REPLY TO REFEREE 1

We thank the Referee for the useful comments. We agree that the current version of the manuscripts perhaps lacks a thorough recognition of previous studies on atmospheric aerosol health effects. We answer to his/her specific comments (reported here in *italics*) below:

"This paper involves an analysis of existing data bases to infer possible toxic components of ambient SOA. The analysis reported is not comprehensive, serving mainly as a proof of concept. The idea is interesting, worthy of publication, but would be more appropriate for this journal if linked to existing knowledge on aerosol chemistry and health effects. My main suggestion with this paper is that a more substantial discussion should be added to put the work in context with known aerosol toxicity, source apportionment, and epidemiology studies. There is a substantial body of published studies that have identified various aerosol sources that are strongly linked to adverse health outcomes, such as incomplete combustion; eg, vehicle tail pipe emissions and biomass burning. These produce SOA and many of these compounds seem to be found in the main factors of this study with high toxicity. In contrast, from other studies biogenic SOA tends to be less clearly associated with health effects (check out the published literature). So why not link Fig 2 in at least a qualitative way with published health studies through a more comprehensive discussion, possibly identifying factors or toxic compounds in this study linked to SOA from incomplete combustion, biogenic VOC SOA etc. Putting these results in a large context of published work would significantly increase the impact of these findings, at least for the readers of this journal."

REPLY: The Reviewer is right: there is massive amount of literature results for anthropogenic and natural aerosol health effects that is simply not referenced in our manuscript. We have addressed this, in part, by adding a range of references to cover studies highlighting distinct aerosol source contributions. On the other hand, we would like to note that much fewer studies have focused on SOA than on combustion aerosols, and mutagenicity is only one in the wide range of the possible endpoints explored in the field of aerosol toxicology. Moreover, a substantial fraction of recent studies focused on properties, such as ROS content or ROS expression, which cannot be considered strictly toxicological endpoints but more specifically refer to the mode of action of toxic agents. Even fewer studies performed source apportionment of the observed toxicity burden of the aerosol. To the best of our knowledge, the only study attempting source apportionment of ambient aerosol mutagenicity is the paper by Hannigan et al. (2005) which is based on a chemical mass balance (CMB) approach employing source profiles uniquely for primary organic aerosol, therefore providing no information on the potential contributions from SOA. The main body of the existing literature on role of SOA on aerosol mutagenic activity refers mostly to nitro-PAHs (and to a lesser extent, to oxo- and hydroxyl-PAHs) (e.g., Enya et al., 1997). References to compounds other than polyaromatics is quite sparse. To go back to the original point raised by the Reviewer, we thus provide below a list a papers dealing with mutagenic effects of SOA but not specifically or uniquely from polyaromatic compounds. We also included some of the recent studies of SOA toxicity targeting ROS expression, because this can be linked to the mechanistic profiles of some classes of oxygenated compounds predicted to be mutagenic by our QSAR approach. The overall picture certainly goes in the direction mentioned by the Reviewer: there is a more consistent evidence of the health effects for the products of anthropogenic combustion emissions than for other sources of organic aerosols (Delfino et al., 2009), although biogenic SOA systems have certainly been subjected to lesser extent. We will summarize these literature findings in a new dedicated paragraph to append to Section 3.3 and, in compact, form in the Conclusions.

Selected literature citations:

