We thank the reviewer for the evaluation of the manuscript. The manuscript was revised according to the reviewers' comments. Here the point-by-point answers are given. The reviewers' comments are in bold and our answers are in normal font. The answers are divided into author's response (AR) and author's changes to manuscript (ACM).

Anonymous Referee #2 Received and published: 4 March 2020

The study by Helin et al. describes in depth the analytical method of a thermal desorption-gas chromatography-mass spectrometry (TD-GC-MS) to quantify a series of Biogenic Volatile Organic Compounds (BVOC). While the core analytical method has been previously used extensively, such a detailed description of the processes and parameters affecting the sampling collection is a valuable addition. The authors focus their investigations to diterpenes, a highly understudied class of compounds that exhibits difficulties in quantification due to their volatility, low abundance, sticky nature and experimental artefacts. The paper reads well but few critical points deserve more attention. Therefore, the paper is suitable for AMT and I could recommend publication but only after addressing the following comments/concerns.

General comments

1. There seems to be a confusion on the terminology that starts from the title itself and extends to various points of the text. Terpenes are a class of compounds that include monoterpenes, sesquiterpenes and diterpenes. However, it's true that in the literature terpenes and terpenoids are frequently used in the same bases. But monoterpenoids, sesquiterpenoids and diterpenoids are actually modified terpenes, with different chemical formulas (eg. Chan et al., 2016). I would recommend changing the title to the more general "Biogenic Volatile Organic Compounds" or "Terpenoids". Consequently, individual corrections on the text over case-based referral to the individual species is needed.

AR: We agree with the Reviewer that in the literature a somewhat mixed use of terpenes and terpenoids is unfortunately common. In the revised manuscript we have changed the naming and added to all Tables the compound categories to which we refer to. This helps the reader to trace all the compounds to which we are referring to. In addition, we changed the referred title part to "…monoterpenes, sesquiterpenes and diterpenes". This was considered to being more representative of the main work.

ACM: The terminology has been changed throughout the manuscript. As an example, the compound categories were added Section 2.1. and Table 1: "The analyte included nine MTs (both hydrocarbon and oxygenated MTs), six SQTs (both hydrocarbon and oxygenated SQTs), three diterpenes (all hydrocarbon DTs) and three other compounds that were categorised here as BVOCs."

Revised Table 1:

Table 1. List of analytes, corresponding retention indices (RI) and analytical figures of merit.

