

Dear reviewer,

We highly appreciate your precious comment on our manuscript and your time. We found the comment can substantially improve the quality of our manuscript and make it possible for an appropriate scientific presentation. We believe that your comments, particularly on the MS data quality control, operation conditions, interpretation of the result, and the possible limitation of this study, were substantially valuable. We have prepared a point-to-point response to the comment. We hope our revision can satisfy both the reviewer and the *ACP* journal.

Reviewer 2

Review of "Evolution of Nucleophilic High-molecular-weight Organic Compounds in Ambient Aerosols" by He et al.

General comments: In this manuscript, the authors present a detailed characterization of the molecular composition of aerosol samples collected throughout the day in China. They use FT-ICR MS analysis to identify molecular formula for each sample and make intercomparisons between the samples. Overall, there is a good deal of data being analyzed and the samples appear interesting. However, there are substantial gaps in the explanation for the methods as well as some of the interpretations that must be addressed before this work is published. I consider these major revisions and these are listed in the specific comments below.

Specific comments:

More detail needs to be included on how the peak identification was carried out. In addition, a peak list that includes the measured m/z , the proposed molecular formula and the corresponding exact mass, and the mass error should be included in supplemental information for each sample. Other items that need to be explained include:

How were isotopes handled? Are they included in the peak identification or removed

initially? If they were removed, what criteria were used?

Response:

In this study, ^{13}C , ^{18}O and ^{15}N isotopic molecules were assigned and analyzed when molecular formula assignment was performed.

Why was sodium not included as a possible component of the molecular formula? Sodium comes from the samples as well as the glassware that is used and it is a very common adduct to see in positive ion mode ESI. If you did not include sodium in your peak identification list I strongly encourage you to go back and re-identify the peaks with it included. It is possible that many of the higher number N containing ions are actually just ions with sodium as the charge carrier.

Response:

The $[\text{M}+\text{Na}]^+$ ion is present in the original mass spectra. The Na adducts ions were also considered in the molecular formula assignment. Considering that the number and relative abundance of $[\text{M}+\text{Na}]^+$ ions are lower than those of $[\text{M}+\text{H}]^+$ ions (Figure 3), only $[\text{M}+\text{H}]^+$ was counted and analyzed.

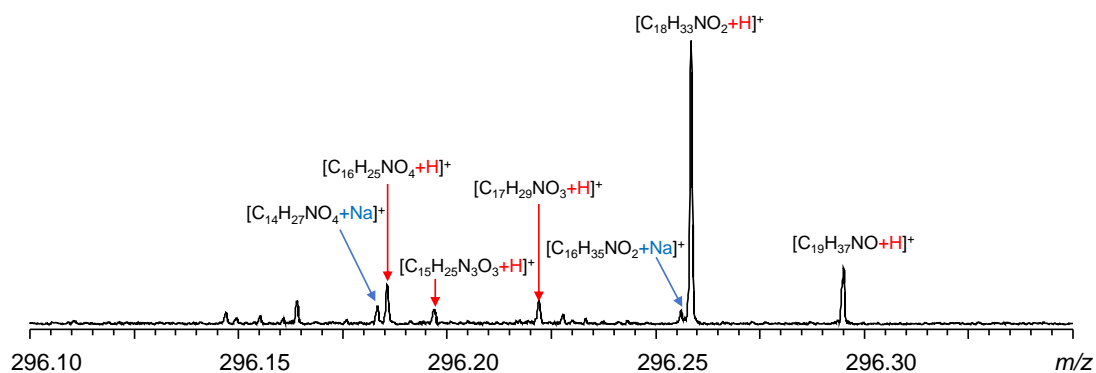


Figure 3 Mass scale-expanded segments at m/z 296 of the (+) ESI FT-ICR mass spectra of sample MEM.

Why was the max O/C set at 1.5? This limits the peak identification of things like sugars and levoglucosan which can come from biomass burning. It is fine if these were not observed, but that limit should not be set at the beginning.

Response:

We set the maximum value of O/C at three before proceeding to molecular formula assignment. Only very few molecules with $O/C > 1.5$ were detected in samples #10 and #11 (please see the single Excel table), and the relative abundances of these molecules are so low that they do not affect the overall conclusions of this paper.

What percentage of the total peaks were identified and what percentage of the total signal?