Citation	Selected results:	Implications for the present study
Kamens et al., Environ. Sci. Technol., 18, 523, 1984.	- Mutagenicity in aged wood smoke increased of a factor of 2 to 10 with respect to fresh smoke.	- Experimental evidence of the mutagenic effects of SOA from combustion sources.
Alves et al., Environmental and Molecular Mutagenesis, 57, 41-50, 2016.	- Episodes of high mutagenicity in the Sao Paulo State (Brazil) seem to occur when the air mass from the rural area of sugar cane production is mixed with air in the region impacted by anthropogenic activities.	- Secondary organic reactions concomitant to both anthropogenic and biogenic SOA formation affect observed mutagenicity of ambient PM in rural areas.
Hannigan et al., Environ Health Perspect., 104, 428–436, 1996.	- Report stresses the importance of proximity to sources of direct emissions of bacterial mutagens but also implies that if 'important mutagen-forming atmospheric reactions occur, they likely occur in the winter and spring seasons as well as the photochemically more active summer and early fall periods.'	- Seasonal variation in mutagenicity, according to different atmospheric reactive conditions
Filep et al., Aerosol and Air Quality Research, 15: 2325–2331, 2015.	- Eco toxicity parameters (cyto and geno) are strongly emission source dependent; the higher the ratio of the biomass burning related carbonaceous aerosol the higher the cytotoxicity and the higher the ratio of the fossil fuel related carbonaceous aerosol the higher the genotoxicity.	- These results, obtained on biomass burning samples, indicate that specific mutagenic effects could change within one PM type – supports potential for composition dependency.
Kroflič et al., Scientific Reports **5**, Article number: 8859, 2015.	- assessment of the impact of low- and semi-volatile aromatic pollutants on the environment due to their atmospheric aqueous phase aging. It reveals that remote biotopes might be the most damaged by wet urban guaiacol-containing biomass burning aerosols. It is shown that only after the primary pollutant guaiacol has been consumed, its	- The study suggests pyrolysis of the polymer lignin during biomass burning leads to semi-volatile organic compounds (SVOC), such as guaiacol (GUA), which then produce nitro-aromatic derivatives.

	probably most toxic nitroaromatic product is largely formed.	
Palacio et al., Mutation Research, 812, 1–11, 2016; Gutiérrez-Castillo et al., Environmental and Molecular Mutagenesis, 47, 199-211, 2006.	- Both organic and water-soluble associated compounds with particulate matter can produce genotoxic effects at concentrations commonly found in urban areas around the world	- Polar organic compounds accounting for SOA composition can contribute to the mutagenic activity of ambient PM.
Barale et al., Environ. Health Perspect., 102 (Suppl. 4), 67–73, 1994.	- Chemical fractionation of aerosol extracts showed that mutagenicity was contributed mostly by polar compounds not PAHs.	- Polar organic compounds accounting for SOA composition can contribute to the mutagenic activity of ambient PM.
Fu et al., Journal of Environmental Science and Health, Part C, 30, 1–41, 2012.	- Photo-oxidation products of polyaromatic comounds lead to the formation of epoxides, endoperoxides and quinones which eventually lead to ROS and DNA damage.	- Evidence for the mutagenic effect of aromatic oxygenated compounds including epoxides, endoperoxides and quinones as in several clusters of compounds predicted to be mutagenic in our paper.
Risom et al., Mutation Research, 592, 119–137, 2005; Oh et al., Mutation Research, 723, 142– 151, 2011. Valavanidis et al., Journal of Environmental Science and Health Part C, 26, 339–362, 2008.	- Oxidative stress caused by PM pollution is genotoxic; - Ambient air PM induces oxidative DNA damage in in vitro systems.	- Experimental evidence of PM-bound ROS-generating organic compounds (not limited to polyaromatic compounds) can be associated to mutagenicity.
Rattanavaraha et al., Atmospheric Environment, 45, 3848-3855, 2011	- ROS expression in aged diesel emissions increased by a factor of 2-4 over fresh diesel particles. - The highest ROS potentials are found when including secondary organic aerosols from an α-pinene, + toluene + an urban HC mixture.	- Several classes of compounds predicted to be mutagenic in our study are characterized by ROS expression among the alerts. Reaction chamber experiments have confirmed the production of redox-active secondary organic compounds in both biogenic and anthropogenic systems.

Tuet et al., Atmos. Chem. Phys., 17, 839–853, 2017.	- Redox-active compounds (measured by the DTT assay) are produced in negligible amounts in isoprene, caryophyllene and pentadecane SOA, in only moderate amounts in α-pinene and monoaromatic SOA, and in much greater amounts in naphthalene SOA.	- Measured ROS expression in SOA qualitatively agree with the predicted mutagenicity of SOA tracers (Figure 2 of our paper), where isoprene, sesquiterpenes, *n*-alkane SOA compounds exhibit no mutagenic effects, while monoterpenes SOA include some mutagenic species and mostly non-mutagenic ones, while low MW PAHs SOA account for the largest share of mutagenic species. These results reinforce the link between ROS expression and mutagenicity.
Verma et al., Environ. Sci. Technol., 49, 4646–4656, 2015.	- Linear regression analysis between ROS generating capacity and OA fractions by AMS-PMF shows that in the SE USA, ROS expression of isoprene SOA is very small while biomass burning OA shows the greatest contribution.	- These results also agree qualitatively with the mutagenic activity of isoprene SOA compounds and of aromatic secondary species presented in our study.

