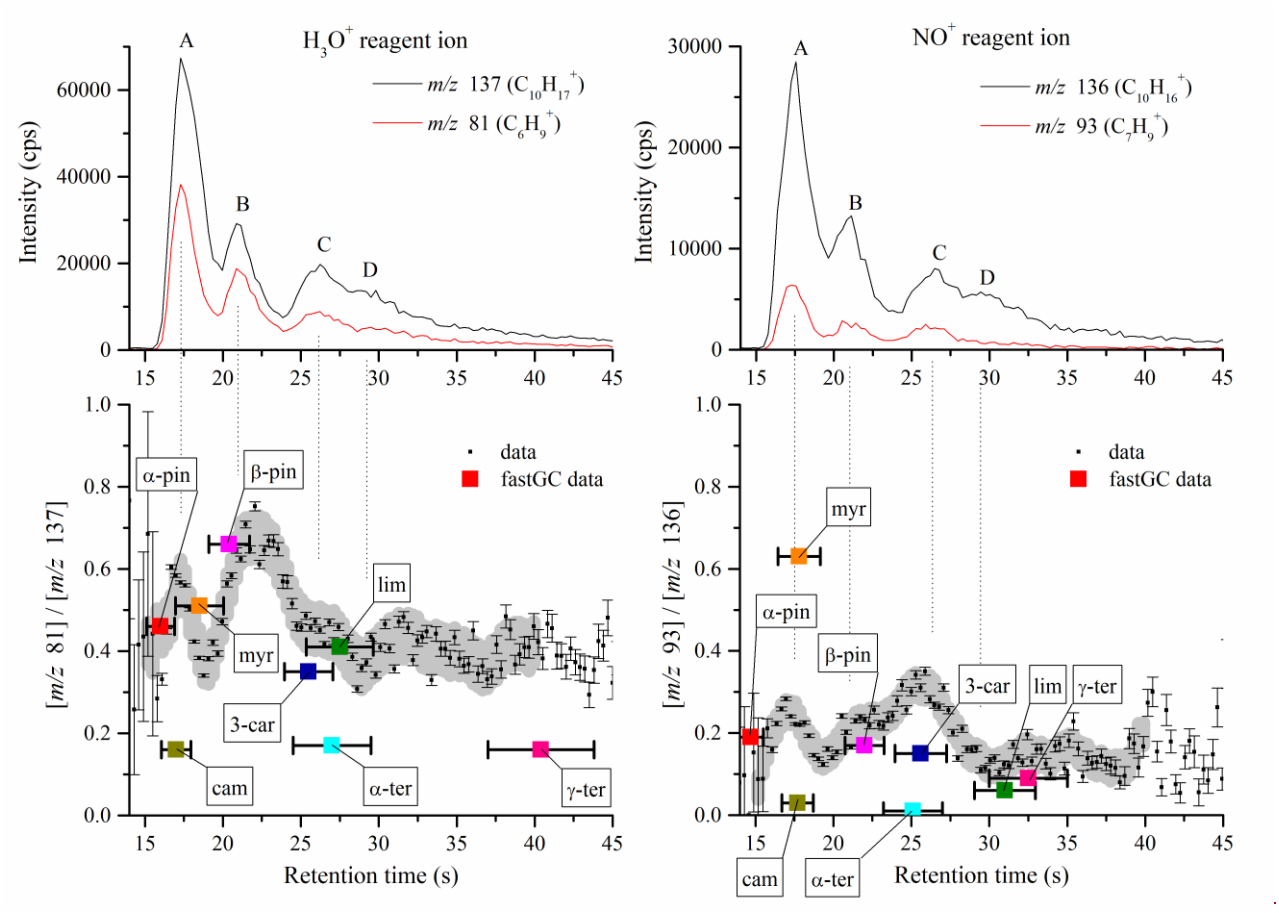
~~Table 1~~ using the MXT-1 column. As observed for both columns, separation can be improved by decreasing the column temperature (see Fig. 3 and Fig. S4 in the Supplement), however this increase the chromatogram width.

Table 2: Ratios of the H_3O^+ and NO^+ reaction product ion signals and the GC retention times, s, for the eight monoterpenes at columns temperature 40 °C. Also given are the saturated vapour pressures in Torr. The standard error of the fast GC $\overline{r_w}$ values for individual monoterpenes estimated by Eq. (5) is less than 5% (except 8.6% for camphene), overall less then ±0.02.

Compound	[*m/z* 81]/[*m/z* 137] H_3O^+		[*m/z* 93]/[*m/z* 136] NO^+		Retention time [s] H_3O^+	NO^+	H_3O^+
Saturated vapour pressure (Torr)	Literature *Schoon* [a] *Wang* [b]	Results *Full scan fast GC MIM*	Literature *Schoon* [a] *Wang* [b]	Results *Full scan fast GC MIM*	MXT-1	MXT-1	MXT-Vol
α-pinene 4.75[e]	0.45 0.64	0.67[c] 0.46[d]	0.05 0.09	0.16[c] 0.19[d]	16	14.7	72
camphene 2.50[e]	0.1 0.16	0.14[c] 0.16[d]	0 0.01	- 0.03[d]	17	17.7	83
β-pinene 2.93[e]	0.52 0.67	0.61[c] 0.66[d]	0.03 0.08	0.12[c] 0.17[d]	20.4	22	106
myrcene 2.09[f]	0.44 0.52	0.72[c] 0.51[d]	0.36 0.62	0.72[c] 0.63[d]	18.5	17.8	134
3-carene 3.72[h]	0.24 0.32	0.39[c] 0.35[d]	0.05 0.1	0.12[c] 0.15[d]	25.5	25.6	142
α-terpinene 1.64[h], 1.66[i]	- 0.11	0.14[c] 0.17[d]	-	0.01[c] 0.01[d]	27	25.1	157
R-limonene	0.30	0.43[c]	0	0.03[c]	27.5	31	170

1.98[g]	0.43	0.41[d]	0.01	0.06[d]			
γ-terpinene	-	0.18[c]	0.08	0.08[c]	40.4	32.5	184
1.07[h], 0.7[j]	0.21	0.16[d]	0.09	0.09[d]			

[a] (Schoon et al., 2003); [b] (Wang et al., 2003); [c] Present result based on SIFT-MS measurement; [d] Present result based on fast GC-SIFT-MS measurement; saturated vapour pressures in Torr at 25 °C are according to [e] (Daubert, 1989), [f] (Haynes, 2014), [g] (Yaws, 1994), [h] (TGSC), [i] (Takasago, 2011), and at 20 °C according to [j] (ChemicalBook, 2016).



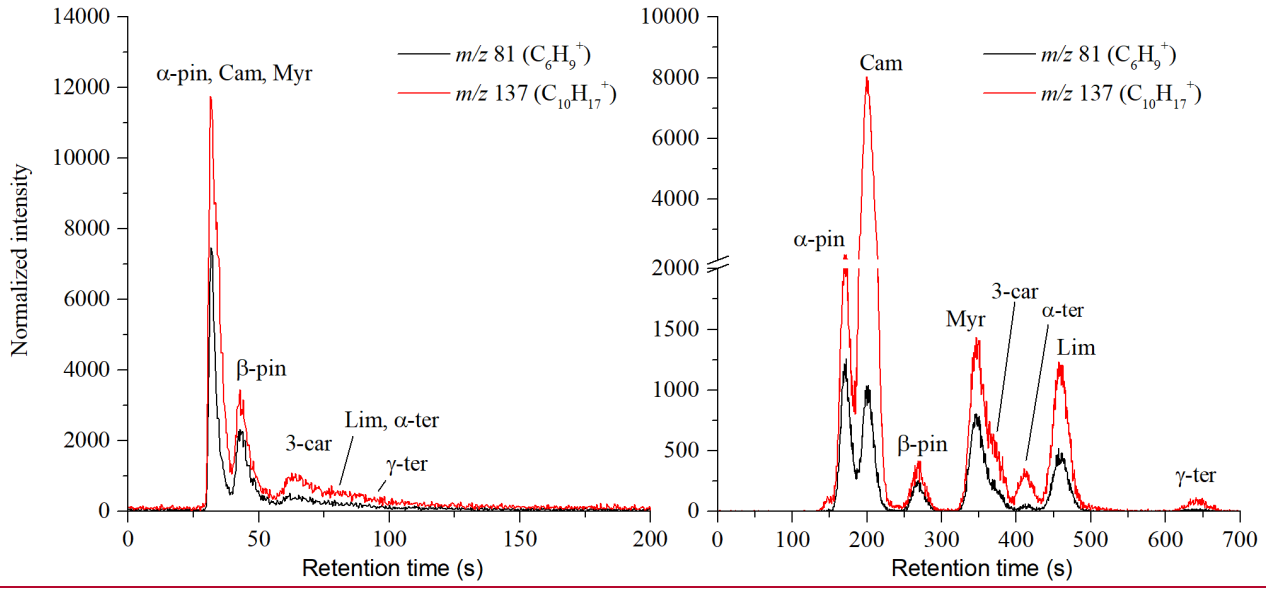


Figure 3: Chromatograms of ~~the~~ mixture of monoterpenes ~~(upper figures) measured by H_3O^+ (left) and NO^+ (right) reagent ions,~~ at room temperature obtained using ~~the~~ MXT-1 column~~. A, B, C, D represent characteristic~~ (left) and MXT-Volatiles column (right). Chromatogram peaks in the MXT-1 column are not fully separated, but separation takes below 150 s compare to 700 s required for MXT-Volatiles column. The signal intensities are the analyte ion count rates normalized to the H_3O^+ reagent ion count rate of 10^6 s^{-1}.

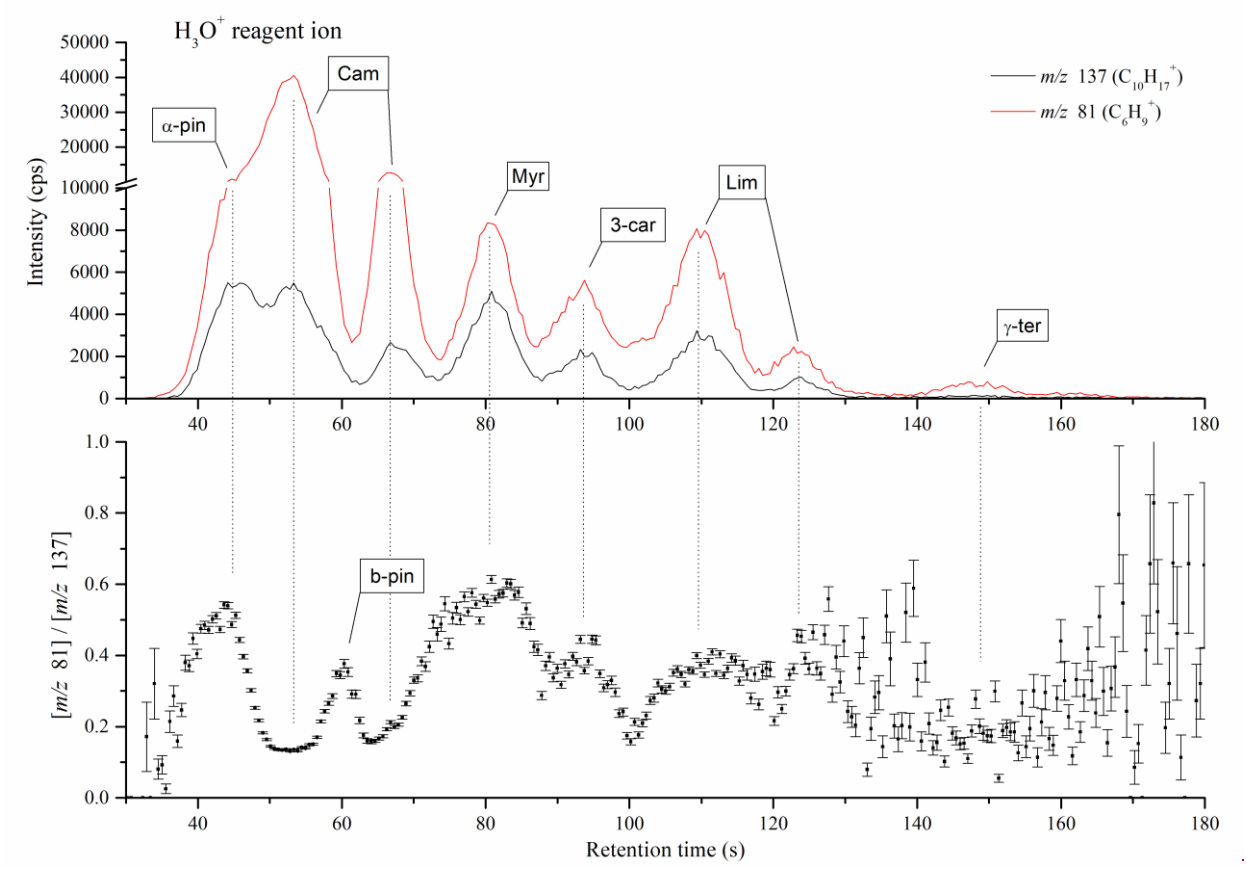
The performance of both MXT-1 and MXT-Volatiles columns were compared by analyses of a gas mixture of eight monoterpenes. For the MXT-1 column, four characteristic GC peaks were identified for both reagent ions, marked as A, B, C and D with retention time of 17.6 s, 20.8 s, 26.3 s and ~30 s for H_3O^+, and 17.5 s, 20.7 s, 26.3 s and ~30 s for NO^+ (see Fig. ~~chromatogram. For each chromatogram, the product ion signal ratio r_i is presented in the lower figures. The grey data background represents the calculated standard deviation of the data by Savizky-Golay smoothing between 15 s and 40 s. The position and value of the ratio for~~ 4). Based on the retention times obtained for individual monoterpenes ~~is based on the fast GC MXT-1 measurements presented in~~(see Table 2 and Fig. S2 in the Supplement), peak A is due to co-elution of α-pinene, camphene and myrcene. Peak B is due to the presence of β-pinene exclusively and peaks C and D are due to the remaining four monoterpenes~~1. –~~, mainly 3-carene and R-Limonen. Note that the individual peak heights are influenced by the monoterpene saturated vapour pressures (see Table 2). Using the MXT-1 column at these conditions it is not possible to achieve separate GC peaks for individual monoterpenes, however qualitative analysis is possible.