Response:

We are sorry that we did not understand the question. The total peaks took up to, of course, 100%.

How were blanks collected and handled in this analysis? How were blank ions removed from the list of identified peaks?

Response:

There are two types of blanks that we handled in the analysis.

The first is the blank from a blank filter for PM_{2.5} sampling. The filter belongs to the same batch of filters for PM_{2.5} collection. This blank can indicate the possible organic residues on the filter. The second is the instrument blank. After the ion source of FT-ICR MS was sufficiently cleaned, the instrumental blank was acquainted with the sample of pure LC-MS methanol.

During the data analysis, we treated the blank with caution by not directly removing the peaks from the blank. Firstly, the sample is from airborne material, and some substances can co-exist in both the blank and samples.

Subtraction between samples and the blank using relative intensity was inappropriate neither due to the competitive nature of ionization between various components in ESI mode. The peaks from the blank might be higher than those in samples, leading to negative values after the subtraction. Therefore, we compared the peaks manually to

determine whether the substances in the samples were originally from the blank.

The paper terms these nucleophilic HMWOC but no justification is provided for the term nucleophilic. Why is this terminology being used here? Organic compounds in atmospheric aerosols are often multi-functional, so care should be used in naming because this study is only reporting molecular formula.

Response:

We agree with the reviewer on this issue. Yes, when we use the term "nucleophilic", we think the positive ESI mode can be more sensitive to the compounds that are nucleophilic, such as ketones, carbonyls, and reduced nitrogen-containing compounds. Indeed, those compounds were often multi-functional. The nucleophilic term has to be used with caution.

Carbon oxidation state is a method used to intercompare organic molecules with carbon, hydrogen, and oxygen atoms in the molecular formula. If you are calculating this for chemicals that contain nitrogen, you need to include estimates of the oxidation state of nitrogen. The analysis for the CHO compounds is fine, but the later analysis for the CHON compounds (Figure 6) needs to be corrected.

Response:

When calculating OSc, the two elements, Nitrogen and Oxygen, were considered. In positive ESI mode, the valence of N was considered as -3. Also, S element was not considered because no S-containing molecules were detected in the sample in positive ESI mode.

The OSc calculated in Figure 6 followed the principle mentioned above.

Figures S1-S3 show some summary information from the study but it is not clear if the data from S2 and S3 are for the shorter window used for FT-ICR analysis or for the whole range shown in Figure S1. Please clarify.

Response:

We are sorry for the confusion. Figures 1-3 were presented for the representative of the data. All three Figures showed the whole duration of the sampling period. We have changed the caption of the figures to clarify.

On page 7-8 the concentration of organic carbon is given with estimates of the secondary organic carbon. How was the secondary organic carbon estimated (what method was used)? Why is the certainty in the concentrations higher than the certainty for the organic carbon (more units past the decimal)? What do the percentages signify that are listed after each value?

Response:

The method we used to estimate the secondary carbon can be found in one of our previous work (Wang et al., 2018). Briefly,

The EC-tracer method has been widely used to estimate SOC (Turpin and Lim, 2001), which can be defined as

$$\text{POC} = (\text{OC}/\text{EC})_{\text{prim}} \times \text{EC} \quad (1)$$

$$\text{SOC} = \text{OC} - \text{POC} \quad (2)$$

where POC, SOC, and OC stand for the estimated primary OC, secondary OC, and measured total OC, respectively.

We have put this part into the SI (lines 60-65).

And the digits were also modified. The percentage of SOC can be used as an indicator to evaluate the overall impact of SOC on the whole OC group.

On page 8 the weighted elemental ratios are discussed. How were these calculated? What is the weighting based on?

Response:

The weighting is based on relative intensity. We have changed the part to "relative

intensity weight." in line 159 and 161.

On page 11 it is noted that "In the (+) ESI mode, the CHO group is favorably detected as carbonyl and ester compounds". I would encourage caution with assigning functional groups based on the ESI mode. It is true that these groups can be seen in (+) mode ESI, but so can any group that can solvate the positive charge. Thus, you should also be able to see things like carboxylic acids and alcohols in (+) mode ESI.