"Finally, it is exposure that determines health effects, that is toxicity times concentration of a given species. A discussion on this would also be very helpful. It would be useful if the authors could provide some idea of typical concentrations of these identified toxic species (or groups), maybe for a range of sites. For example, maybe only a small fraction of the biogenic SOA leads to a toxic substance, but maybe the concentration of these species are very high (or low) making it a potentially important (or not) species."

REPLY: Most of the SOA studies quoted in Table S1 focus on the identification of specific organic compounds on the basis of mass spectrometric analyses. However, the paucity of authentic standards makes quantitation challenging. As a result, the literature provides rather sparse information on the actual abundance of SOA markers in atmospheric samples. Most consistent data refer to well-studied systems such as α-pinene SOA and isoprene SOA. For instance, Kristensen et al. (2014) found that the most common α-pinene SOA tracers, including pinonic, hydroxyl-pinonic and pinic acids (mtr_05, mtr_06 and mtr_07 in Table S1), terpenyllic, terebic and diaterpenyllic acids (mtr_11, mtr_12 and mtr_13), 3-methyl-1,2,3-butanetricarboxylic (MBTCA) (mtr_17), together with their dimers (mtr_15, mtr_16), overall account for 10 to 15% of SOA mass. Similarly, Lin et al (2013) showed that isoprene SOA tracers can represent 12 – 14% of ambient particulate organic matter in an environment characterized by strong isoprene emissions. The contributions of the diverse isoprene SOA species varied a lot, with much greater shares from methyl-tetrols (iso_01 in Table S1), C5-alkene triols (iso_04) and their sulfate esters (iso_02) than from 2-methyl-glyceric acid and its derivatives (iso_06, iso_07). No concentration data are available for the majority of the compounds listed in Table S1, making any attempts of exposure assessment impossible for them. We would like to clarify, however, that the toxicity predictions conducted in our study are useful mainly for *hazard identification*, which is only the first step of risk assessment. Clearly, additional information on concentrations and exposure, as well as

compound-specific dose-response functions are required to characterize the health risk associated with the SOA compounds predicted to be mutagenic in this study. On the other hand, hazard identification is necessary to guide the subsequent steps of risk assessment, including the development of adequate analytical techniques to determine the concentrations of specific chemical compounds according to a priority list.

"Minor comment. The format is a bit strange: Why two Introduction sections? Table 1 has no caption."

REPLY: That is an error: Section 2 is in fact "Methods" not again "Introduction". It is also true that Table 1 has only footnotes not a proper caption. The table reports a summary of the observed mutagenic properties of the 13 organic compounds for which experimental data could be found in the literature.

REFERENCES:

Delfino et al., Air Pollution Exposures and Circulating Biomarkers of Effect in a Susceptible Population: Clues to Potential Causal Component Mixtures and Mechanisms, Environ. Health. Perspect., 117, 152-156, 2009.

Enya et al., 3-Nitrobenzanthrone, a Powerful Bacterial Mutagen and Suspected Human Carcinogen Found in Diesel Exhaust and Airborne Particulates, Environ. Sci. Technol., 31 (10), 2772–2776, 1997.

Hannigan et al., Source Contributions to the Mutagenicity of Urban Particulate Air Pollution, J. Air & Waste Manage. Assoc. 55, 399-410, 2005.

Kristensen et al., Dimers in α-pinene secondary organic aerosol: effect of hydroxyl radical, ozone, relative humidity and aerosol acidity, Atmos. Chem. Phys., 14, 4201–4218, 2014.

Lin et al., Investigating the influences of SO_2 and NH_3 levels on isoprene-derived secondary organic aerosol formation using conditional sampling approaches, Atmos. Chem. Phys. 13, 8457–8470, 2013.