Compound	CAS Number	Formula	RI[a]	Quan-tify m/z ion	LOQ in pg (in pptv)[b]	Repeatability (RSD%)	Inter-mediate precision (RSD%)	U[c] (%)	U[d] (%)
α-Pinene (MT)	7785-70-8	$C_{10}H_{16}$	945 (942)	93	34 (1.2)	2.6	3.8	16	18
camphene (MT)	79-92-5	$C_{10}H_{16}$	963 (960)	93	14 (0.5)	1.6	4.0	16	18
β-pinene (MT)	19902-08-0	$C_{10}H_{16}$	989 (988)	93	13 (0.5)	4.2	3.2	17	20
3-carene (MT)	498-15-7	$C_{10}H_{16}$	1018 (1017)	93	18 (0.6)	2.0	3.3	16	17
p-cymene (MT)	99-87-6	$C_{10}H_{14}$	1033 (1031)	119	20 (0.7)	2.0	4.2	17	19
limonene (MT)	5989-54-8	$C_{10}H_{16}$	1039 (1037)	68	62 (2.2)	1.5	3.8	17	17
1,8-cineol (MT)	470-82-6	$C_{10}H_{18}O$	1044 (1042)	154	27 (0.9)	2.6	4.7	18	20
terpinolene (MT)	586-62-9	$C_{10}H_{16}$	1094 (1093)	121	65 (2.3)	2.5	5.0	17	18
linalool (MT)	78-70-6	$C_{10}H_{18}O$	1100 (1100)	71	88 (2.8)	2.4	4.8	18	18
4-acetyl-1-methylcyclohexene (4-AMCH) (BVOC)	6090-09-1	$C_9H_{14}O$	1140 (1139)	95	120 (4.2)	2.7	6.3	19	20
nopinone (BVOC)	38651-65-9	$C_9H_{14}O$	1154 (1153)	83	29 (1.0)	2.7	5.3	18	21
bornylacetate (BVOC)	5655-61-8	$C_{12}H_{20}O_2$	1295 (1294)	95	49 (1.2)	2.4	6.2	20	23
longicyclene (SQT)	1137-12-8	$C_{15}H_{24}$	1401 (1401)	94	35 (0.8)	2.4	4.9	18	21
isolongifolene (SQT)	1135-66-6	$C_{15}H_{24}$	1422 (1422)	161	38 (0.9)	2.5	5.7	19	22
β-caryophyllene (SQT)	87-44-5	$C_{15}H_{24}$	1445 (1444)	93	105 (2.5)	2.2	3.5	17	22
β-farnesene (SQT)	18794-84-8	$C_{15}H_{24}$	1457 (1456)	69	198 (4.7)	5.0	5.9	20	22
α-humulene (SQT)	6753-98-6	$C_{15}H_{24}$	1481 (1480)	93	33 (0.8)	2.1	4.2	18	24
caryophyllene oxide (SQT)	1139-30-6	$C_{15}H_{24}O$	1612 (1612)	79	340 (7.5)	18.9	8.6	24	28
cembrene (DT)	1898-13-1	$C_{20}H_{32}$	1959 (1959)	93	287 (5.1)	4.0	9.5	27	52
ent-kaurene (DT)	562-28-7	$C_{20}H_{32}$	2106 (2106)	257	518 (9.3)	2.9	7.7	26	42
3-methylene-5-α-androstane (3-MA)[e] (DT)	not assigned	$C_{20}H_{32}$	2153 (2152)	257	383 (6.9)	3.0	7.2	-	-

[a] RI as Kovats indices (and arithmetic indices in brackets)

[b] LOQ in pptv calculated by using sample volume of 5 L (at T=298 K and p=1 atm)

[c] U (%) for ambient air application

[d] U (%) for branch enclosure application

[e] The purity of this crude standard was set to being 100% (allowing the determination of LOQ, but unknown uncertainty)

2. My most serious concern has to do with the striking claim that there is no need for heating the Teflon lines in field applications, in contrast to almost all experimental approaches in the field to date. That conclusion was derived performing sampling recovery tests over which a second sample was taken subsequent to the original (L159: “Inlet line recovery was calculated simply by the relationship of analytes found in the second tube divided by the amounts in the first tube.”) One would naturally expect that when the line is kept at the same temperature, artifacts cannot be evaluated properly. The losses on the line could be better illustrated if after the first sample, a subsequent “blank” sample over a now heated line is taken. In this approach, the amount of VOCs stuck in the tube could be quantified and the proper temperature for heating up the lines could be ascertained.

AR: We agree with the Reviewer. The original sentence was too generalizing. It was meant that the Teflon FEP sampling line does not need to be extensively heated. It was not intended to mean that non-heated Teflon FEP lines could be used e.g. during cold spring. We revised this part so that our results better corroborate the statement and removed the field applications part from the sentence. In the revised manuscript there is no claim of non-heated lines usage in field measurements anymore.

ACM: “The results indicate that under the applied conditions the Teflon FEP sampling lines do not need to be heated excessively above room temperature (22±1 °C)”

3. Similar to the previous comment, the authors describe in S1.5 the problems that arise with the replacement of the Teflon line. It seems that a new line is causing more artefacts comparing to a used one, indicating some saturation over the line surfaces. In general, the inlet line experiments should be explained in greater detail. The claim that Teflon lines should remain unheated for field applications has the potential to mislead future research and therefore it cannot become acceptable unless strong and well described proof of concept is presented.

AR: The reviewer is correct. In the original manuscript we had a sentence that was too generalizing, and it could have been interpreted in a wrong way. It was not our intention to mislead the reader. The sentence was modified as shown in above point 2. ACM part.

