

“Observations of fluorescent and biological aerosol at a high-altitude site in Central France”/Gabey et al.

The paper centres on a very short campaign to detect bioaerosol (and other) found at high-altitude in France. As such it does provide further interesting data, particularly regards the use of the WIBS real-time approach in a field campaign, but I am not sure if any wide-ranging or definitive conclusions about airborne bacteria, pollen, spores etc can be made from the limited study period employed. (*v.i.* at one point, 3049.25, an important conclusion is given based on just two nights of comparison.) Essentially there must be some more clarification about the way the experiments were performed, why they were performed how the different detection approaches complement each other in particle quantification, sizing/shaping and finally the contribution that the work makes to our understanding of airborne bioaerosol distributions in Europe. Detailed comments/questions are found below.

3033. 16. As discussed in the manuscript at a later point the two excitation wavelengths utilized in WIBS do not uniquely excite tryptophan/NAD(P)H. (NB. some consistency here please on NADH-NAD(P)H use in the paper). Hence I would leave these assignments out here in the abstract...the issue requires the nomenclature discussion given in the main body of text.

3033. 20. Gives two fluorescent populations of interest found: 3 μm and 2-3 μm but no real message as to what species they correspond to as an important conclusion from the results.

3034. 4 The statement here represents the first reference to the important contribution of non-PBA to the WIBS3 data. Hence it does require the listing of exactly what fluorescent, non-PBA are likely here and what discriminatory WIBS signals they give if the suggested species are introduced to the instrument (say back in the laboratory). This would help pin down some of the important interference issues raised in this paper

3034.19 No “that” in the sentence

3038.4. What were the typical (or high-low limits) of the wind speeds measured at pdD over the campaign period? Is the particle entry point tubing vertical to the WIBS or bent to a right angle? In other words can the incoming PBA be blown away by the wind if it is a vertical arrangement because the WIBS does not pump more effectively than the wind speed? What is the length of tubing used between entry point and WIBS? What is the tubing made from? How high above the ground is the particle inlet point? These matters can lead to major discrepancies between reported “measurements” and actual ambient concentrations unless they have been accounted for. Could/did a right angle capture, for example, provide differing counts to a vertical arrangement on a windy day? That is why knowing the wind speeds present at the site over the campaign period is particularly important to the ultimate counting conclusions of this manuscript. If the wind speeds were high(ish) and only a vertical arrangement was used then the data reported is likely suspect.

3039.2 Could there be an expansion of what is actually meant by EFM uncertainties here....particularly recognition errors? Does this statement indicate issues between bacterial types or between a bacterium and a fungal spore for example? What were the actual proportions of the different types of biological particle observed here over the campaign? There must have been some assignments and size/shape information.

3040.17. I think it is a mistake for this paper to only briefly discuss the AF data...shown in Figure 3. It could give a lot of information here about bacteria, other PBA or non-PBA. Bacteria are generally spherical when at about 1 μm diameter....that is close in size and shape to certain fluorescent chemical interferants (although these are often $<0.5 \mu\text{m}$ and spherical). However many bacteria are rod-like at the larger dimensions....maybe 3 μm long and 0.5-1.5 μm wide. (What species are 5 μm in diameter as given on 3042.24?) What clues as to identity are actually given by the EPF results here? (There must be some if only % spherical to % rod-like data). Does the WIBS measure some of these PBA on the axis or off the axis thereby giving two different apparent "sizes" for the same species? Also see next point.

3045.24. Reference is made here to Figure 3 and the small feature at AF11 and the larger one at AF20. First of all surely the AF11 peaks are not small in comparison to AF 20! What sizes of particle do these two AF features refer to? In any case AF11/AF20 values also surely indicate spherical particles. (The range of AF is 0-100 from Figure 3). Thus AF20 is still a low value and indicates spherical....but if you have 3 μm sized **bacteria** then these are more likely rod-like! In summary, I think there should be a more coherent and connected discussion in this paper of AF/size/identity using WIBS plus EPF...it would help the reader. Section 4.1 discussion does not include any quantification of the PBA general types likely here or even make comparison to any quantitative EPF results.....that should themselves be made more explicit (section 2.3). I began to think that bacteria were not important at all here as currently written.....and I should at least be able to understand that by this point in the text. Is it possible that no bacteria were detected by WIBS3? This might explain the discrepancy referred to in Section 4.2 regards "clustering". What evidence is there that airborne bacteria cluster to large sizes in the WIBS3...from actual lab experiments? (See next point).

3047.9. Reference is made to WIBS3 limited sensitivity at small sizes. What does this mean...what do the calibration curves look like for sizes <2 and $>2 \mu\text{m}$? In other words if you cannot detect $<2 \mu\text{m}$ particles effectively then you will not detect many bacteria...even some spores....likely important here....and what does that all mean to the overall analysis given in this paper?

3048.23. The diurnal cycle referred to generally has a relationship to relative humidities. What were the RH as a function of day/time measured here?. Such data is important...as well as temperatures.

3050.3 Is enough known about the relative intensities of fluorescence for any airborne PBA to say that all bacteria can be discriminated from fungal spores etc by signals originating from the two excitations? Again....the two excitation wavelengths used in WIBS do not correspond to tryptophan and NAD(P)H contents entirely....many other absorbers can be present in different ratios for different species.

3050.23 How can the cluster technique come up with a distribution set at 1.5 μm if WIBS3 is not sensitive at $<2 \mu\text{m}$? This point, in a sense, goes to the heart of the issue that must be clarified in this paper for readers. Is this set being assigned as non-PBAP because they are non-fluorescent even though the WIBS3 is not sensitive to detecting such particles? What were the previous laboratory characterizations, using WIBS3 that leads to the conclusion made about Cluster1?

3051.7. Data in table 1 do not indicate high AF for clusters 1 and 3....they are likely spherical at 22 and less...Cluster 2 at 33 is perhaps on the edge of being slightly asymmetric. At a size of 7.6 μm , could cluster 3 be associated with any pollen or spores monitored by the EFM approach?

How did the team calibrate the WIBS3 for AF relationship to shape for the types of PBA proposed here e.g. *P.syrengae* ?

My summary conclusion for this paper is that it will have to make a more convincing case for showing that in this campaign the WIBS3 instrument detected any bacteria at all, and if so what types they were...including the likelihood of them becoming agglomerated.....possibly by parallel laboratory studies on the WIBS3. Once this is established then the important point that this paper makes about non-fluorescent particle contribution to the overall burden in this campaign in France can be better evaluated. Currently I cannot do this but with a clarified manuscript I hope to be able to attempt to do so because it represents a very important topic for on-line analyses of ambient bioaerosol.