~~**retention times are determined by the fast GC conditions and do not depend on which SIFT-MS reagent ion is used.**~~

~~The quality of the separation could be increased by using hydrogen or helium as a carrier gas and by a faster sample injection, as demonstrated by Materic et al. (Materić et al., 2015) with fastGC-PTR-MS by where complete separation of monoterpenes was achieved. As observed for both columns, separation can be as well improved by decreasing the column temperature (see **Fig. S3** in the Supplement), however this may increase the chromatogram width and thus decrease sensitivity of the technique. Additional sensitivity can be obtained from increasing the injection time, which will, however, increase the peak width. In the~~

~~present experiment we used heated columns isothermally to the temperature app. 40 °C. due to the behaviour of the MXT-1 column. For higher temperatures, the monoterpene chromatogram peaks coalesced. For lower temperatures a significant influence of the lab air temperature fluctuations was apparent. At these conditions for MXT-1 column, monoterpenes are not fully separated and thus, fast GC with MXT-1 column alone (at 40 °C) provides only qualitative analysis.~~

The MXT-Volatiles column facilitates identification of all monoterpenes present in the mixture for temperatures close to the room temperature (see Fig. ~~S3 in the Supplement).~~3). For the MXT-Volatiles tests, the sampling mode was extended to 12 s, representing the collection of approximately 0.6 mL of the monoterpene mixture headspace. ~~A noticeable effect of ambient~~ At column temperature ~~on the rate of passive column cooling was observed resulting in changes of the column temperature profile and thus in variations of~~40 °C, the monoterpene ~~retention times.~~peaks are well separated, however, α-pinene and camphene are likely to co-elute as they are usually very intensive. It is interesting to note that the chromatogram (see Fig. ~~S3~~S4 in the Supplement) changes with the temperature of the column and additional peaks appear at higher temperatures probably as a result of presence of different conformers. It thus seems that at the column temperature ~45 °C using 20 V heating voltage (see Fig. 4) in the mixture chromatogram the small β-pinene is hidden behind the second camphene peak and the α-terpinene peak also disappears (see also the fragmentation analyses later in section 4.2).



~~**Figure 4: A chromatogram for a prepared mixture of monoterpenes obtained with the MXT-Volatiles column as derived by SIFT-MS using H_3O^+ reagent ions. For the chromatogram, the product ion signal ratio r_i is presented in the bottom figure. This chromatogram was obtained at the column temperature ~ 45 °C using 20 V heating voltage (separation is faster compare to data on Table 1).**~~

~~**4.2 Use of the NO^+ reagent ions for improved selectivity**~~

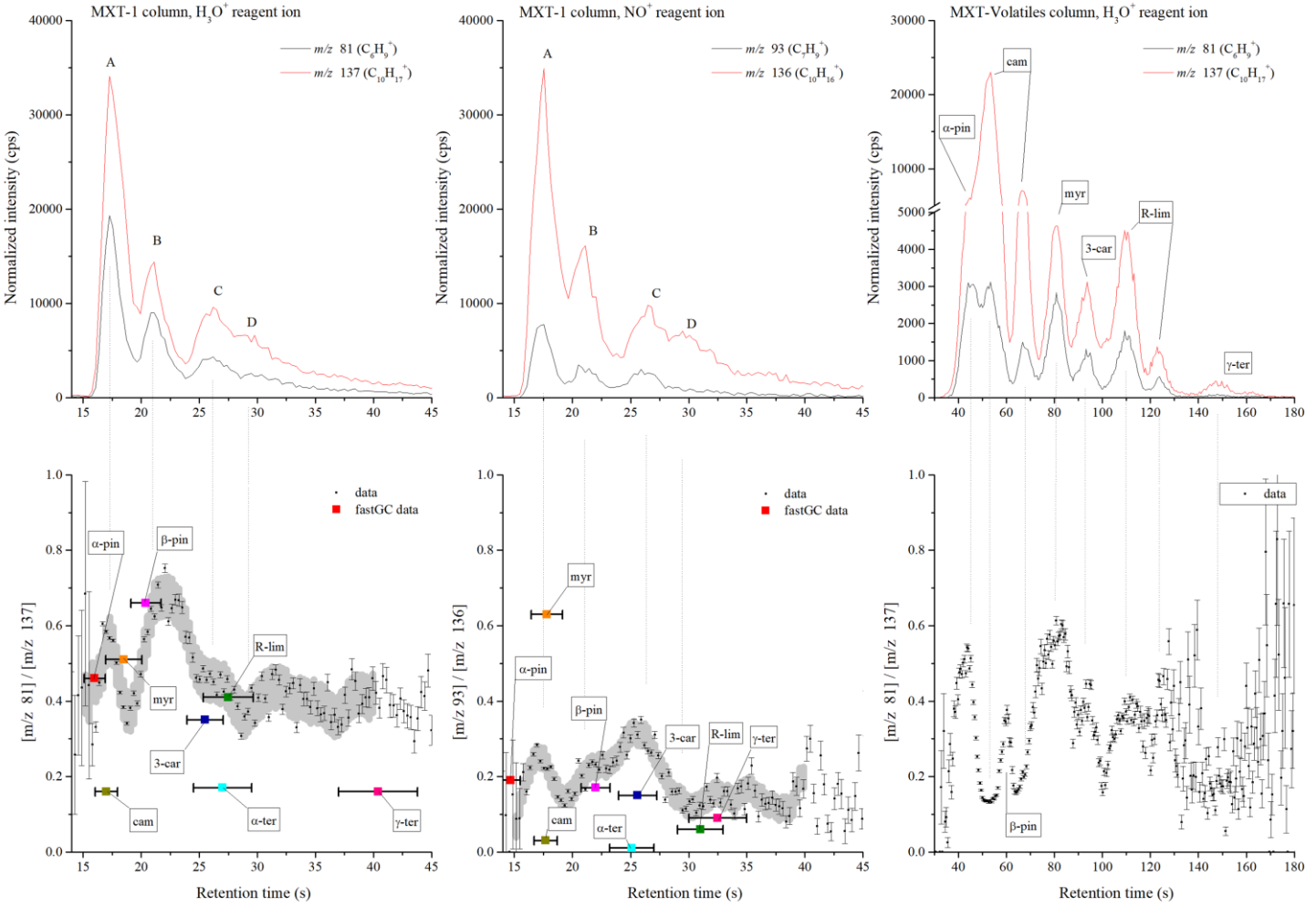


Figure 4: Chromatograms of the mixture of monoterpenes (upper figures) measured by H_3O^+ (left) and NO^+ (right) reagent ions, obtained using the MXT-1 column. A, B, C, D represent characteristic peaks in the chromatogram. For each chromatogram, the product ion signal ratio r_i is presented in the lower figures. The grey data background represents the calculated standard deviation of the data by Savizky-Golay smoothing between 15 s and 40 s. The position and value of the ratio for individual monoterpenes is based on the fast GC MXT-1 measurements presented in Table 1. Note that the retention times are determined by the fast GC conditions and do not depend on which SIFT-MS reagent ion is used. The signal intensities are the analyte ion count rates normalized to a reagent ion count rate of 10^6 s^{-1}.

4.2 Analysis of product ratio and use of the NO^+ reagent ions

The inadequate separation of monoterpenes due to a short column or high temperature (as the case of MXT-1 column) can be mitigated by ~~using an additional reagent ion and by~~ the analysis of the product ion signal ratios r_i (see Sec. ~~3.2).~~3.2) and additionally by using an additional reagent ion. It may be possible to improve identification of myrcene or camphene (often co-eluted with a-pinene) as well as of other monoterpenes by exploiting different ion chemistry of the NO^+ regent ions. These data in combination with H_3O^+ data allow identification of compounds on the basis of the ratios of in total four different product ions. Use of NO^+ regent ions was applied only on MXT-1 column, as full separation of monoterpenes using H_3O^+ reagent ions was not achieved and thus retention time cannot be effectively used as parameter for their identification. Hoverer, as will be presented, use of the NO^+ regent ions brings additional benefits and thus it may be a valuable source of information even for fully separated chromatograms. Note that the retention times are determined by the fast GC conditions and do not depend on which SIFT-MS reagent ion is used (see Table ~~1~~ 2).

~~It must be kept in mind, that monoterpenes are not the only BVOCs emitted by plants. Especially when plants are physically damaged, they emit so called „leaf aldehydes" such as 2-, and 3-hexenal (Tani et al., 2003). Ion chemistry of these two aldehydes differs in SIFT-MS. Whilst the reaction of 2-hexenal with H_3O^+ proceeds as a proton transfer forming a product ion at *m/z* 99 (100 %), it has been found that reaction of cis-3-hexenal with H_3O^+ results in H_2O elimination producing a dominant fragment at *m/z* 81 (Španěl et al., 1997). To avoid an overlap of 3-hexenal with monoterpenes, it is thus more reliable to use the product ion at *m/z* 137. Another possibility is to choose NO^+ as a precursor ion, where the product ions of 3-hexenal (*m/z* 97, 69 and 74) do not overlap with those of monoterpenes (*m/z* 92, 93 and 136) (Wang et al., 2003).~~

~~The same approach can be applied to other isomeric or isobaric molecules present in environment like isoprene that form a product ion at *m/z* 69, overlapping with the second hydrate of methanol that is also emitted by plants (12% of global BVOC emissions) (Španěl et al., 1999).~~

The $\bar{r_w}$ values (see Table 2) obtained from the SIFT-MS FS data and the MIM data for the fast GC peaks for most of the isomers are in good agreement. However, the ratios obtained for α-pinene and myrcene are somewhat variable between the FS and MIM data and they also differ somewhat from the literature values~~.~~ (α-pinene from 0.45 to 0.67 for H_3O^+, myrcene from 0.44 to 0.72 for H_3O^+). This may be caused by different humidities of the samples, as discussed ~~-~~in Section 3.1., where it was seen that increase of humidity ~~lowered~~lower the $\bar{r_w}$ values. ~~The greatest fluctuation was observed for myrcene, which was not found to be sensitive to humidity in contrast to~~In fast-GC setup, water retention time is much lower as the ~~sensitive compounds (β pinene, R-Limonene and 3 carene for H_3O^+ regent~~retention time for monoterpenes, thus water influence on ion~~, and~~ chemistry is for most monoterpenes negligible. Slightly affected can be α-pinene~~, β-pinene NO^+ regent ion).~~ as he is the first one presented in the chromatogram. Therefore, only $\bar{r_w}$ values obtained using the fast CG will be used for further study. The standard error of the fast GC $\bar{r_w}$ values for individual monoterpenes estimated by Eq. (5) (using the MXT-1 column) is less than 5% (except 8.6% for camphene) and is smaller than the observed variability between the methods. The $\bar{r_w}$ values for MXT-Volatiles column were similar to those obtained with MXT-1 column, as expected.

~~The~~Analysis of $\overline{r_w}$ values can be now used to improve identification of monoterpenes in measured mixtures. For MXT-1 column, the $\overline{r_w}$ values for peaks A, B, C and D were calculated as 0.49, 0.63, 0.45, 0.40 respectively for H_3O^+ and as 0.21, 0.21, 0.27, 0.14 for NO^+. Based on these ratios (using fast GC data from Table ~~1~~2), peak B could clearly be assigned as β-pinene. However, the remaining peaks contain several isomers and thus the $\overline{r_w}$ values are not providing unique identifications. So dynamic variations of r_i needed to be investigated to see if they can provide additional information.

The time profile r_w in chromatogram is shown in the bottom part of Fig. ~~3~~4. To recognize trends in these data, Savizky-Golay smoothing (Savitzky and Golay, 1964) was used (second polynomial order across 10 data points, OriginPro 9.0 (OriginPro, 2018). Also plotted (grey area in Fig. ~~3~~4) is the standard deviation of the data points from the smoothed line in the interval of retention times from 15 s to 40 s. Note that this standard deviation is greater than the standard error of the data points, possibly due to a lower accuracy of data at the longer retention times. The standard deviation allows assessment of the significance of the changes in $r_i = f_i/g_i$.

According to the elution time, the first chromatographic peak A consist of three monoterpenes: α-pinene, camphene and myrcene. For the H_3O^+ reagent ions, the $\overline{r_w}$ value corresponds to both α-pinene and myrcene considering the $\overline{r_w}$ value for peak A (0.49) or r_w close to the peak maxima (0.55–0.6). However, a more obvious difference between α-pinene and myrcene is observed ~~with~~using the NO^+ reagent ions. The value of the weighted mean ratio for the peak A (0.21) is close to the ratio for α-pinene. In the maxima of peak A, however, r_w approaches the value of 0.3, which is close to the value expected for a combination of both these monoterpenes (0.32, considering the data from fast GC measurement and the vapour pressure in Table ~~1~~2). For camphene, r_w in the chromatograph did not reach the low values expected for both reagent ions. However, its presence is clearly visible as a dip in r_w situated between the peaks A and B. In the absence of camphene, the ratio should linearly move to values characteristic for the peak B without any dip. The depth of the dip does not reach the ratio value expected for camphene due to a persistent tails of the peaks for both α-pinene and myrcene.

