Interactive comment on "The atmospheric potential of biogenic volatile organic compounds from needles of White Pine (Pinus strobus) in Northern Michigan" by S. Toma and S. Bertman

We would like to thank both reviewers for their time and insightful comments. Both reviewers were thorough and thoughtful and have made some excellent suggestions. Some of the suggestions are beyond the scope of this study. Most are very helpful and have been incorporated in a revised manuscript.

Our main changes are:

1. *Figure S1 was moved from the supplementary material and inserted in the main paper as Figure 1. Therefore, original Figures 1 and 2 are changed to Figures 2 and 3.*
2. *We modified Figure 2 (Figure 3 in the current version) Figure S3 (Figure S2 in the current version)*
3. *At least 9 new citations were added.*
4. *All suggested grammatical and editorial changes were made and the revised manuscript has been further proofread.*

Suggestions from Anonymous Referee #2

This study examines the influences of location within the canopy, year, and phenotypic differences on needle concentrations of terpenoid volatile organic compounds. It attempts to apply these results to changes in atmospheric chemistry, and it also presents some interesting results of a little studied, highly reactive sesquiterpene species found within the needles. This latter result is of special interest, given recent papers on the unknown OH reactivity in forests (e.g. Sinha et al., 2010). However, I think that for this paper to be published, the introduction and some of the results sections may need to be rewritten. Many references are needed to motivate the methods adopted within this paper. Finally, the many ideas contained within this paper seem very scattered, and the order of presentation of the data is not clear. For the rest of this review, I refer to pages by the last two letters, e.g. 26852, Line 10 will be replaced by P52L10, and I use MT to abbreviate 'monoterpenes', as in the paper.

Abstract: When reporting percentages, it would help to state the number of samples.

We agree and have added the number of samples after the reported percentages on P50L12. The wording now is "About 15 % (n=13) of the trees showed high levels".

Introduction: If the potential for VOC to serve as a source of aerosols is relevant for this study, I recommend devoting a paragraph to this subject, and citing additional sources, such as Hao et al.(2011 -see references). It would be extremely helpful to clearly delineate when the subject is 'BVOC content' of a forest, meaning the concentrations of BVOC found within leaves, BVOC emissions, meaning the amount of BVOC emitted from leaves into the ambient atmosphere, and BVOC ambient concentrations, which is the concentration of BVOC found in the atmosphere. In the introduction, these ideas appear together, without clearly explaining the link between them.

Additionally, it is unclear if the focus is on 'forest health' or 'air quality and production of aerosols' (P51L29).

We revised the first paragraph of the Introduction to clarify the important potential for VOC serving as a source of aerosols

"Biogenic volatile organic compounds (BVOC) lead to the formation of tropospheric ozone in the presence of NOx (Atkinson, 2000) as well as to formation and growth of secondary organic aerosol (SOA) (Andreae, 2009; Laothawornkitkul et al., 2009). The total global estimated emission rate of BVOC is about 1.2×10^{15} g C per year, while the global anthropogenic VOC emission rate is about 1.0×10^{14} g C per year (Muller, 25 1992; Guenther et al., 1995). The BVOC emissions in North America are dominated by isoprene (51%), terpenes (31 %), and oxygenated VOC (16 %), based on OH reactivity (Guenther et al., 2000). The organic component of aerosols is the largest single fraction with an average of 45% (Zhang et al., 2007) and recent research suggests that most of the OA mass is secondary (Jimenez et al., 2009). BVOC are believed to be the dominant contributors to global SOA formation due to their large global emissions and high reactivity with major oxidants in the atmosphere (Nga et al., 2006). The large mass of emission and atmospheric reactivity makes estimation of BVOC emission rates important. Foliage is the largest source of total global BVOC emissions, accounting for over 90% (Guenther et al., 1995). Hao et al. (2011) directly measured SOA yields from oxidation of real plant emissions and suggest that the specific chemical profile of BVOC emissions affects the SOA outcomes. Specific BVOC compound composition depends on the type of ecosystems, plant species composition, and environmental conditions. Peñuelas and Llusia (2001) suggested that emissions are controlled by plant VOC concentration, VOC vapor pressure or the resistance to VOC diffusion to the atmosphere. The factors controlling BVOC emissions are complex, and it is still difficult to evaluate their relationship."

Materials and Methods: I recommend stating in this section that the forests are generally identical, as noted later in the paper on P53L10.

The section was revised to state this.

"Two forest sites located within a kilometer of each other in the same environment with similar soil conditions and with similar forest cover types were sampled."

If you need to refer to something in the supplement multiple times, it may be best to include it in the paper, so it may be best to include figure S1 in the actual paper, rather than the supplement.

As suggested, we have moved Figure S1 into the body of the paper as Figure 1 to more clearly indicate the proximity of the samples in the forest. This proximity is one argument we are making against environmental induction of chemotypic variation.

Section 3.1: It is not evident to this reviewer how the different biosynthetic pathways relate to the ambient summer temperatures. This last paragraph needs to be clarified, and should probably have additional sources cited.