The inlet line experiments are explained quite thoroughly in Section 2.3.3 and in the schematic illustrations of the setups presented in the supplementary (the information is presented so that others can reproduce the experiments). This was not really expanded, since upon revision, no missing information could be identified. Also, in that Section 2.3.3 another study by Hellen et al. (2012) is cited, where a similar experimental setup was used.

Regarding the Supplementary S2.5 (in the revised version it is Text S2), it is true that the line changes initially caused a drop in the overall recovery levels. The lines and connections were checked a few times to identify leaks, which were thought to be the reason, but we could not verify the leaking for sure. It was left a bit open question was the improvement overtime really caused by the “saturation” as the reviewer puts it or by the done leak checking open/closing connections. Since the leak checking information was not really scientific nor fitted the text, it was not included originally in the S2.5. However, due to this comment, it was considered to being relevant and was added to the supplement text.

Change to supplement: “It is unknown if the initially observed drop in overall recoveries was caused by some active surfaces in the Teflon line (which became saturated over time) or by potential loose connections upon line changes (connections were open/closed separately a few times to investigate possible leaking).”

4. There is a lot of useful information, results and discussion in the supplementary information. Sometimes, it becomes challenging to follow the text while browsing through Figures, Supplementary Figures, additional discussion and references. I would recommend to bring forward the supplementary discussion, including several of the figures.

AR: In the original manuscript, we attempted to keep the main text and figures as compact as possible and to present all complementary information in the supplementary information (SI). We wish to keep it so for the most part. However, in view of one of the Reviewer #1 comments and this comment, the supplementary figure showing the cuvette results was raised from the SI to the manuscript in order to help the reader better transit between the text and the plot for results interpretation purposes. In addition, upon revision, it was considered that the real sample chromatograms could fit in the manuscript and that these could help the reader in visualizing the results, thus those were also raised from the SI to the manuscript.

ACM: Three supplementary Figures were raised to the manuscript (now these are Figs. 3, 6 and 7). The Fig. 3 shows the cuvette recovery results, while Figs. 6 and 7 are the chromatograms from pine needles and spruce twigs extraction experiments, respectively (revised by adding blank chromatograms also to those).

5. In order to derive some quantitative emissions of diterpenes from needles, the experiments (both in lab and field) were performed under unrealistically high temperatures. While this is acceptable for validating the method, it should be further noted that these are stored compounds coming for the pools of the plant material and further highlight the reasons for this approach. Some investigations between the vapor pressure and measured emissions would be nice addition to the paper, if possible.

AR: Thank you for pointing this out. The information about the emissions coming from storage pools was added to justify the approach. This was added to Sections 2.4 and 3.2. (related also to Reviewer #1 comments).

In general, we agree with the Reviewer that some investigation between vapor pressure and measured emissions could be nice addition, however, we are not sure how it would fit into this manuscript and what it would contribute eventually to the overall manuscript. As an example, in Figs. AR1 and AR2 are presented the plots of emissions rates as a function of vapor pressures from the pine needles and spruce twigs experiments, respectively. The vapor pressures (in mmHg at 25 °C) were predicted values obtained via EpiSuite (https://www.epa.gov/tsca-screening-tools) and are presented only for those compounds that could be tentatively identified. The emission rates are presented at 30 °C and 60 °C as observed in the laboratory experiments performed in our study (e.g. in Fig. AR1 one of the tentatively identified diterpenes was emitted only at 60 °C, therefore the point is missing from the 30 °C results). In the plots, both x- and y-axis are presented in logarithmic scales.

As can be seen in these Figs. AR1 and AR2, the emission rates are the highest for the most volatile compounds investigated (monoterpenes) and the lowest for the least volatile compounds investigated (diterpenes). In principle, this is a fairly trivial result and does not necessarily tell anything meaningful about, for example, the terpene content of the plant storage pools. There could be also some uncertainty in the predicted vapor pressure values used here. In the end, we decided not to present these plots in the manuscript. In our opinion, the added value would have been minor and it would have not fitted into the main scope of our study.