Peak B in the chromatograms is identified as β-pinene by its retention time. The $\overline{r_w}$ values for the H_3O^+ and NO^+ reagent ions are 0.63 and 0.21, respectively. The values r_w are similar to $\overline{r_w}$ and slightly higher than to the fast GC standard values for β-pinene (see Table ~~1~~2).

Peaks C and D are not clearly separated in the chromatogram. For the H_3O^+ reagent ions, the $\overline{r_w}$ value is similar for both peaks; thus, the presence of limonene, 3-carene or α-terpenine is likely since the $\overline{r_w}$ values for the peaks C (0.45) and D (0.4) are comparable with the analyte signal ratios (see Table ~~1~~2) for limonene and 3-carene. A lower r_i for α-terpinene might be observed as a dip similar to camphene. However, the observed dip in r_i at the D peak is not so statistically significant as the dip for camphene, and the vapour pressure for both α- and γ-terpinene are lower than other monoterpenes. Analysis of the C and D peaks using the NO^+ reagent ion shows a clearer difference between them. The calculated $\overline{r_w}$ for the peak C (0.27) as well as the maximum r_i (0.35) are, unexpectedly, much higher than for the remaining monoterpenes. This can be explained only by the influence of myrcene or by the presence of impurities in the form of an additional monoterpene in the mixture (for example ocimeme has high r_i of 0.62 (Wang et al., 2003)). Amongst the eight monoterpenes, 3-carene has the highest r_i within

the retention time of peak C. The second peak D (0.14) can be then associated with R-limonene, which has a low r_i (0.06) for NO^+ reagent ions, with some contribution by α-terpinene. The presence of γ-terpenine is not visible due to its low vapour pressure, but there may be some contribution in the D peak, but much smaller than the contribution by limonene.

To summarize, combining analyses using both H_3O^+ and NO^+ reagent ions with dynamic variations of r_i allows the identification of α-pinene, camphene and myrcene in peak A followed by the presence of β-pinene in peak B exclusively. Peak C is characterized as 3-carene and peak D as R-limonene and/or α-terpinene. γ-terpenine contributes only weakly due to its low vapour pressure and has no recognisable response in the chromatogram compared to the remaining monoterpenes.

Analysis of the $\bar{r_w}$ values for MXT-Volatiles column is more simple due to better separation of peaks. Value of r_i clearly change for different monoterpenes, according the expected $\bar{r_w}$ values for individual monoterpenes. Usefulness of the r_i analysis for MXT-Volatiles column can be observed in analysis of β-pinene, which is featureless compare to the camphene. Camphene, additionally, produce second chromatographic peak, which can be easily incorrectly associated with β-pinene. Analysis of the r_i show values below 0.2 for both peak maxima, characteristic for camphene. Presence of β-pinene is visible as increase of r_i value up to 0.4 at retention time 60 s.

4.3 Tree samples investigation using the MXT-1 column

To test how fast GC-SIFT-MS is applicable for analyses of real biological samples, VOC emissions were analysed from three fresh coniferous tree needle samples: spruce, fir, and pine as shown in Fig. 5. MS obtained using the H_3O^+ regent ion are shown in Fig. S3 in Supplement. Based on the results of the above GC data for standard monoterpene mixtures, the chromatograms were divided into three areas. The first part characterized by the presence of α-pinene, camphene and myrcene between retention times of 12-18 s, the second part characterized by the presence of β-pinene with retention times between 18-25 s and the third part characterized by presence of 3-carene and limonene~~/α-terpenine~~ with retention times between 25-40 s. The $\bar{r_w}$ values were calculated for the selected regions as follows

- Spruce: The first region of the main peak 0.35 (H_3O^+), 0.11 (NO^+). Note that the very low $\bar{r_w}$ for NO^+ indicates the absence of Myrcene. The $\bar{r_w}$ value for H_3O^+ is lower than expected for β-pinene and higher than expected for camphene. Therefore, the first peak is formed mainly from α-pinene, perhaps with small amount of camphene. The second region of a small peak 0.38 (H_3O^+) and 0.14 (NO^+). $\bar{r_w}$ for H_3O^+ is lower than expected for β-pinene and higher than that for camphene, ~~low $\bar{r_w}$ for NO^+ indicates~~ signal therefore belongs to the ~~absence~~decay of ~~myrcene; 3-carene is thus the best match.~~α-pinene. The signal ~~increase~~ratio 0.38 (H_3O^+), 0.14 (NO^+) in the third region ~~may~~ indicates ~~trace~~ presence of R-limonene~~.~~ or 3-carene.
- Fir: The chromatogram shows two intense peaks. The calculations of $\bar{r_w}$ for the first region (0.40 for H_3O^+, 0.14 for NO^+) and for the second region (0.56 for H_3O^+, 0.15 for NO^+) indicate the presence of both α-pinene and β-pinene.

The decreasing $\overline{r_w}$ for the H_3O^+ reagent ions in the last part (0.48 for H_3O^+, 0.19 for NO^+) indicates the presence of ~~an additional monoterpene,~~ 3-carene.

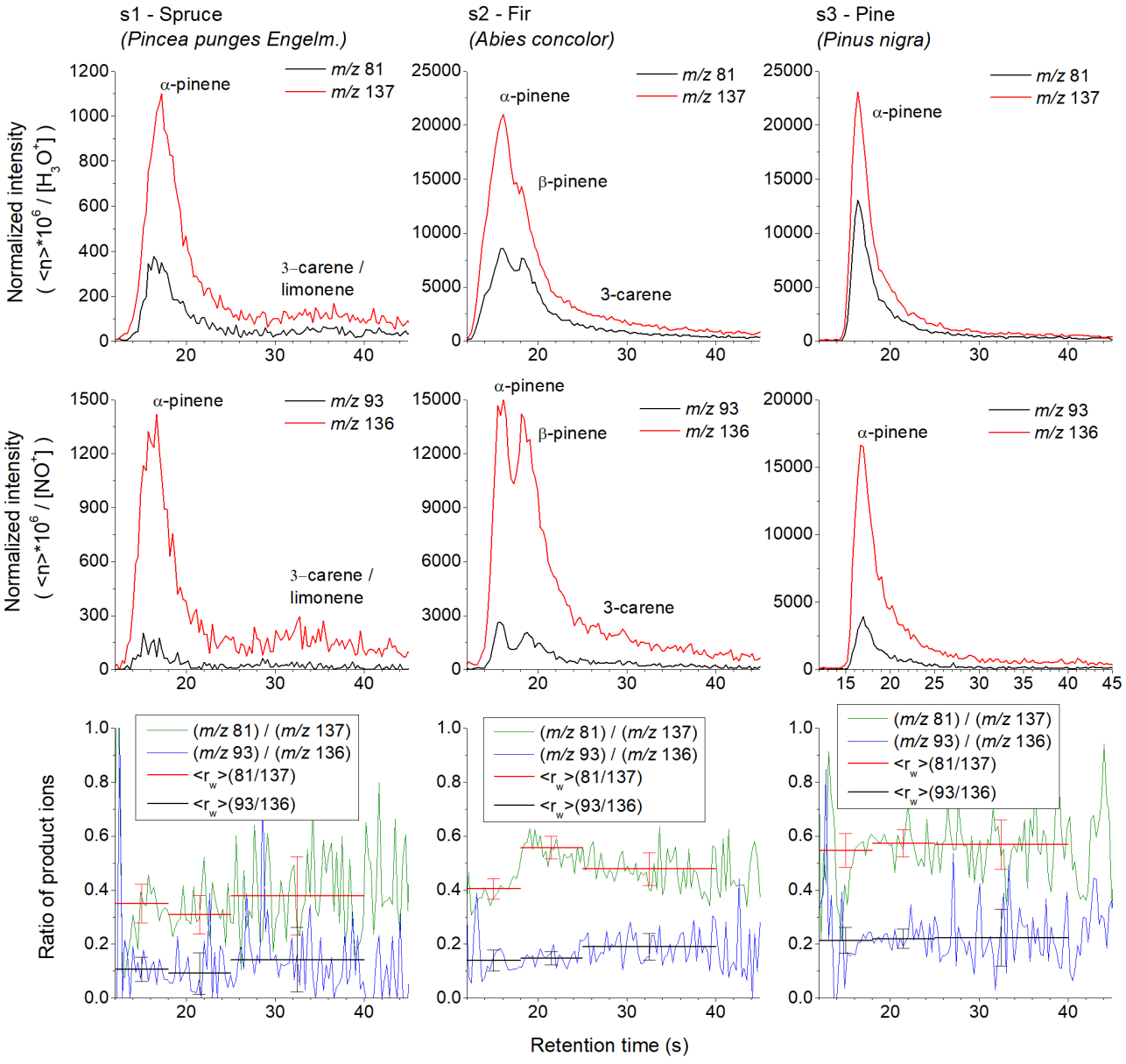
- Pine: Chromatogram contains only one peak. $\overline{r_w}$ is stable for both reagent ions for all retention times (0.55 for H_3O^+, 0.21 for NO^+ for the first sector; 0.57 for H_3O^+, 0.22 for NO^+ for the second sector; 0.57 for H_3O^+, 0.22 for NO^+ for the third sector). Together with the retention time of the peak (16.4 s) this certainly corresponds to α-pinene.

Concentrations of individual monoterpenes were calculated according to the procedure described in Section 3.3 for all selected regions. Calculation of monoterpene concentrations depends primarily on the individual reaction rate constants (see Table S1 in Supplement), which change from 2.3 to 2.6 for H_3O^+ and from 2.0 to 2.3 for NO^+ (in units of $10^{-9}cm^3s^{-1}$). Incorrect identification of monoterpened will thus lead to maximum 20% error in the concentration calculation. According to the $\overline{r_w}$ values in selected regions, the most representative rate constant was adopted to calculated monoterpene concentration in the selected region (see Table 3).

Table 3: Calculated concentrations of monoterpenes (in ppm and %) in the headspace over coniferous needles in selected regions of chromatogram obtained using MXT-1 column at column temperature 40 °C, using injection time 1.8 s and column flow 3 sccm. Rate constant used for calculation of concentration in selected regions was chosen according to $\overline{r_w}$ analysis.

Sample	Concentration (ppm, %)			
	12-18s	18-25s	25-40s	Sum 12-40s
Spruce (H_3O^+)	11.0[A], **42%**	9.0[A], **35%**	5.2[R], 5.9[3], **23%**	25.2 [A,R], 25.9 [A,3]
Spruce (NO^+)	14.5[A], **50%**	6.6[A], **23%**	7.4[R], 7.7[3], **27%**	28.5 [A,R], 28.8 [A,3]
Fir (H_3O^+)	177[A], **32%**	274[B], **49%**	95[R], 107[3], **19%**	546 [A,B,R], 558 [A,B,3]
Fir (NO^+)	117[A], **31%**	191[B], **51%**	74[R], 77[3], **18%**	372 [A,B,R], 375 [A,B,3]
Pine (H_3O^+)	195[A], **55%**	112[A], **31%**	43[R], 49[3], **14%**	350 [A,R], 356 [A,3]
Pine (NO^+)	128[A], **48%**	100[A], **37%**	38[R], 41[3], **15%**	266 [A,R], 269 [A,3]

Calculations were performed using the reaction rate constants for [A] α-pinene, [B] β-pinene, [R] R-limonene or [3] 3-carene.



~~4.4 Tree samples analyses using the MXT-Volatiles column~~

~~Similar experiments were conducted also using the MXT-Volatiles column. The retention times for the individual monoterpenes were taken from the standard data obtained at the same column temperature (40 °C). The headspaces of the~~

prepared tree needle samples were sampled for 6 s, representing a volume of 0.3 mL. The chromatograms obtained for the spruce, fir and pine samples are shown in Fig. 6 and represent the means of analyte ion count rates from 5 consecutive runs normalized to a constant reagent ion count rate of 10^6 s^{-1}.