The reviewer is right that we do not have evidence about how biosynthetic pathways might be specifically driven by ambient temperature. However, temperature varied greatly from year to year and we are suggesting that this environmental factor is a representative of different overall growing conditions that might have influenced the absolute levels of compounds measured. Given that monoterpenes and sesquiterpenes are produced in different parts of the cell, they could be differently sensitive to environmental conditions. We are not aware of citations that specifically look at temperature effects on partitioning of BVOC into MT and SQT. Environmental conditions appear not, though, to affect the ratio of limonene/pinene. That is, the same trees expressed Mode II in all years despite large variation in environmental conditions.

Section 3.2: Note that although the light environments are different, there can also be a temperature difference of several degrees between the top and bottom of a forested environment (e.g. Gu et al., 1999).
If the number density of insect larvae varies with altitude, you should provide some evidence.

We have included the idea of temperature differences by height in forests as suggested, "A temperature gradient of several degrees can exist between the top and bottom of a forested environment (e.g. Gu et al., 1999)."

The suggestion that insect number density might have an influence on terpene levels is purely hypothetical/speculative and not an inference drawn from our data. We did not measure insect density. To avoid the suggestion that we have those data, we removed reference to this idea from the manuscript.

Additionally, it would be interesting to see a direct comparison of overstory and understory trees, I note that only in 2008 are the 'bottom' overstory concentrations of MT+SQT close to the concentrations in the understory. Are the concentrations from the 'lower' overstory trees statistically significantly different from the concentrations in the understory trees? If not, this would lend confidence to the conclusion about the changes in terpene concentrations as the understory becomes the overstory.

This is a good idea. A comparison of needles from the bottom of canopy trees and needles from understory trees for total MT, total SQT, and total BVOC in 2008-2010 using non-parametric Mann-Whitney Tests shows no statistical difference with $P<0.05$. We have included this phrase in the text to support the idea that terpene concentrations will change as the understory becomes the overstory.

Section 3.3: It makes sense to note that the difference between BVOC concentrations reported in the gas-phase and BVOC concentrations measured in needles might be due to the volatility of different terpenes, since to first order, the high concentrations where the BVOCs are produced result in diffusion to the lower concentration areas (the ambient atmosphere) where the BVOCs are deposited or reacted away. A big-leaf model (e.g. Pleim and Ran, 2009) might help make this matter more clear.

The specific profile of different terpenes will be influenced by differences in volatility, and non-volatile compounds seen in the needles could be absent from the gas phase, but it is unlikely to

find a compound in the gas phase if it is not seen in the needles. So our data represent a baseline "pool" of compounds that might be seen in the air of the forest. It is important to measure BVOC in the gas phase and in needles at same time, which we are working on. While big-leaf modeling would be very interesting, we feel it is beyond the scope of this work. We have revised this section to include the idea of volatility differences influencing gas phase concentrations.

Section 3.4: I strongly recommend that this section be extended, in light of the many papers about missing OH reactivity above forests. The authors state 'little is known about its gas-phase properties'. I recommend the authors leave no stone unturned in searching the literature for references to the atmospheric effects. Some additional background, or some calculations, would strengthen your conclusions.

We have found several references to germacrene D-4-ol in various literatures (including ecological and synthetic organic), with varying levels of sometimes contradictory evidence of its structure (primarily MS). This compound is cited several times in analyses of essential oils. It has been reported twice in leaves and sap of pines, but there are no reports we can find discussing measurements of gas-phase properties or atmospheric presence. Estimated/calculated physical properties suggest that it is a high-boiling compound with a vapor pressure much lower (5 orders of magnitude) than that of the monoterpenes and about 300x lower than that of germacrene D. While this suggests that the compound may not be found in high concentration in the gas phase, it could exist on forest surfaces and contribute to OH reactivity. Also, dehydration of this compound yields germacrene, which could volatilize more readily. We have extended this section, as suggested.

Section 3.5: The conclusions of this section – that there can be phenotypic differences in production of BVOCS within a tree population is interesting, but this section needs significant improvement. It would help, first of all, to present some of the literature that have also found genotypic variation in BVOC production within a population of trees.

Plant species can exhibit variation in VOC profile between individuals in populations, which is of interest largely to the essential oils industry (e.g. Keszei et al., 2010; Tamir et al., 2011). For instance, six chemotypes of foliar monoterpenes were observed in the Australian Tea Tree (Keszei et al., 2010) and the frequency of chemotypes in this plant varied by sampling site, however sesquiterpene formation was independent of the monoterpene chemotype of an individual. Similar chemotypic variation in high emitting trees, such as the pines, could affect atmospheric chemistry depending on the extent and magnitude of the variation. Establishing a genetic basis for gemotypic variation can be challenging (Shelton et al., 2002), although expression of the genes involved in BVOC synthesis positively correlate with their emission rates. There is still much to be learned about the regulation of BVOC synthesis rates (Laothawornkitkul et al. (2009). Because there is a high degree of genetic variability in white pine, which occupies a wide distribution over very diverse environments (Buchert, 1994), it is reasonable to expect variation in BVOC production within a population. We have revised this section to include these ideas.