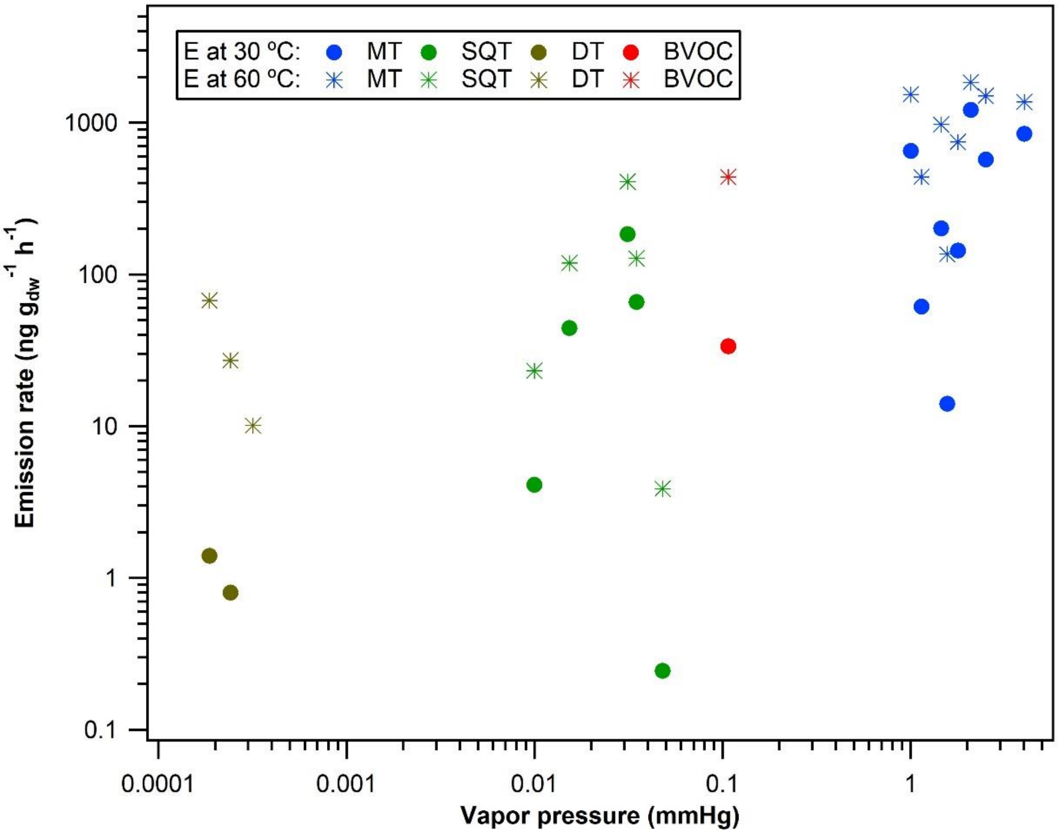

Figure AR1. Relationship between emission rates (E) and vapor pressure in the emissions from pine needles laboratory experiments.