- For spruce, four peaks were observed in the chromatogram. The first peak with a retention time of 68 s corresponds to α pinene with $\bar{r}_{tt}$ of 0.60 for H_3O^+ and 0.24 for NO^+ regent ions. The tailing edge of the first peak shows a decrease of $\bar{r}_{tt}$ (0.29 for H_3O^+, 0.14 for NO^+) due to a small contribution by camphene. The second peak corresponds to β-pinene, characterized by a retention time of 94 s with $\bar{r}_{tt}$ of 1.05 for H_3O^+ and 0.50 for NO^+. The standard deviation in r_{tt} was unfortunately substantial (±0.6 for H_3O^+, ±0.73 for NO^+). The position of the third peak corresponds to myrcene. The $\bar{r}_{tt}$ values (0.43 for H_3O^+, 0.41 for NO^+) were again imprecise due to the low intensity and do not fully agree with the unique $\bar{r}_{tt}$ for myrcene (see Table 1). The observed weak peak could therefore be due to other monoterpenes other than those eight included in Table 1. The last peak corresponds to 3-carene with $\bar{r}_{tt}$ as 0.48 for H_3O^+ and 0.16 for NO^+ reagent ions

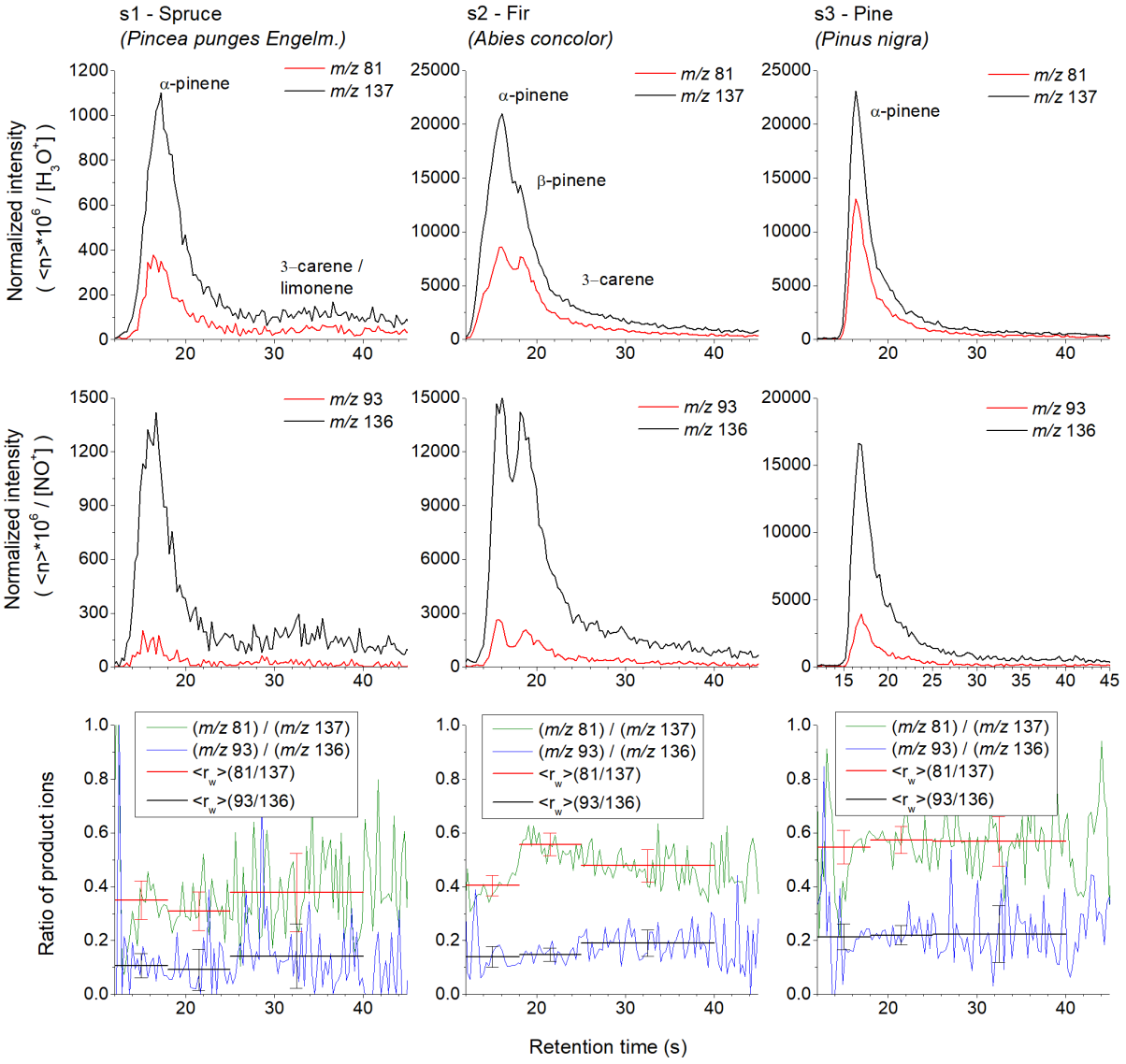
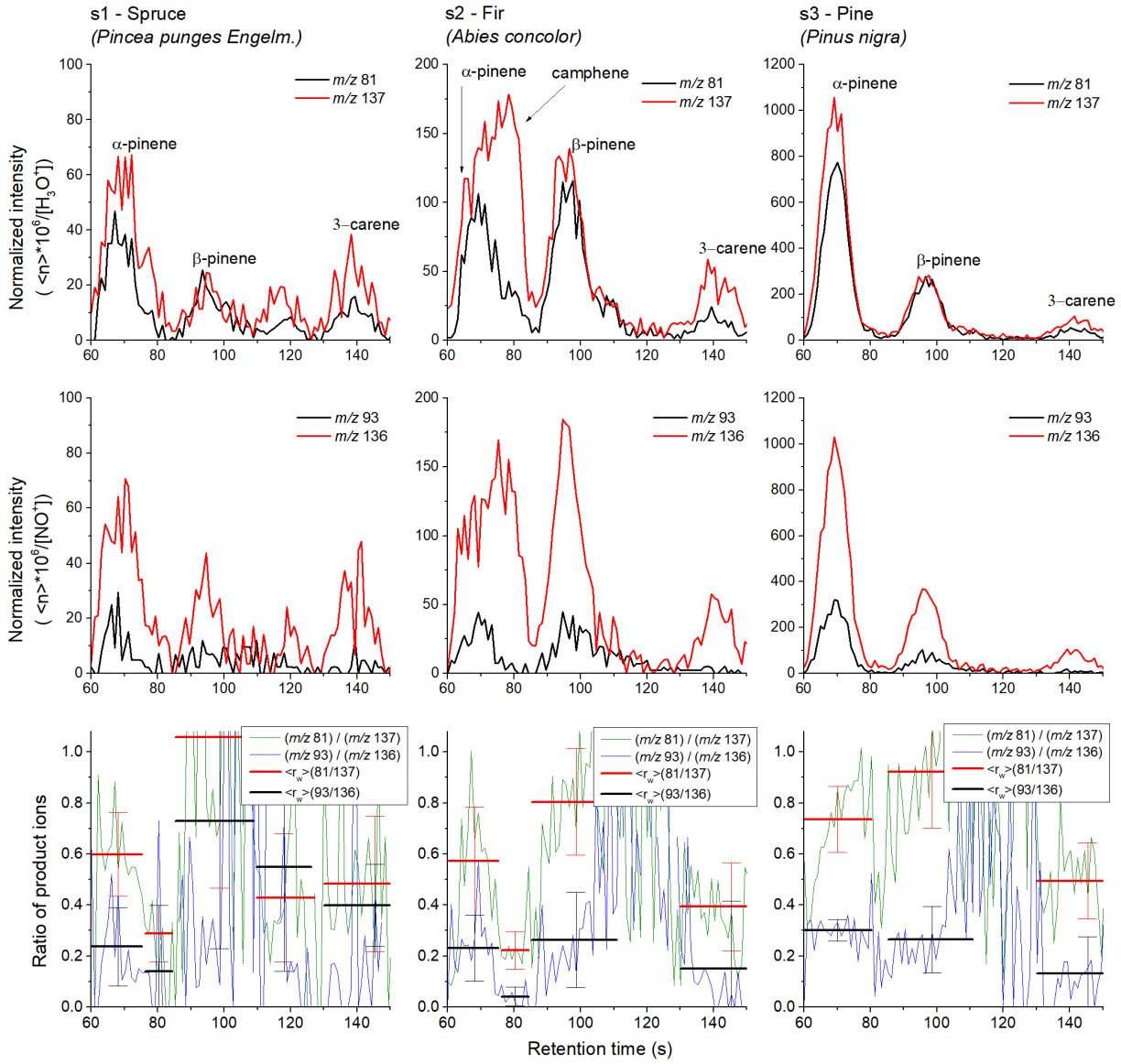


Figure 5: Chromatograms derived using the product ions for the reactions of H_3O^+ (upper row) and NO^+ (lower row) reagent ions with monoterpenes obtained for the three investigated pine tree samples (s1, s2 and s3) using the MXT-1 column. The signal intensities are the analyte ion count rates normalized to a reagent ion count rate of 10^6 s^{-1}. The black and red curves represent $C_6H_9^+$ (*m/z* 81) and $C_{10}H_{17}^+$ (*m/z* 137) product ions for H_3O^+ and $C_7H_9^+$ (*m/z* 93) and $C_{10}H_{16}^+$ (*m/z* 136) product ions for NO^+ reagent ions. The last row shows calculated ratios of product ions r_i for both reagent ions (green and blue curves) and for peaks areas calculated $\overline{r_w}$ (red and black).



4.4 Tree samples analyses using the MXT-Volatiles column

Similar experiments were conducted also using the MXT-Volatiles column, although on a different set of coniferous samples. The retention times for the individual monoterpenes were taken from the standard data obtained at the same column temperature (40 °C). The headspaces of the prepared tree needle samples were sampled for 6 s, representing a volume of

0.3 mL. The chromatograms obtained for the spruce, fir and pine samples are shown in Fig. 6 and represent the means of analyte ion count rates from 5 consecutive runs normalized to a constant reagent ion count rate of 10^6 s^{-1}.

- Spruce: In the chromatogram, four peaks were observed . The first peak with a retention time of 68 s corresponds to α-pinene with $\bar{r_w}$ of 0.60 for H_3O^+ and 0.24 for NO^+ regent ions. The tailing edge of the first peak shows a decrease of $\bar{r_w}$ (0.29 for H_3O^+, 0.14 for NO^+) due to a small contribution by camphene. The second peak corresponds to β-pinene, characterized by a retention time of 94 s with $\bar{r_w}$ of 1.05 for H_3O^+ and 0.50 for NO^+. The standard deviation in r_w was unfortunately substantial (±0.6 for H_3O^+, ±0.73 for NO^+). The position of the third peak corresponds to myrcene. The $\bar{r_w}$ values (0.43 for H_3O^+, 0.41 for NO^+) were again imprecise due to the low intensity and do not fully agree with the unique $\bar{r_w}$ for myrcene (see Table 2). The observed weak peak could therefore be due to other monoterpenes other than those eight included in Table 1. The last peak corresponds to 3-carene with $\bar{r_w}$ as 0.48 for H_3O^+ and 0.16 for NO^+ reagent ions

Fir: In the chromatogram, three peaks are presentwhere the first is due to both α-pinene and camphene.~~**Figure 6: SIFT-MS selected ion mode/fast GC/SIFT-MS chromatograms for monoterpene emissions from pine tree samples (s1, s2 and s3) obtained using the MXT-Volatiles column. The upper and lower rows were obtained using H_3O^+ and NO^+ reagent ions respectively. The signal intensities are the analyte ion count rates normalized to a reagent ion count rate of 10^6 s^{-1}. The black and red curves stand for monitored ions $C_6H_9^+$ (m/z 81) and $C_{10}H_{17}^+$ (m/z 137) for H_3O^+ reagent ions of $C_7H_9^+$ (m/z 93) and $C_{10}H_{16}^+$ (m/z 136) for NO^+ reagent ions respectively. The last row shows calculated ratios of product ions r_t for both reagent ions (green and blue curves) and for peaks areas calculated $\overline{r_w}$ (red and black).**~~

- ~~The chromatogram for the fir sample shows three peaks, where the first is due to both α pinene and camphene.~~ Transition of $\bar{r_w}$ from the left (0.57 for H_3O^+, 0.23 for NO^+) to the right (0.22 for H_3O^+, 0.04 for NO^+) part of the first peak is clearly visible on the Fig. 6 in middle column. The first peak of the fir sample thus consists of two isomers. The second peak is due to β-pinene ($\bar{r_w}$ 0.80 for H_3O^+, 0.26 for NO^+) and the third peak by 3-carene ($\bar{r_w}$ 0.39 for H_3O^+, 0.15 for NO^+).
- ~~The pine sample chromatogram~~Pine: Chromatogram shows three clear peaks of α-pinene (0.73 for H_3O^+, 0.30 for NO^+), β-pinene (0.92 for H_3O^+, 0.26 for NO^+) and 3-carene (0.49 for H_3O^+, 0.13 for NO^+) with just a very small and statistically insignificant indication of camphene. The retention times for α-pinene, β-pinene and 3-carene were 69.6 s, 97 s and 141 s, respectively.

Some differences can be seen between the results from the MXT-1 and MXT-Volatiles columns. The most significant difference is the presence of a camphene peak in the fir sample headspace, and the presence of β-pinene and 3-carene in the pine sample headspace when the MXT-Volatiles column was used. However, samples were collected at different times of the year and the character of the samples was also different (only needles for MXT-1 and whole twigs for the MXT-Volatiles analyses). Different sample sources could cause differences in monoterpene concentration as well (see Table 4).

Table 4: Calculated concentrations of monoterpenes (in ppm and %) in the headspace over coniferous twigs in selected regions of chromatogram obtained using MXT-Volatiles column at column temperature 40 °C, using injection time 6 s and column flow 3 sccm. Rate constant used for calculation of concentration in selected regions was chosen according to $\overline{r_w}$ analysis.