Response:

We appreciate this comment. Indeed, esters and ketones can be detected in +ESI mode. Our target compounds, however, commonly contained multiple oxygen atoms that is likely in -COOH or -OH functional groups. Indeed, it is inappropriate to name those complex compounds as ester for ketone. What we can confirm is that there is at least one or even more functional groups like ester or ketones.

For a full description, we changed the sentence to lines 185-187:

"In the (+) ESI mode, the CHO groups are favorably detected as carbonyl or ketones with - CHO functional groups, but we can not simply assign those compounds as carbonyl and ester compounds because it was highly likely to contain -COOH or -OH in one molecule. "

On page 11 a comparison is made between the locations in the O/C and H/C Van Krevelen space the ions are observed and the locations for hydrocarbons, lipids, lignin, etc. It is stated that "Saturated hydrocarbons, ... and carbohydrates were detected in CHO". I recommend more careful language here as you have not positively identified these chemicals as belonging to these classifications. What you have done is measure molecular formula that have the same O/C and H/C ranges as chemicals that are found in these groups.

Response:

We totally agree to this comment. We certainly can not simply assign those compounds as "Saturated hydrocarbons, ... and carbohydrates." The classification in VK diagram

is widely used in previous studies. The classification is more used as references to tell audiences what kind of molecules and their environmental impacts. The criteria of the classification is also used in the work of Bianco et al., 2018.

We have reworded the sentence to lines 192-195:

"We used Van Krevelen diagrams and the criteria from Antony et al. 2014. The classification of Saturated hydrocarbons, unsaturated hydrocarbons, lipids, lignin, and polyphenols is only used as a reference for a better understanding of the distribution of organic molecules and the corresponding environmental impacts."

References

Bianco, A., Deguillaume, L., Vaitilingom, M., Nicol, E., Baray, J.L., Chaumerliac, N., Bridoux, M., 2018. Molecular Characterization of Cloud Water Samples Collected at the Puy de Dome (France) by Fourier Transform Ion Cyclotron Resonance Mass Spectrometry. *Environ. Sci. Technol.* 52(18), 10275-10285.

Throughout this text comparisons are made between the numbers of different chemical groups that are observed in different samples. However, FT-ICR MS with direct ESI is a non-quantitative technique with large matrix effects. Please include a statement somewhere in the text that acknowledges this and clarifies the level of uncertainty that can be expected.

Response:

We totally agree. We have added the statement in the conclusion as a discussion of the limitations of the study (Section 4):

"This work is an attempt to evaluate the evolution of high molecular weight organics using FT-ICR MS. Since the technique is a non-quantitative, and the matrix effect cannot be ignored. Those facts lead to considerable uncertainties."

On page 13 it is noted that "Figure S4 implies that the highest carbon oxidation state (OSc) occurred in compounds with a number of carbon atoms around 20". This is not what I observe in the figure, please modify this text.

Response:

We double-checked the Figure, and we have changed the "20" to "15". The changes were also made in the revision (line 211).

On page 14: "CHO with 20 carbon atoms was observed with more oxygen in the Morning and Afternoon samples than that in MEM and Night". I do not observe this in Figure 3. If this difference is present, I suggest including numbers or a table in the supplemental because it is not visibly obvious in the figure.

Response:

We have added a table in an Excel file containing those data in supplementary, please see the attachment.

Page 14: "Then, at Night, the highly oxygenated compounds (O number >6, DBE > 5, and carbon atoms >50)..." Why are these called highly oxygenated? That is terminology that is used in atmospheric chemistry for compounds with significantly higher O/C values than what is observed here.

Response:

Yes, at this part, the description might be misleading. In the original version, we want to say the organic compound containing more oxygen atoms was found.

We have changed the sentences to lines 220-221:

"Then, at Night, organic compounds containing more oxygen atoms were found (O number >6, DBE>5 and carbon atoms >50) appeared in the nighttime."

For Figure 4, it is difficult to read and interpret this and the other KMD figures. This will be especially true for readers who are red/green colorblind. I recommend changing how this data is presented so that differences between the samples are more observable.

For the KMD analysis comparing CO and COO it is stated that these correspond to the formation of carbonyl and carboxylic acids. However, the formation of carbonyls and carboxylic acids through oxidation reactions does not involve the simple addition of

CO or COO, there are losses of other atoms. It is fine to use these as KMD basis, but the chemical reactions that are underlying these trends are not correctly discussed in this manuscript.

