

Interactive comment on “Molecular genetics and diversity of primary biogenic aerosol particles in urban, rural, and high-alpine air” by V. Després et al.

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GENERAL REMARKS.

Overall, this paper on the molecular diversity of primary biogenic aerosol particles is an important contribution to the small but growing data base on the nature of biological aerosols. The most original contribution of the work is the estimation of the amount of DNA inhaled daily by an average human in an urban site. What is the potential impact of this DNA, and what is the possibility for recombination of this DNA (in free form or in cells) in aerosols? These are new questions that will make for lively discussion in the future. (I am not asking the authors to address these questions here.)

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This work also addresses the issue of DNA contamination of sampling materials. This is a VERY important question that has not been addressed elsewhere. The authors have presented data indicating that “clean” filters might harbour significant amounts of DNA. However, their arguments could be improved if they demonstrated that this DNA can be amplified. If the background DNA cannot be recognized by primers used in a study of aerosol biological diversity, then does this contamination really introduce a serious error and the need for decontamination procedures?

The authors do not attempt to make any statistical comparisons about diversity among the different sites. There are some anecdotal remarks about observed differences between sites, but otherwise the effort to sample at different locations is not well valorized. Could the authors improve on this part of the work with existing data (i.e. other data that might not have been presented here)? Otherwise, it is not clear why such an effort was made to sample at different locations.

The authors present data about the taxonomic identity of the DNA sequences detected here. I understand that it would be unusual for the authors not to include this data in a table: it serves as a point of reference. However, it is somewhat redundant with existing information: the authors note that the taxonomic groups detected are rather typical of aerobiological studies. Remarks should be added that some of this DNA likely corresponds to non viable organisms as no attempt was made by the authors to use selective techniques to isolate DNA in so-called "viable" cells (for example: Nocker et al 2006, J. Microbiol. Methods 67: 310-320).

SPECIFIC COMMENTS. 1) The title needs a minor change. ‘Molecular genetics’ is the study of the molecular structure and function of genes. However, this paper addresses only the molecular diversity of aerosols. Hence ‘genetics’ should be removed from the title.

2) pg. 352, line 11: This paragraph is about anomalies in characterization of air samples due to contaminants and chemical modifications. Hence, shouldn’t “On the other

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hand” be changed to “Furthermore” as the information that follows is not a contradiction to what preceded but rather an additional illustration?

3) pg. 352, line 23-24. Remove the word “biological”. When is an organism not ‘biological’?

4) On pg. 354 the authors indicate that they did not decontaminate the TSP filters. It should be explained why these filters were not decontaminated whereas the others were.

5) pg. 355, line 1: ‘Aliquot’ is generally used to describe a volume. Here the word is used to describe a section cut from the filter. It would be best to state: “PM2.5 filter pieces”.

6) pg. 356, lines 25-26: The description of how data were normalized is not clear.

7) Concerning statistical analysis and data related to identity of sequences: did all sequences belong to known groups? Were there unidentifiable sequences? This is not clearly presented.

8) pg. 359, Atmosphere aerosol samples: Information is presented about the adequacy of the DNA extraction kit. Were there any measures of efficiency whereby decontaminated filters were seeded with a known amount of DNA followed by extraction? This would be useful.

9) pg. 362. This section on blank and background samples provides information about the presence of DNA on filters before they are used for sampling and that which might accumulate during handling after sampling. Data is given about the quantity of DNA. Is data available about the identity of this DNA? It would be useful to know what organisms it corresponded to. Was there any attempt to amplify this DNA? If it could not be amplified, then it could be argued that it might not necessarily be an important source of error for diversity studies.

10) pg. 363, Sequences and phylogeny: In this section the authors present the identi-

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ties of the sequences detected based on blasting in the NCBI data base and on phylogenetic analyses. It must be understood that all of these identities are the best HYPOTHETICAL identities based on similarities with known organisms. In this light, it is VERY misleading to bring up names of members of some of these large groups that have high “sales value” - such as *Bacillus anthracis*. There is no data to support that such organisms were in samples. Why not suggest the name of any other random *Bacillus* species or other spore-former?

11) pg. 366, lines 10-20: The experimental design does not allow any statistical comparisons to be made among the different sampling locations. Is there any way that the data can be better exploited so as to test hypotheses (via statistical analyses) about differences in locations?

12) pg. 367, lines 3-4: The authors state that “These findings are consistent with the results of bacterial sequence analyses in air particulate matter at urban and rural locations Ě”. They should add: “and with most previous studies using culture-based methods” (and cite the appropriate references).

13) pg. 368, lines 15-23: This result about the absence of fungal DNA is very surprising!!

14) pg. 370: Animal sequences. In this section the authors again make a misleading remark related to the identity of their sequences. The Alveolata: Apicomplexa, otherwise known as the Sporozoa, are common parasites of insects and vertebrates. Hence there is a large list of possible examples that the authors could cite to illustrate this type of organism. I do not think that it is appropriate to single out a human pathogen when there is no specific data. In the long run this could lead to dangerous consequences for this field of research and does not favour comprehension of microbiology by the general public.

TECHNICAL COMMENTS 15) pg. 360, line 7: change “were” to “we” and eliminate the comma after “calculated”. This will read better.

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16) pg. 362, first line: change “have been” to “were”. Line 4: “in the databank OF THE National Center”

17) References: There are many inconsistencies in the format used to list the references, especially concerning the use of capital letters in titles.

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