Sample	Concentration (ppm, **%**)				
	α-pinene	Camphene	β-pinene	3-carene	Sum
Spruce (H_3O^+)	0.97, **46%**	0.21, **10%**	0.46, **22%**	0.48, **22%**	2.12
Spruce (NO^+)	0.74, **36%**	0.26, **13%**	0.56, **27%**	0.49, **24%**	2.05
Fir (H_3O^+)	2.51, **31%**	1.46, **18%**	2.9, **36%**	1.17, **15%**	8.04
Fir (NO^+)	1.97, **28%**	1.29, **19%**	2.80, **40%**	0.88, **13%**	6.94
Pine (H_3O^+)	15.5, **65%**	nd	5.95, **25%**	2.29, **10%**	23.74
Pine (NO^+)	13.7, **65%**	nd	5.45, **26%**	1.83, **9%**	20.98

nd – no data

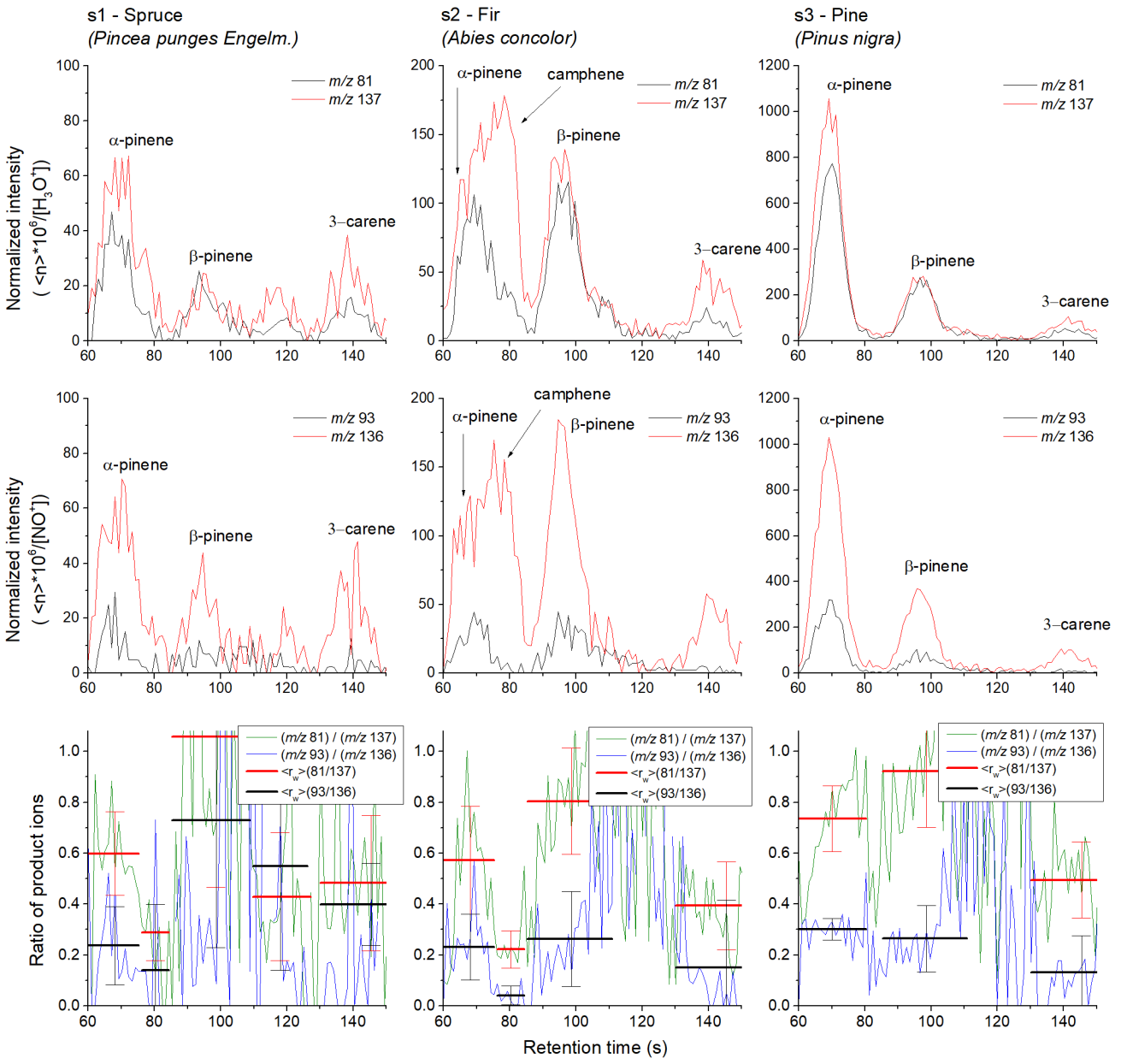


Figure 6: SIFT-MS selected ion mode/fast GC/SIFT-MS chromatograms for monoterpene emissions from pine tree samples (s1, s2 and s3) obtained using the MXT-Volatiles column. The upper and lower rows were obtained using H_3O^+ and NO^+ reagent ions respectively. The signal intensities are the analyte ion count rates normalized to a reagent ion count rate of 10^6 s^{-1}. The black and red curves stand for monitored ions $C_6H_9^+$ (*m/z* 81) and $C_{10}H_{17}^+$ (*m/z* 137) for H_3O^+ reagent ions of $C_7H_9^+$ (*m/z* 93) and $C_{10}H_{16}^+$ (*m/z* 136) for NO^+ reagent ions respectively. The last row shows calculated ratios of product ions r_i for both reagent ions (green and

blue curves) and for peaks areas calculated $\overline{r_w}$ (red and black). The signal intensities are the analyte ion count rates normalized to a reagent ion count rate of 10^6 s^{-1}.

4.5 Comparison with previous studies

The present tests indicate that using the fast GC-SIFT-MS combination, it is possible to achieve ~~only qualitative~~ analysis of the monoterpene mixture ~~with a~~. The limit of ~~the~~ detection ~~about 100 ppb.~~was determined for α-pinene and R-limonene from analysis of a calibration curve as three times the standard error of predicted intercept value divided by the slope of the calibration regression line (Graus et al., 2010). α-pinene and R-limonene were chosen as they have the lowest and the highest ration rate constants for proton transfer (2.3 for α-pinene and 2.6 for R-limonene, in $10^{-9}cm^3s^{-1}$). When the reagent ion count rate was 10^6 c/s and 12 seconds sampling interval was used, the detection limit of the current setup was found to be 16.3 ppbv for α-pinene and 19.5 ppbv for R-limonene, using the column temperature 40 °C. For column temperature 69 °C, limit of detection for α-pinene decreased to 6.1 ppbv. This is inferior to the previously described limit of the detection up to 1-2 ppb and full separation achieved by fastGC-PTR-MS systems (Materić et al., 2015; Pallozzi et al., 2016~~), which achieved full separation with limit of the detection up to 1-2 ppt.~~).

However, one advantage of SIFT-MS is the possibility to use another reagent ion as well as analysis of product ion ratios can be helpful. The combination of the data from the two reagent ions together with the analyses of the product ion signal ratios r_i can be shown to improve the identification of monoterpenes, especially identification of camphene and myrcene. It must be kept in mind, that monoterpenes are not the only BVOCs emitted by plants. Especially when plants are physically damaged, they emit so called „leaf aldehydes“ such as 2-, and 3-hexenal (Tani et al., 2003). Ion chemistry of these two aldehydes differs in SIFT-MS. Whilst the reaction of 2-hexenal with H_3O^+ proceeds as a proton transfer forming a product ion at *m/z* 99 (100 %), it has been found that reaction of cis-3-hexenal with H_3O^+ results in H_2O elimination producing a dominant fragment at *m/z* 81 (Španěl et al., 1997). To avoid an overlap of 3-hexenal with monoterpenes, it is thus more reliable to use the product ion at *m/z* 137. Another possibility is to choose NO^+ as a precursor ion, where the product ions of 3-hexenal (*m/z* 97, 69 and 74) do not overlap with those of monoterpenes (*m/z* 92, 93 and 136) (Wang et al., 2003). ~~,~~Aside from potentially better selectivity, a benefit of employing the NO^+ regent ions in atmospheric analysis is quantification of isoprene, which for H_3O^+ regent ions interferes with furan, C5 aldehydes and 2-methyl-3-buten-2-ol (Karl et al., 2012; Karl et al., 2014), and overlapping with the second hydrate of methanol that is also emitted by plants (12% of global BVOC emissions) (Španěl et al., 1999). The same approach can be applied to other isomeric or isobaric molecules present in environment. The last benefit of using SIFT-MS compare to other techniques is that calculation of VOC concentration in the sample depends only on the known physical constants, reaction rate constant and ions abundance. The system therefore does not require complicated calibration procedures.

The results obtained from the ~~present study~~analysis of leaf headspace samples in the terms of monoterpene composition agree well ~~with~~other studies in the published literature ~~reports~~. Because the emission from plans depends on various physical parameters, here we compare only monoterpene composition. In a previous study (Mumm et al., 2004) of the volatiles emitted

by *Pinus nigra* needles, 35 terpenoid compounds were identified, with the following being most abundant: α-pinene (45%), β-phellandrene (9%), limonene (8%), β-pinene (5%) and 3-carene (2%). Holzke et al. (2006) studied diurnal and seasonal variation of monoterpenes and sesquiterpenes from Scots pine. The main isomers they observed were α-pinene, β-pinene and 3-carene, which represented 90% of the total terpene emission. A similar study on monoterpene emissions from boreal Scots pine showed that the most abundant monoterpenes measured above the forest and from the canopy were α-pinene and 3-carene (Räisänen et al., 2009).

Kainulainen et al. (Kainulainen et al., 1992) investigated the effect of drought and waterlogging stress on needle monoterpenes of *Picea abies* (spruce). In the controlled group, the most abundant monoterpenes were camphene (22%), limonene (14%), α-pinene (9%) and myrcene (6%). In the emission from Southern and Central Sweden (Janson, 1993) the following isomers were most abundant: α--pinene (60-70%), camphene (10%), limonene (10%) and 3-carene (4%). Analysis of spruce samples (Zavarin et al., 1975) studied cortical oleoresin from *Abies concolor* (fir) that were collected in 43 different localities in order to analyse their composition for the monoterpenoid fractions. They concluded that the production of camphene and 3-carene varied geographically. In the study of (Pureswaran et al., 2004) they focused on quantitative variations in monoterpene vapours in four species of conifers, concluding that the four species (Douglas-fir, Lodgepole pine, Interior spruce and Interior Fir) did not differ qualitatively but there were significant differences in their quantitative profiles. For example, Coastal Douglas fir needle samples contained 10% of α-pinene, 31% of Sabinene and 40% of β-pinene, and in samples of interior Douglas fir the most abundant isomers were bornyl acetate (26%), camphene (25%), α-pinene and β-pinene (both 15%).

In presented study, we detected presence of α-pinene, β-pinene, camphene and 3-carene, representing common emissions emitted from pine, spruce and fir samples. The present results thus agree with the usually reported composition of terpenes emitted from pine trees and their parts.

5 Summary and conclusions

A new method has been developed that Addition of a fast GC pre-separation to SIFT-MS allows quantitative analyses of individual monoterpenes in mixtures using SIFT-MS enhanced by chromatographic pre-separation. As a pre-separation module,at the expense of some loss of sensitivity. The bespoke electrically heated fast GC systems was constructed by which pre-separation of the isomers was for this study achieved separation in retention times shorterless than 45 s for ana 5 m MXT-1 column and shorterless than 180 s for ana 5 m MXT-Volatiles column. Individual at 40 °C. The identification of monoterpenes were identified and analysedwas aided by SIFT-MS fromusing the information on the ratios of the analyteproduct ion signals generated in their reactions with of both H_3O^+ and NO^+ reagent ions. Thus, using bothIt was shown that combining the SIFT-MS analyteproduct ion ratios and the GC retention times, six 7 of 8 monoterpenes were identified in a mixture of eight were identified using the MXT-1 column whilst all eight monoterpenes were identified using the MXT-Volatiles column. To demonstrate the applicability of this unique analytical application of this novel combination to real samples, theof fast GC with SIFT-MS, volatile monoterpenes in the headspace of emissions from spruce, fir and pine

~~needles~~samples were analysed. ~~All headspace samples clearly contained~~ α-pinene ~~and~~was identified together with a lower amount of β-pinene and 3-carene. A significant contribution of camphene was also observed in the fir sample headspace. The fast GC SIFT-MS combination can thus be a step towards atmospheric analyses of monoterpenes that should resolve individual compounds due to their different reactivity with the OH radicals.

A weakness of the current fast GC setup is the relatively poor temperature stability caused by a strong dependence on the laboratory ambient temperature. However, this can surely be improved by active temperature feedback to control the column temperature. The flow rate through 5 m long and 0.28 mm i.d. column was about ten times lower than the conventional flow rate used in direct SIFT-MS analyses and this resulted in commensurate worsening of the limit of detection. This could be in future resolved by using a wider column or by using multiple capillaries in parallel. A clear advantage of SIFT-MS has been shown to be in the possibility to use several different product ions to determine different fragmentation ratios from data obtained for H_3O^+ and NO^+ at the same retention time to improve the identification of compounds.

~~The present study demonstrated that the combination of SIFT-MS determination of fragment ion fractions together with low resolution pre-separation technique allows analyses of monoterpenes in short periods of time. This novel idea of a fast GC-SIFT-MS combination could broaden the application of SIFT-MS to *in situ* trace gas analyses of complex mixtures such as ambient air and exhaled breath.~~

Data availability

All data are available upon request from the corresponding author (Michal Lacko).

Author contribution

ML and NW crated experimental hardwere and provided experiments with MXT-1 column, ML, KS and PP then provided experiments with MXT-Volatiles column. PS and ML provdided data reatment and paper preparation.

Competing interests

The authors declare that they have no conflict of interest.

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References

Allardyce, R. A., Langford, V. S., Hill, A. L., and Murdoch, D. R.: Detection of volatile metabolites produced by bacterial growth in blood culture media by selected ion flow tube mass spectrometry (SIFT-MS), Journal of microbiological methods, 65, 361-365, 2006.

Amelynck, C., Schoon, N., and Dhooghe, F.: SIFT Ion Chemistry Studies Underpinning the Measurement of Volatile Organic Compound Emissions by Vegetation, Current Analytical Chemistry, 9, 540-549, 2013.

Atkinson, R., Aschmann, SM., and Pitts Jr, JN.: Rate constants for the gas-phase reactions of the OH radical with a series of monoterpenes at 294±1 K. International Journal of Chemical Kinetics, 18, 287-299, 1986.

Atkinson, R., Aschmann, SM., and Arey, J.: Rate constants for the gas-phase reactions of OH and NO_3 radicals and O_3 with sabinene and camphene at 296±2 K. Atmospheric Environment Part A: General Topics, 24, 2647-2654, 1990.

Atkinson, R., Hasegawa, D., and Aschmann, SM.: Rate constants for the gas-phase reactions of O_3 with a series of monoterpenes and related compounds at 296±2 K, International Journal of Chemical Kinetics, 22, 871-887, 1990.

Atkinson, R., and Arey, J.: Gas-phase tropospheric chemistry of biogenic volatile organic compounds: a review, Atmospheric Environment, 37, ~~197-219~~S197-S219, 2003.