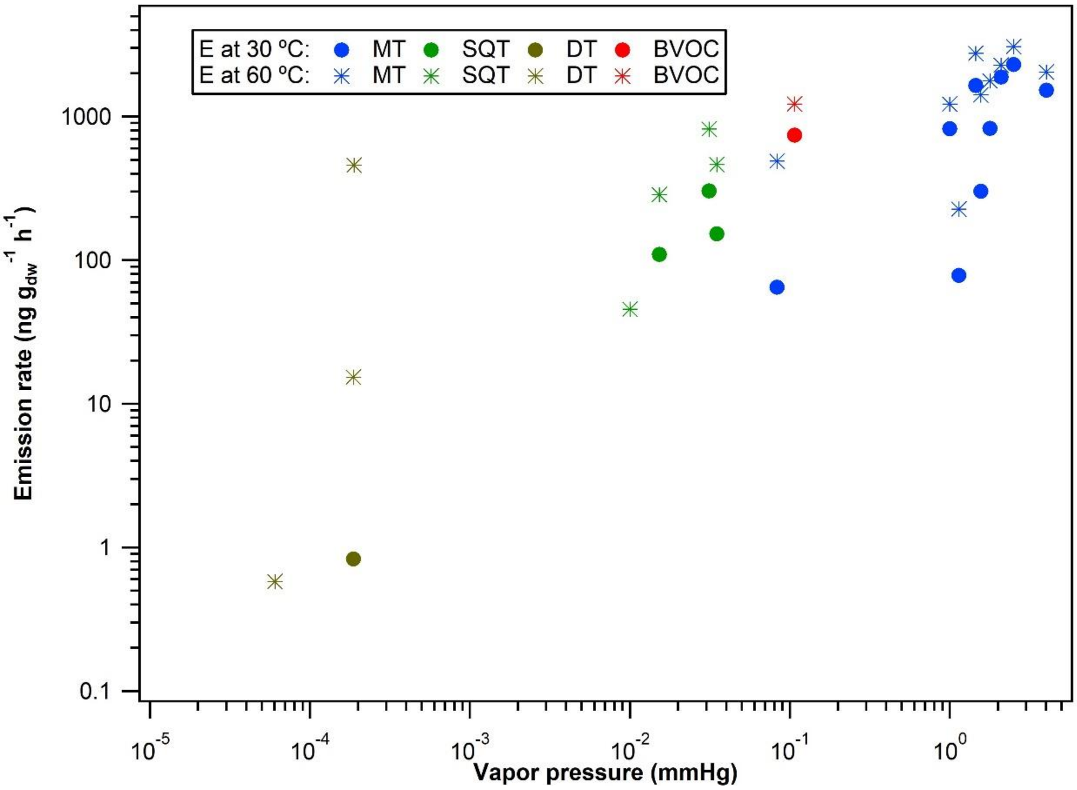

Figure AR2. Relationship between emission rates (E) and vapor pressure in the emissions from spruce twigs laboratory experiments.

ACM: The beginning of Section 2.4 was rewritten and modified:
"Two types of real sample applications were selected in order to test the analytical method performance, to identify compounds emitted by boreal forest tree species and to quantify the emissions from a branch enclosure. The first sample application was a dynamic headspace sampling of compounds emitted by needles/twigs in a closed chamber upon heating, and the second sample application was a study of emissions from a branch enclosure in field conditions. The main emphasis on real sample applications was set on the identification and study of DTs, although MTs and SQTs were also monitored. Based on previous studies, it was considered that since the DTs seem to be emitted from plant storage pools and since temperature has been shown to be a driving force for DTs emissions (Haberstroh et al., 2018;Matsunaga et al., 2012;Yáñez-Serrano et al., 2018), heating of the plant materials would be an efficient way for releasing the DTs into gas-phase. Thus, the studied plant material samples were firstly heated to temperatures ranging from 30 °C to up to 70 °C in order to obtain presumably detectable amounts of DTs to allow their identification and quantification."

In the second paragraph of Section 3.2 the following was added:
"As explained in Section 2.4., in the following experiments the plant material samples were heated in order to characterize the DTs potentially emitted by the studied tree species and to further test the TD-GC-MS method performance in view of real sample analysis. The plant material samples were heated to facilitate the release of DTs from the plant storage pools into the gas-phase."

Specific comments

L34-40. The authors probably refer to the comparative reactivity method. If so, the reference of Kovacs and Brune is misplaced. Instead the study by Sinha et al (2008) should be cited.

AR: Yes, it should be Sinha et al. (2008) not Kovacs and Brune.

ACM: Reference Kovacs and Brune removed and was changed to Sinha et al. (2008).

L93. Please make a comment on how the commercial adsorbent tubes compare with the custom-made ones.

AR: In this work, the commercial and custom-made tubes were used randomly. For example, the intermediate precision (n=22) and blanks (n=10) used for LOQ determination contain results from both types of tubes. This information was added.

ACM: "The commercial and in-house prepared tubes were used collectively and randomly in this work."