Response:

Thanks for the heads up. The color scheme that we used there are friendly to the readers who are red/green colorblind because we have them checked as the request of the ACP journal. We totally agree with the reviewer about the chemical reactions when forming -CO or COO groups. The following statement has been added to the text when discussing the limitations of our work (Section 4):

"When discussing the evolution of HMWOCs, it is noticeable that the formation of higher oxidative groups, such as carbonyl or carboxylic acid groups, commonly were accompanied by loss of other atoms such as hydrogen or even breaking up of the carbon chain. However, those processes are difficult to be illustrated using the FT-ICR MS method."

On page 15: "Those values were recommended for biomass-burning OA in (+) ESI mode (Song et al., 2022)." What does this statement mean and how does it relate to the results listed immediately above?

Response:

Sorry for the confusion. We have wanted to stated that those groups were most likely from biomass-burning emissions. And in previous studies, HMWOCs with those parameters were attributed as being from biomass burning.

We have reworded the sentence to lines 247-248:

"The HMWOCs in the range of above parameters were likely from biomass burning, as reported in a previous study by Song et al., 2022."

Please be careful with assigning functional groups to the molecular formula identified throughout the paper. For example, on page 16 it is stated that the CHONs were unsaturated organonitrates. While that may be true, I don't see any fragmentation

analysis that demonstrates these groups are present. Thus, the text needs to be modified to be more careful of the assignments.

Response:

We used the criteria from Tang et al. (2020) and Song et al. (2022) . It might be arbitrary to say that CHONs with H:C values between 1.7 and 2.5 and AI< 0.15 were unsaturated organonitrates. A proper attribution as nitrogen-containing organic compounds would be more appropriate because we did not exclude molecules with only one or two oxygen atoms. Also, the H:C and AI values can be influenced by C=C, C=O, and ONO₂, NO₂ groups. Therefore, we have changed the statement as (lines 250-253):

These CHONs were nitrogen-containing, and most were with –NO₂ or –ONO₂, namely unsaturated organonitrate. The details of CHON compounds with higher O/N ratios (≥ 3) are discussed in the following text.

On page 16 a series of changes are listed but these are not clearly visible in Figure 5. I suggest adding a table or some additional figure (possibly in the supplemental) to demonstrate that these statements are true.

Response:

The average molecular weight decreased during the daytime, from 418 Da in MEM to 402 in Morning, and 399 in Afternoon, then increased to 405 at Night. O/C_w varied from 0.13 to 0.14, while H/C_w was between 1.74 to 1.82. O/C_w was lower than 0.18, and H/C_w was higher than 1.5 (Table 1).

On page 18 it is stated that "The nighttime chemistry strongly removed the CHON₁ CH₂-homologues with m.w. >400." I do not see this in the figure.

Response:

Sorry for your confusion. Our statement is (lines 282-283):

The nighttime chemistry strongly removed the CHON₁ CH₂-homologues with m.w. >600 (Figure 7c).

A citation of Figure 7c was added, which might be helpful.

On page 19: "the carboxylic acid functional group can be removed due to the reaction with acids such as HNO₃ and H₂SO₄, or be neutralized by NH₃ in the particle phase."

I am very confused by this statement. What chemical reactions are occurring between carboxylic acids and nitric or sulfuric acid? Do you have references for this chemistry? How would neutralization of the acid impact the measurements in this case? The only impact I can see is a change in solubility (possibly) but the carboxylic acid should still be present.

Response:

We are sorry for the ambiguous interpretation of the result.

1. Reactions between nitric acid and carboxylic acid can form organonitrate. For example, Lim et al. (2016) reported organonitrate formation from the reactions between glyoxal and nitric acid in a chamber study. The reaction occurred on wet aerosols via HNO₃ uptake. In this work, we found a similar process can occur on large organic molecules in the aerosol phase.
2. Likewise, carboxylic acid can also react with sulfuric acid to form Organosulfate in the aerosol phase (Darer et al., 2011).
3. Carboxylic acid can react with ammonia to form ammonium salt or reduced N-containing particles like pyridine (Zarzana et al., 2012). This process might not impact the measurement but on the environment because the products commonly can absorb light to alter the optical properties of aerosols.