Chameides, W., Fehsenfeld, F., Rodgers, M., Cardelino, C., Martinez, J., Parrish, D., Lonneman, W., Lawson, D., Rasmussen, R., and Zimmerman, P.: Ozone precursor relationships in the ambient atmosphere, Journal of Geophysical Research: Atmospheres, 97, 6037-6055, 1992.

ChemicalBook: https://www.chemicalbook.com/ChemicalProductProperty_EN_CB4443087.htm, .

OriginPro 9.0 OriginLab Corporation, One Roundhouse Plaza, Suite 303, Northampton, MA 01060, United States. 1800-969-7720.: https://www.originlab.com/, 2018.

Daubert, T. E.: Physical and thermodynamic properties of pure chemicals: Data compilation Hemisphere Pub. Corp., New York, 1989.

de Gouw, J., and Warneke, C.: Measurements of volatile organic compounds in the earth's atmosphere using proton-transfer-reaction mass spectrometry, Mass Spectrometry Reviews, 26, 223-257, 2007.

Ellis, A. M., and Mayhew, C. A.: Proton transfer reaction mass spectrometry: principles and applications, John Wiley & Sons, 2013.

Fehsenfeld, F., Calvert, J., Fall, R., Goldan, P., Guenther, A. B., Hewitt, C. N., Lamb, B., Liu, S., Trainer, M., and Westberg, H.: Emissions of volatile organic compounds from vegetation and the implications for atmospheric chemistry, Global Biogeochemical Cycles, 6, 389-430, 1992.

Garcia, G. A., Nahon, L., and Powis, I.: Near-threshold photoionization spectroscopy of the mono-terpenes limonene and carvone, International Journal of Mass Spectrometry, 225, 261-270, 2003.

de Gouw, J. A., Goldan, P. D., Warneke, C., Kuster, W. C., Roberts, J. M., Marchewka, M., Bertman, S. B., Pszenny, A. A. P., and Keene, W. C.: Validation of proton transfer reaction-mass spectrometry (PTR-MS) measurements of gas-phase organic

compounds in the atmosphere during the New England Air Quality Study (NEAQS) in 2002, Journal of geophysical research: atmospheres, 108, D21, 2003.

Graus, M., Müller, M., and Hansel, A.: High resolution PTR-TOF: quantification and formula confirmation of VOC in real time, Journal of the American Society for Mass Spectrometry, 21, 1037-1044, 2010.

Haynes, W. M.: CRC handbook of chemistry and physics, CRC press, New York, 2014.

Holzke, C., Hoffmann, T., Jaeger, L., Koppmann, R., and Zimmer, W.: Diurnal and seasonal variation of monoterpene and sesquiterpene emissions from Scots pine (Pinus sylvestris L.), Atmospheric Environment, 40, 3174-3185, 2006.

Janson, R. W.: Monoterpene emissions from Scots pine and Norwegian spruce, Journal of geophysical research: atmospheres, 98, 2839-2850, 1993.

Jordan, A., Haidacher, S., Hanel, G., Hartungen, E., Herbig, J., Märk, L., Schottkowsky, R., Seehauser, H., Sulzer, P., and Märk, T. D.: An online ultra-high sensitivity Proton-transfer-reaction mass-spectrometer combined with switchable reagent ion capability (PTR+SRI−MS), International Journal of Mass Spectrometry, 286, 32-38, 2009.

Kainulainen, P., Oksanen, J., Palomäki, V., Holopainen, J., and Holopainen, T.: Effect of drought and waterlogging stress on needle monoterpenes of Picea abies, Canadian Journal of Botany, 70, 1613-1616, 1992.

Karl, T., Hansel, A., Cappellin, L., Kaser, L., Herdlinger-Blatt, I., and Jud, W.: Selective measurements of isoprene and 2-methyl-3-buten-2-ol based on NO^+ ionization mass spectrometry, Atmospheric Chemistry and Physics, 12, 11877–11884, 2012.

Karl, T., Kaser, L., and Turnipseed, A.: Eddy covariance measurements of isoprene and 232-MBO based on NO^+ time-of-flight mass spectrometry, International Journal of Mass Spectrometry, 365-366, 15-19, 2014.

Kesselmeier, J., and Staudt, M., Biogenic Volatile Organic Compounds (VOC): An Overview on Emission, Physiology and Ecology, Journal of Atmospheric Chemistry, 33, 23-88, 1999.

Kulmala, M., Suni, T., Lehtinen, K., Maso, M. D., Boy, M., Reissell, A., Rannik, Ü., Aalto, P., Keronen, P., and Hakola, H.: A new feedback mechanism linking forests, aerosols, and climate, Atmospheric Chemistry and Physics, 4, 557-562, 2004.

Lindinger, W., Hansel, A., and Jordan, A.: On-line monitoring of volatile organic compounds at pptv levels by means of proton-transfer-reaction mass spectrometry (PTR-MS) medical applications, food control and environmental research, International Journal of Mass Spectrometry and Ion Processes, 173, 191-241, 1998.

Malásková, M., Henderson, B., Chellayah, P., Ruzsanyi, V., Mochalski, P., Cristescu, S. M., and Mayhew, C. A.: Proton transfer reaction time-of-flight mass spectrometric measurements of volatile compounds contained in peppermint oil capsules of relevance to real-time pharmacokinetic breath studies. Journal of breath research, 2019.

Materić, D., Lanza, M., Sulzer, P., Herbig, J., Bruhn, D., Turner, C., Mason, N., and Gauci, V.: Monoterpene separation by coupling proton transfer reaction time-of-flight mass spectrometry with fast GC, Analytical and bioanalytical chemistry, 407, 7757-7763, 2015.

Matisová, E. and Dömötörová, M.: Fast gas chromatography and its use in trace analysis, Journal of Chromatography A, 1000, 199-221, 2003.

Mumm, R., Tiemann, T., Schulz, S., and Hilker, M.: Analysis of volatiles from black pine (Pinus nigra): significance of wounding and egg deposition by a herbivorous sawfly, Phytochemistry, 65, 3221-3230, 2004.

NIST WebBook Chemie, NIST Standard Reference Database Number 69, U.S. Secretary of Commerce, National Institute of Standards and Technology: Gaithersburg, MD: http://webbook.nist.gov/chemistry/ cited in January 2019.

Nolscher, A. C., Yanez-Serrano, A. M., Wolff, S., de Araujo, A. C., Lavric, J. V., Kesselmeier, J., and Williams, J.: Unexpected seasonality in quantity and composition of Amazon rainforest air reactivity, Nat Commun, 7, 10383, 2016.

Pallozzi, E., Guidolotti, G., Ciccioli, P., Brilli, F., Feil, S., and Calfapietra, C.: Does the novel fast-GC coupled with PTR-TOF-MS allow a significant advancement in detecting VOC emissions from plants?, Agricultural and Forest Meteorology, 216, 232-240, 2016.

Pureswaran, D. S., Gries, R., and Borden, J. H.: Quantitative variation in monoterpenes in four species of conifers, Biochemical systematics and ecology, 32, 1109-1136, 2004.

Pysanenko, A., Španěl, P., and Smith, D.: Analysis of the isobaric compounds propanol, acetic acid and methyl formate in humid air and breath by selected ion flow tube mass spectrometry, SIFT-MS, International Journal of Mass Spectrometry, 285, 42-48, 2009.

Räisänen, T., Ryyppö, A., and Kellomäki, S.: Monoterpene emission of a boreal Scots pine (Pinus sylvestris L.) forest, Agricultural and forest meteorology, 149, 808-819, 2009.

Rhoderick, G. C., and Lin, J.: Stability assessment of gas mixtures containing monoterpenes in varying cylinder materials and treatments, Analytical chemistry, 85, 4675-4685, 2013.

Rinne, J., Ruuskanen, T. M., Reissell, A., Taipale, R., Hakola, H., and Kulmala, M.: On-line PTR-MS measurements of atmospheric concentrations of volatile organic compounds in a European boreal forest ecosystem, Boreal environment research, 10, 425-436, 2005.

Romano, A., Fischer, L., Herbig, J., Campbell-Sills, H., Coulon, J., Lucas, P., Cappellin, L., and Biasioli, F.: Wine analysis by Fast GC proton-transfer reaction-time-of-flight-mass spectrometry, International Journal of Mass Spectrometry, 369, 81-86, 2014.

Savitzky, A., and Golay, M. J.: Smoothing and differentiation of data by simplified least squares procedures, Anal. Chem., 36, 1627-1639, 1964.

Schoon, N., Amelynck, C., Vereecken, L., and Arijs, E.: A selected ion flow tube study of the reactions of H_3O^+, NO^+ and O_2^+ with a series of monoterpenes, International Journal of Mass Spectrometry, 229, 231-240, 2003.

Shestivska, V., Antonowicz, S. S., Dryahina, K., Kubišta, J., Smith, D., and Španěl, P.: Direct detection and quantification of malondialdehyde vapour in humid air using selected ion flow tube mass spectrometry supported by gas chromatography/mass spectrometry, Rapid Communications in Mass Spectrometry, 29, 1069-1079, 2015.

Shestivska, V., Nemec, A., Dřevínek, P., Sovová, K., Dryahina, K., and Španěl, P.: Quantification of methyl thiocyanate in the headspace of Pseudomonas aeruginosa cultures and in the breath of cystic fibrosis patients by selected ion flow tube mass spectrometry, Rapid Communications in Mass Spectrometry, 25, 2459-2467, 2011.

Shestivska, V., Spanel, P., Dryahina, K., Sovova, K., Smith, D., Musilek, M., and Nemec, A.: Variability in the concentrations of volatile metabolites emitted by genotypically different strains of Pseudomonas aeruginosa, Journal of Applied Microbiology, 113, 701-713, 2012.

Smith, D., Španěl, P., Holland, T. A., Al Singari, W., and Elder, J. B.: Selected ion flow tube mass spectrometry of urine headspace, Rapid communications in mass spectrometry, 13, 724-729, 1999.

Smith, D., and Španěl, P.: Selected ion flow tube mass spectrometry (SIFT-MS) for on-line trace gas analysis, Mass spectrometry reviews, 24, 661-700, 2005.

Smith, D., and Španěl, P.: Ambient analysis of trace compounds in gaseous media by SIFT-MS, Analyst, 136, 2009-2032, 2011a.

Smith, D., and Španěl, P.: Direct, rapid quantitative analyses of BVOCs using SIFT-MS and PTR-MS obviating sample collection, TrAC Trends in Analytical Chemistry, 30, 945-959, 2011b.

Smith, D., Sovová, K., and Španěl, P.: A selected ion flow tube study of the reactions of H_3O^+, NO^+ and $O_2^{+\bullet}$ with seven isomers of hexanol in support of SIFT-MS, International Journal of Mass Spectrometry, 319, 25-30, 2012.

Song, M., Xia, Y., and Tomasino, E.: Investigation of a Quantitative Method for the Analysis of Chiral Monoterpenes in White Wine by HS-SPME-MDGC-MS of Different Wine Matrices, Molecules, 20, 7359-7378, 2015.

Španěl, P., Ji, Y., and Smith, D.: SIFT studies of the reactions of H_3O^+, NO^+ and O_2^+ with a series of aldehydes and ketones, International journal of mass spectrometry and ion processes, 165, 25-37, 1997.

Španěl, P., and Smith, D.: Selected ion flow tube studies of the reactions of H_3O^+, NO^+, and O_{2+} with several aromatic and aliphatic hydrocarbons, International Journal of Mass Spectrometry, 181, 1-10, 1998.

Španěl, P., Davies, S., and Smith, D.: Quantification of breath isoprene using the selected ion flow tube mass spectrometric analytical method, Rapid communications in mass spectrometry, 13, 1733-1738, 1999.

Španěl, P., Dryahina, K., and Smith, D.: A general method for the calculation of absolute trace gas concentrations in air and breath from selected ion flow tube mass spectrometry data, International Journal of Mass Spectrometry, 249-250, 230-239, 2006.

Španěl, P., and Smith, D.: Advances in On-line Absolute Trace Gas Analysis by SIFT-MS, Curr. Anal. Chem., 9, 525-539, 2013.

Takasago: http://www.takasago.com/cgi-bin/pdf/alphaterpinene.pdf, 2011, cited in January 2019.

Tani, A., Hayward, S., and Hewitt, C.: Measurement of monoterpenes and related compounds by proton transfer reaction-mass spectrometry (PTR-MS), International Journal of Mass Spectrometry, 223, 561-578, 2003.

TGSC, The Good Scents Company Information System: http://www.thegoodscentscompany.com/, cited in January 2019.

Thompson, J. M., and Perry, D. B.: A new system of refillable and uniquely identifiable diffusion tubes for dynamically generating VOC and SVOC standard atmospheres at ppm and ppb concentrations for calibration of field and laboratory measurements, Journal of Environmental Monitoring, 11, 1543-1544, 2009.

Van Kempen, G. M. P., and Van Vliet, L. J.: Mean and variance of ratio estimators used in fluorescence ratio imaging, Cytometry Part A, 39, 300-305, 2000.

Wang, T., Španěl, P., and Smith, D.: Selected ion flow tube, SIFT, studies of the reactions of H_3O^+, NO^+ and $O2^+$ with eleven $C_{10}H_{16}$ monoterpenes, Int. J. Mass Spec., 228, 117-126, 2003.

Yaws, C. L.: Handbook of Vapor Pressure. Vol 3: C8-C28 Compounds, Gulf Pub Co., Houston, 1994.

Zavarin, E., Snajberk, K., and Fisher, J.: Geographic variability of monoterpenes from cortex of Abies concolor, Biochemical Systematics and Ecology, 3, 191-203, 1975.