I question the statement that trees of both modes were within 2m of each other at UMBS, since not all of the trees used in Figure 2 seem to be labeled on the map in Figure S1. To fix this, you may want to label all the trees in the map, produce a table with each tree number, which analysis it was used in, and what its 'mode' was in Figure 2.

We have labeled all Mode II trees in Figure 2 (new Figure 3) and have made sure that all Mode II trees are clearly marked in Figure S1 (new Figure 1). The distance between trees of different modes that were sampled varied, so not all Mode II trees had a measured Mode I tree within 2m, but some did.

Additionally, it would help to explain why 2008 was not included in the analysis. It seems like different years and trees are used for different analyses, and little scientific motivation is provided. As an example, tree S16 is present in the seasonal trend study, but S41 is not especially since it was one of the more clearly 'defining' trees for the second mode.

The original observation that some trees have anomalous limonene levels was made in 2008, but we did not achieve GC resolution between limonene and phellandrene. Therefore the 2008 data, while showing the same overall trends, are slightly complicated by the overlapping peaks. We resampled in 2009 and 2010 and modified the analytical methods to give baseline separation and therefore cleaner data. Data from multiple months in a season (Figure S3 (new Figure S2)) were collected only during 2010. Additional information was added to the caption of "conditions of GCMS analysis" in the supplementary material to clarify this, and we have included all samples in the revised Figures 2 and S3 (new Figures 3 and S2).

For all of the line fits in Figure 2 and referenced Figure S3, you should indicate the goodness of fit. You only label two trees that have a 'very different' ratio of Limonene/ α- pinene so this makes it somewhat difficult to accept the conclusions.

We have included in Figure 2 (new Figure 3) all the trees measured multiple times over the course of one season. These data clearly show that the trees do not switch expression of limonene between Mode I and Mode II over the course of a season. We've listed the R^2 for each fit in this Figure and Figure S3 (new Figure S2).

I strongly suggest that the authors adopt the term 'OH reactivity'(e.g. Sinha et. al, 2010) instead 'loss of hydroxyl radical. First, the terms presented are 'loss rates', and not absolute losses. Second, since OH is produced quickly during the day, and is also quickly reacted away, it may be more accurate to use the term 'OH reactivity' and not 'OH loss'.

As suggested, we have changed all references of 'loss of hydroxyl radical' to 'OH reactivity'.

What is the basis for scaling BVOC emissions with needle concentration and temperature based on Raoult's law? It would help to provide some background as to the validity, and the potential drawbacks, of application of this method.

Raoult's Law was a starting point to think about the relationship of partial pressure, concentration, and temperature. Emission rates of terpenes from needles are exponentially

dependent on temperature, like the vapor pressures of pure liquids. This is at least an implication that the behavior can be modeled on principles that are derived for ideal solutions. We expect deviations from ideal behavior of BVOC in needles, since we are dealing with more than just intermolecular forces of pure substances. Terpenes are stored together in vacuoles, and assuming that the resistance to movement through the leaf tissue is similar for a given class (e.g. MT) partial pressure outside of the vacuole will mirror partial pressure outside the needle.

Summary Describe what the different behavior between MT and SQT was, and what you mean by 'annual change'.

This subject was discussed in the manuscript on P53L16-P53L21 and the data are in Table 1. We have included the following in the summary:

"In 2009, total MT showed 32% larger concentration and a wider range of standard deviations than in 2008 and 2010. In contrast, total SQT species were 65% lower in concentration in 2009 than in 2008 and 2010."

References to be added to manuscript:
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Gu, L. et al.: Micrometeorology, biophysical exchanges and NEE decomposition in a two-story boreal forest – development and test of an integrated model, Agricultural and Forest meteorology, 94, 125-148, 1999
Hao, L. Q., et al.: Mass yields of secondary organic aerosols from the oxida- tion of α-pinene and real plant emissions, Atmos. Chem. Phys., 11, 1367-1378, doi:10.5194/acp-11-1367-2011, 2011
Jimenez, J. L. et al.: Evolution of Organic Aerosols in the Atmosphere, Science, 326, 1525-1529, doi: 10.1126/science.1180353, 2009
Keszei, A., Hassan, Y., and Foley, W. F.: A Biochemical interpretation of terpene chemotypes in Melaleuca alternifolia, J. Chem. Ecol., 36, 652-661, doi: 10.1007/s10886-010-9798-y, 2010
Martin, D. M., Gershenzon, J., and Bohlmann, J.: Induction of volatile terpene biosynthesis and diurnal emission by methyl jasmonate in foliage of Norway spruce, Plant Physiol., 132, 1586-1599, 2003
Peñuelas, J., and Llusia, J.: The complexity of factors driving volatile organic compound emissions by plants, Biologia Plantarum, 44, 481-487, 2001.
Shelton, D., et al.: Genetic control of monoterpene composition in the essential oil of Melaleuca alternifolia (Cheel), Theor Appl Genet, 105, 377-383, doi:10.1007/s00122-002-0948-7, 2002
Tamir, H., et.: Intraspecific variation of Chiliadenus iphionoides essential oil in Israel, Chemistry & Biodiversity., 8, 1065-1082, 2011