L154-155. Zero air generators and carbon cartridges have frequently shown that are not completely scrubbing the VOCs. Have you tested the efficiency of these methods? How do they compare between them?

AR: In our case both purification systems worked fine for our purpose, at least they scrubbed the BVOCs we were investigating. In this work, we did not compare the different purification setups.

This is said in the manuscript in Section 2.5: "In both real sample applications, the C_{in} was negligible (or <LOQ) for all compounds, and was subsequently set to zero in the calculations."

L182&L239. Please use equations in separate lines so they are clearly distinguishable from the rest of the text.

AR: Done.

ACM: Equations were added to separate lines.

L375. O3 removal. Have you tested other scrubbers? Why the heated stainless steel in the optimal solution of ozone removal and not e.g. KI scrubbers or Na2S2O3 infused quartz filters? Please refrain from strong claims that are not supported by experiments.

AR: Discussion and references on ozone removal techniques was added. Changes were made to avoid too strong claims, e.g. the "optimal solution" was replaced with "good solution".

ACM: This part was added "Several different kind of ozone traps and filters have been used to trap ozone, but most of them are not suitable even for MTs and SQTs (Polmann et al. 2005, Calogirou et al. 1996). In addition, some of the ozone removal techniques have very short ozone removal capacity and they have to be changed frequently and therefore are not ideal for online sampling (Bouvier-Brown et al. 2009, Fick et al., 2001)."

In addition the sentence was modified a bit "Based on the ozone removal results presented in Hellén et al. (2012) and the inlet recovery results presented in this study (Fig. 2d), heated stainless-steel line could be a good solution for ozone removal especially in online sampling." to avoid too strong claims.

The following were added to References:

Calogirou, A., Larsen, B.R., Brussol, C., Duane, M., Kotzias, D., 1996. Decomposition of terpenes by ozone during sampling on Tenax. Analytical Chemistry 68,1499-1506.

Fick, J., Pommer, L., Andersson, B., Nilsson, C., 2001. Ozone removal in the sampling of parts per billion levels of Terpenoid compounds: an evaluation of different scrubber materials. Environmental Science and Technology 35, 1458-1462.

L386. Please make a comment on the saturated peaks of Sup.Fig.19-20.

AR: Those are not saturated peaks, the chromatograms are just scaled in such a way that all the peaks are visible. The comment clarifying this is present in the respective captions, e.g.: "The chromatogram is scaled for presentation purposes, peak number 3 is not showed in full height in order to visualize the other peaks more clearly." These Figures are now in the manuscript, Figs. 6 and 7. No changes were made regarding this since the information is in the captions.

L463. Similar to my major comments, it has not been sufficiently demonstrated that the ambient sampling DTs does not require heating lines. One potential experiment to help towards this direction would be to sample through a long inlet line the analyte mixture at different temperatures and present a plot of inlet temperature vs signal.

AR: This sentence was removed and the paragraph was revised. The last sentence of the paragraph was changed.

ACM: "These sampling recovery results of DTs were promising, since they demonstrated that the DTs were not lost in excessive amounts in the sampling lines tested."

References Chan, A. W. H., Kreisberg, N. M., Hohaus, T., Campuzano-Jost, P., Zhao, Y., Day, D. A., Kaser, L., Karl, T., Hansel, A., Teng, A. P., Ruehl, C. R., Sueper, D. T., Jayne, J. T., Worsnop, D. R., Jimenez, J. L., Hering, S. V., and Goldstein, A. H.: Speciated measurements of semivolatile and intermediate volatility organic compounds (S/IVOCs) in a pine forest during BEACHON-RoMBAS 2011, Atmos. Chem. Phys., 16, 1187–1205, https://doi.org/10.5194/acp-16-1187-2016, 2016.

Sinha, V., Williams, J., Crowley, J. N., and Lelieveld, J.: The Comparative Reactivity Method – a new tool to measure total OH Reactivity in ambient air, Atmos. Chem. Phys., 8, 2213–2227, https://doi.org/10.5194/acp-8-2213-2008, 2008.