Addition of a fast GC to SIFT-MS for ~~analyses~~analysis of individual monoterpenes in mixtures

Supplementary Material

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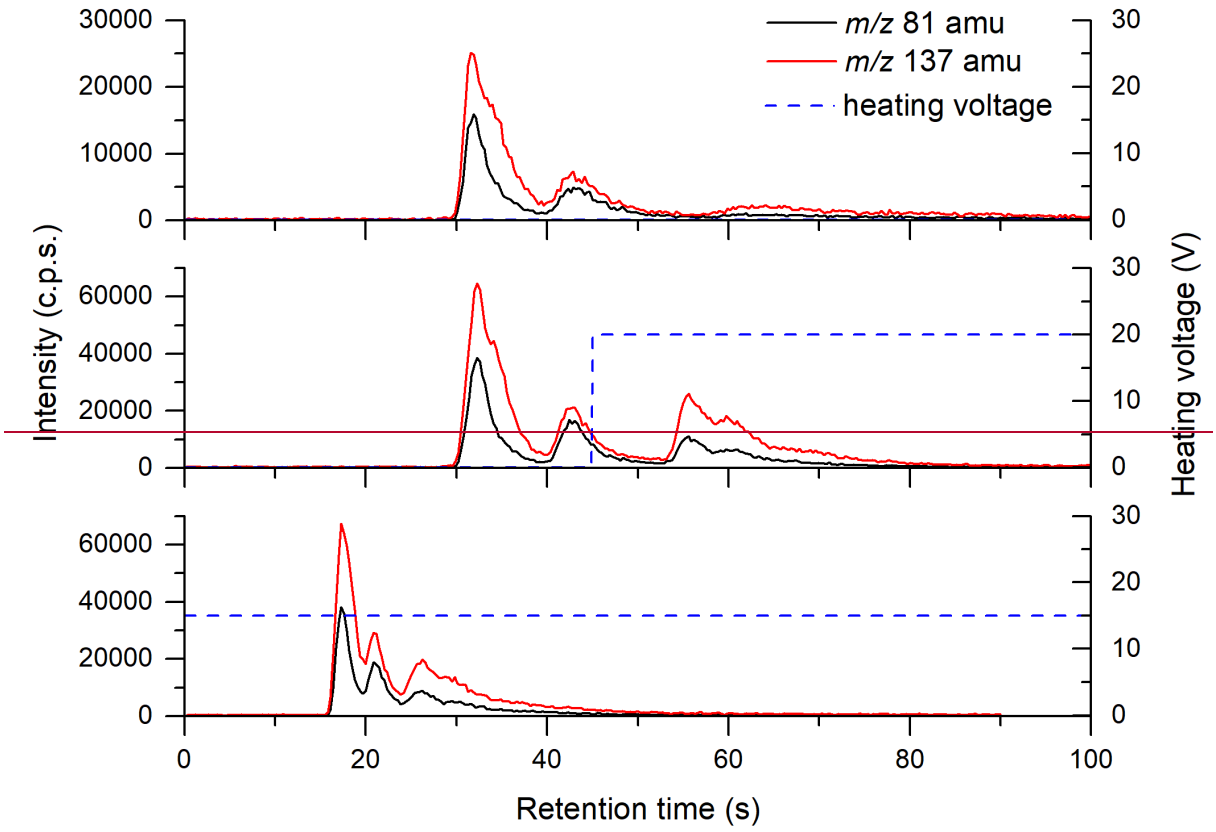
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m/z 137 amu
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Heating voltage (V)
Retention time (s)
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20000
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60000
40000
20000
0
30
20
10
0
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20
40
60
80
100

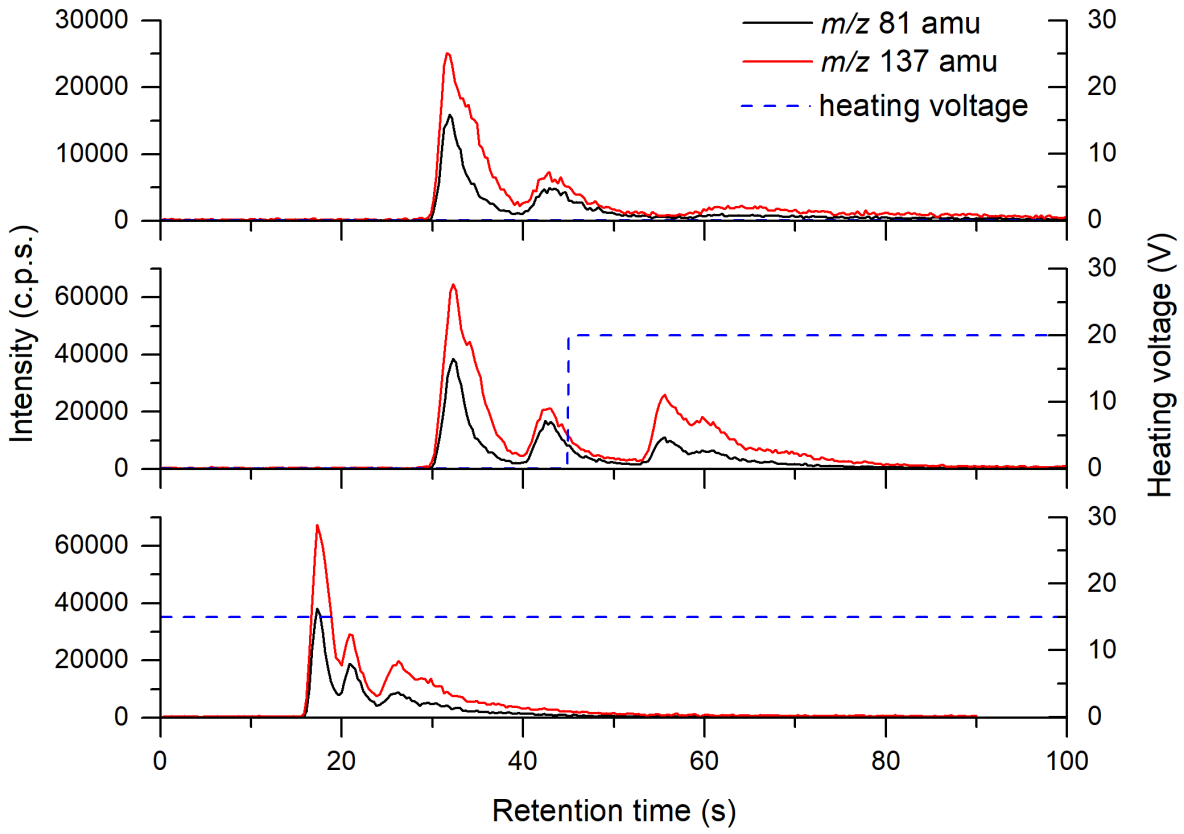


Figure S1: Chromatograms of a monoterpene mixture analysed by the MXT-1 column for different profiles of the heating voltage. Profiles were analysed by SIFT-MS using the H_3O^+ reagent ion.

Table S1: Summary of reaction rate constants and branching ratios of investigated monoterpenes. All presented rate constants have units of $10^{-9}cm^3s^{-1}$. Only significant products are given, for witch branching ratios are at least 10%.

	H_3O^+			NO^+			$O_2^{+\cdot}$		
	~~rate~~*k*	*m/z*	Products (b.r.)	*k*	*m/z*	Products (b.r.)	*k*	*m/z*	Products (b.r.)
α-pinene	2.3[a], 2.4 [a*]	81 137	$C_6H_9^+$ (30[a], 39[b]), $C_{10}H_{17}^+$ (67[a], 61[b])	2.0[a], 2.0[a*], 2.3[b]	92 136	$C_7H_8^+$ (16[b]), $C_{10}H_{16}^+$ (85[a], 77[b])	2.0[a], 1.9[a*], 2.1[b]	92 93 121	$C_7H_8^+$ (18[a], 22[b]), $C_7H_9^+$ (52[a], 56[b]), $C_9H_{13}^+$ (12[a], 12[b])
β-pinene	2.4[a], 2.6 [a*]	81 137	$C_6H_9^+$ (33[a], 40[b]), $C_{10}H_{17}^+$ (64[a], 60[b])	2.1[a], 2.2[a*], 2.1[b]	136	$C_{10}H_{16}^+$ (93[a], 89[b])	2.1[a], 2.1[a*], 2.0[b]	93 121 136	$C_7H_9^+$ (56[a], 19[b]), $C_9H_{13}^+$ (49[b]), $C_{10}H_{16}^+$ (11[a])
R-limonene	2.6[a], 2.6[a*]	81 137	$C_6H_9^+$ (22[a], 29[b]), $C_{10}H_{17}^+$ (73[a], 68[b])	2.2[a], 2.2[a*], 2.2[b]	136	$C_{10}H_{16}^+$ (91[a], 89[b])	2.2[a], 2.1[a*], 2.2[b]	68 92 93 94 107 121 136	$C_5H_8^+$ (10[b]), $C_7H_8^+$ (10[b]), $C_7H_9^+$ (26[a], 30[b]), $C_7H_{10}^+$ (11[a], 12[b]), $C_8H_{11}^+$ (11[b]), $C_9H_{13}^+$ (14[a], 13[b]), $C_{10}H_{16}^+$ (11[a], 11[b])
3-carene	2.3[a], 2.4[a*]	81 137	$C_6H_9^+$ (19[a], 24[b]), $C_{10}H_{17}^+$ (78[a], 76[b])	2.1[a], 2.0[a*], 2.2[b]	136	$C_{10}H_{16}^+$ (86[a], 81[b])	2.0[a], 2.0[a*], 1.9[b]	92 93 121	$C_7H_8^+$ (11[b]), $C_7H_9^+$ (41[a], 45[b]), $C_9H_{13}^+$ (20[a], 20[b]),

								136	$C_{10}H_{16}^+$ (14^a)
myrcene	2.6^a, 2.7^{a*}	81 137	$C_6H_9^+$ (26^a, 30^b), $C_{10}H_{17}^+$ (59^a, 58^b)	2.3^a, 2.2^{a*}, 2.2^b	92 93 136	$C_7H_8^+$ (11^b), $C_7H_9^+$ (22^a, 34^b), $C_{10}H_{16}^+$ (61^a, 55^b)	2.2^a, 2.2^{a*}, 2.2^b	69 92 93	$C_5H_9^+$ (10^b), $C_7H_8^+$ (70^b), $C_7H_9^+$ (61^a)
camphene	2.4^a, 2.6^{a*}	81 137	$C_6H_9^+$ (14^b), $C_{10}H_{17}^+$ (88^a, 86^b)	2.1^a, 2.1^{a*}, 2.3^b	136 166	$C_{10}H_{16}^+$ (87^a, 79^b), $NO^+C_{10}H_{16}$ (11^b)	2.0^a, 2.1^{a*}, 2.2^b	93 107 121	$C_7H_9^+$ (13^a, 19^b), $C_8H_{11}^+$ (10^b), $C_9H_{13}^+$ (44^a, 49^b)
α-terpinene		81 137	$C_6H_9^+$ (10^b), $C_{10}H_{17}^+$ (87^b)	2.0^b	136	$C_{10}H_{16}^+$ (87^a, 99^b),	2.0^b	93 121 136	$C_7H_9^+$ (16^b), $C_9H_{13}^+$ (42^b), $C_{10}H_{16}^+$ (33^b)
γ-terpinene		81 137	$C_6H_9^+$ (17^b), $C_{10}H_{17}^+$ (81^b)	2.1^b	135 136	$C_{10}H_{15}^+$ (18^b), $C_{10}H_{16}^+$ (87^a, 75^b),	1.9^b	92 93 121 136	$C_7H_8^+$ (12^b), $C_7H_9^+$ (46^b), $C_9H_{13}^+$ (21^b), $C_{10}H_{16}^+$ (14^b)

[a] (Schoon et al., 2003); [b] (Wang et al., 2003); [c] Present result based on SIFT-MS measurements; [d] Present result based on fastGC-SIFT-MS measurements; * theoretical data based on the method of Su and Chesnavitch (Su and Chesnavich, 1982); b.r. stands for branching ratio; Dimension of rate constants is $10^{-9}cm^3s^{-1}$.