We have modified the following statement to clarify our purpose on this part (lines 291-295):

"...and the carboxylic functional group can be due to the reaction with acids such as HNO₃ and H₂SO₄ to form organonitrate and organosulfates or be neutralized by NH₃ to form ammonium salt or reduced N-containing compounds in the particle phase (Lim et

al., 2016; Darer et al., 2011; Zarzana et al., 2012). In previous studies, those processes were proven to occur in small carboxylic acids. In this work, we can observe a similar process on HMWOCs"

Page 22 it is noted that the CHON compounds with N greater than 8 occurred in both day and night samples and the relative intensities for CHN compounds between 20 and 50 were enhanced. Where is this shown and how is it consistent with the KMD analysis shown in Figure 9?

Response:

I suppose the reviewer meant CHN compounds. The N number and relative intensity of high N-containing compounds were enhanced at night, as we can see from Figure 8. In Figure 9, in different KMD plots, we also overserved the enhancement of reduced nitrogen-containing compounds. Hence, the results of both Figures 8 and 9 were consistent.

On page 23 it is noted that you expect acid/base reactions between ammonia and carbonyls. I believe you mean carboxylic acids here as that was what was probed in Dinar et al., 2008. If you do mean carbonyls, please provide more information because this statement is unclear.

Response:

Sorry for the typo. We meat "carboxylic acids," and the statement has been changed (line 354).

For the atmospehric implications section, but also througout the manuscript. Previous work has been done looking at ESI (+) of organic aerosols with FT-ICR or Orbitrap analysis. How do the results presented here compare to the trends observed in those prior works?

Response:

We are grateful for this comment that can clarify the meaning of our work. First, in atmospheric chemistry studies, the ESI (+) is uncommonly used because most studies

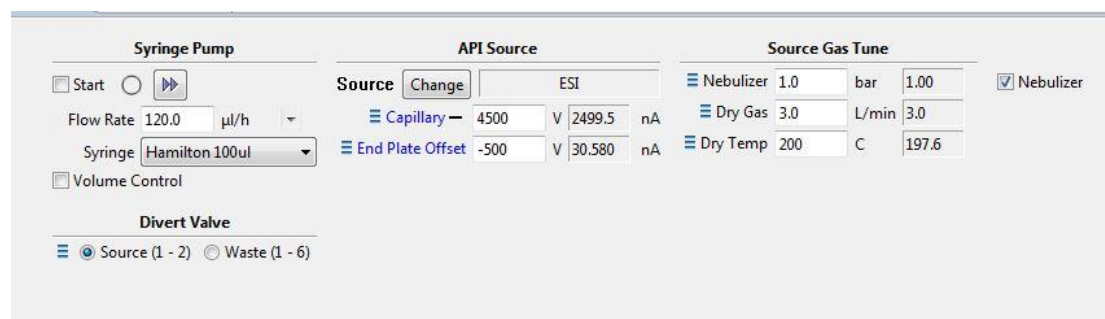
were interested in water-soluble brown carbon aerosols, which were commonly used ESI (-) in FT-ICR MS analysis (Li et al., 2022; Han et al., 2022; Qi et al., 2021; Han et al., 2021). Secondly, in most FT-ICR or Orbitrap studies on related subjects, typically daily samples of PM_{2.5} between different days, the trends between samples were hardly considered, and most of the work focused on a comparison of samples in a daily bias. Thirdly, to our knowledge, we might be advanced in the studies of HMWOCs in PM_{2.5} in a typical daily cycle. All of this reason made our comparison with previous studies difficult.

Minor comments

The capillary voltage on page 6 is negative but this is positive ion mode ESI.

Response:

In the positive ESI mode, capillary voltage is negative. Please see the snapshot below.



The color scale on Figure 3 is repeated with very small text in the Night. Sample

Response:

We have made changes.

In figure 5, what is the color bar?

Response:

The color bar is AI values, we have added it in the revision.

On page 23 it is stated that ammonia and biogenic SOA can form light-absorbing organonitrates. The chemicals that are produced through this chemistry are not generally organonitrates, they are organonitrogen compounds. Nitrates are a specific functional group and should not be confused with the nitrogen containing compounds found in things like pyridine.

Response:

We have changed the statement to "light-absorbing nitrogen-containing organics."(line 339)

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