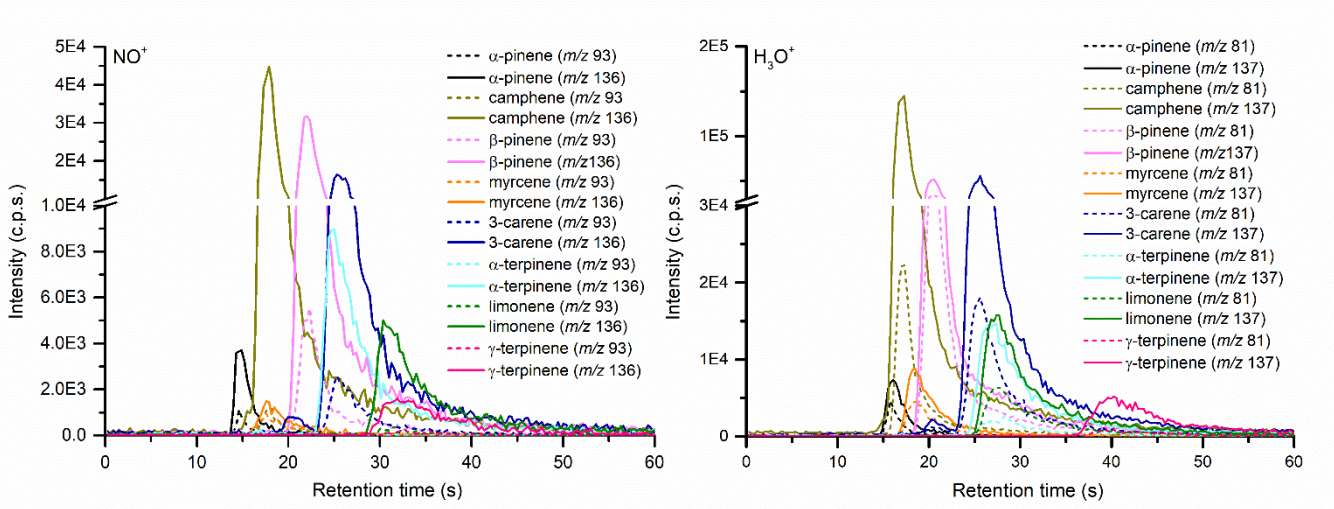
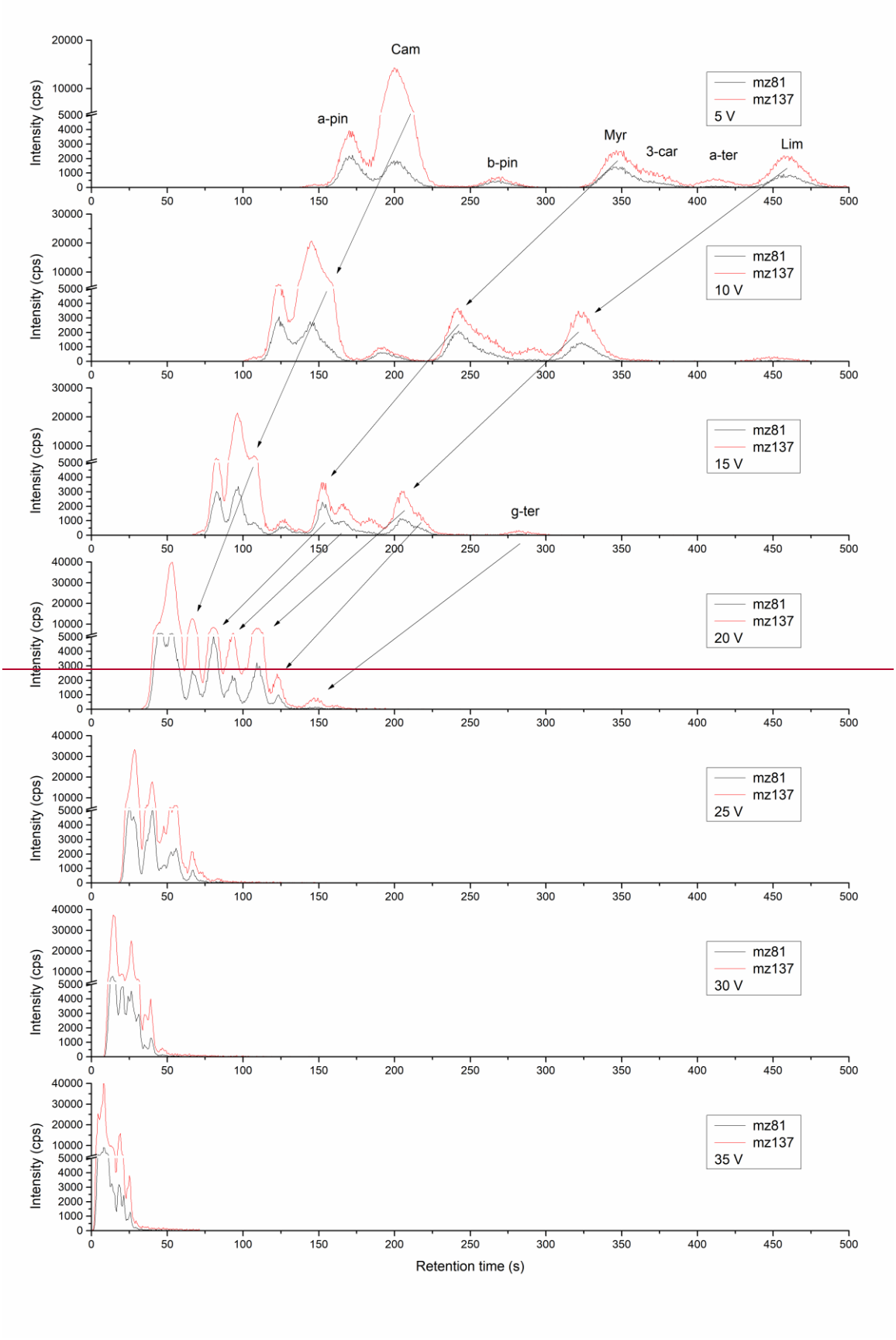


Figure S2: Chromatograms of individual monoterpenes analysed using the MXT-1 column at a constant temperature of column ~40 °C. The profile is associated with the profile shown in the bottom of Figure S1. Profiles were analysed by SIFT-MS using the H_3O^+ reagent ion. Intensity of α-pinene was reduced.


Cam
a-pin
b-pin
Myr
3-car
a-ter
Lim
g-ter
mz81
mz137
5 V
10 V
15 V
20 V
25 V
30 V
35 V
Intensity (cps)
Retention time (s)

~~Figure S3~~

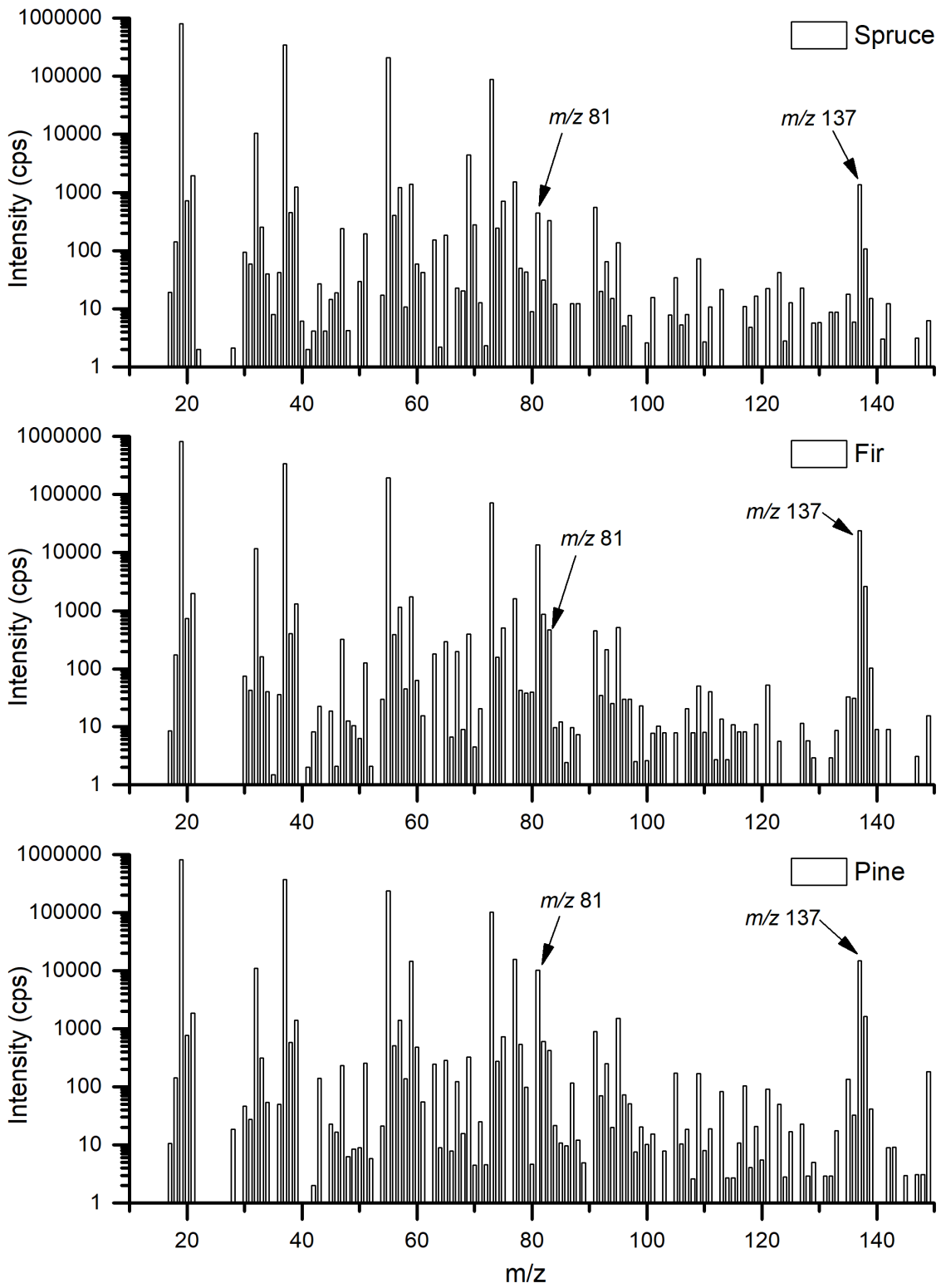


Figure S3: SIFT-MS spectra of coniferous samples analysed by H_3O^+ reagent ions. The marked ions with m/z 81 and m/z 137 were used for analysis of monoterpenes.

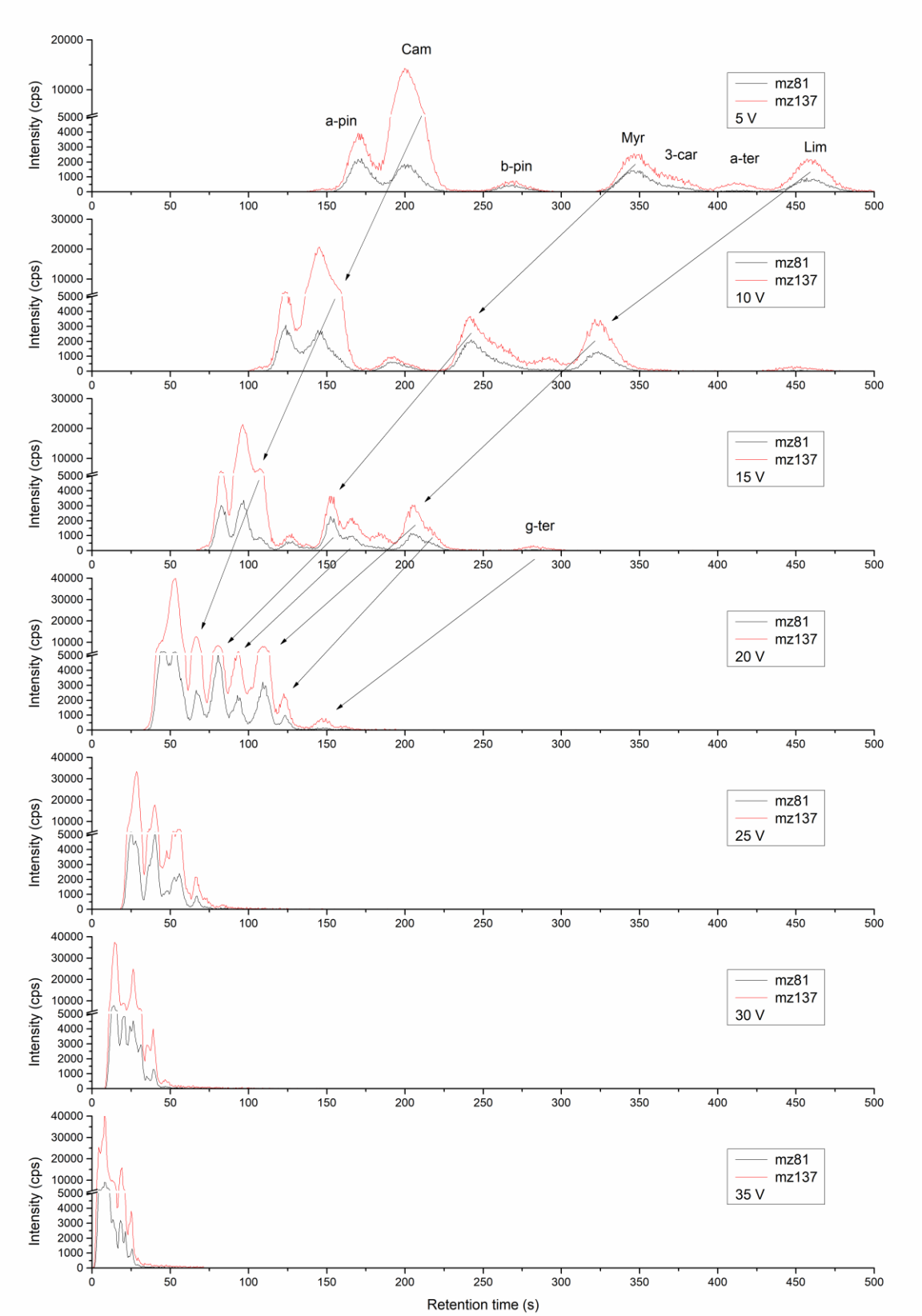


Figure S4: Chromatograms of a monoterpene mixture analysed by the MXT- Volatiles column for different heating voltages. Profiles were analysed by SIFT-MS using the H_3O^+ reagent ion.

~~Figure S4: Sample no.~~

~~1 (Pinecea punges)~~

Figure S5: Sample no. 1 (Pincea punges)

Figure S6: Sample no. 2 (Abies concolor)

Figure ~~S6~~S7: Sample no. 3 (Pinus nigra)

References

Schoon, N., Amelynck, C., Vereecken, L., and Arijs, E.: A selected ion flow tube study of the reactions of H_3O^+, NO^+ and O_2^+ with a series of monoterpenes, International Journal of Mass Spectrometry, 229, 231-240, 2003.

Su, T., and Chesnavich, W. J.: Parametrization of the ion–polar molecule collision rate constant by trajectory calculations, The Journal of Chemical Physics, 76, 5183-5185, 1982.

Wang, T., Španěl, P., and Smith, D.: Selected ion flow tube, SIFT, studies of the reactions of H_3O^+, NO^+ and O_2^+ with eleven $C_{10}H_{16}$ monoterpenes, Int. J. Mass Spec., 228, 117-126, 2003.