Answer to the anonymous Reviewer #1

Comments

The manuscript describes measurements of aerosols in tropical regions, on the island of Puerto Rico. The authors had the interesting idea to shed light on the aerosol composition in San Juan, the capital city of Puerto Rico, and try to identify the role of biological aerosols as CCN. The authors used various measurement techniques (WIBS, CCN counter, CPC, Burkard trap) to characterize the aerosols with temporal resolution over a period of 8 days. The main findings of the paper can be summarized as follows:

1. Most aerosol particles in the region are emitted from human emission sources, e.g. cooking and traffic. However, the authors were able to distinguish those from the bioaerosol fraction which was much smaller in number concentration (using fluorescence and fungal morphology data).
2. The daily trend is different for the measurement methods, e.g., different aerosols categories and sizes follow different trends in time (due to primary and secondary aerosol formation mechanisms).
3. High relative humidity triggers bioaerosol emissions, likely fungal spore production.
4. Most fungal spores present in the area are from the phylum Basidiomycota or Ascomycota. Those contribute to the fraction of fluorescent particles detected with WIBS and follow a trend which peaks in the early morning hours.
5. There is no evidence for a correlation between the CCN number concentration and the number of fluorescent bioaerosol. This suggest that bioaerosols emitted from tropical urban areas do not significantly contribute to cloud condensation in tropical clouds.

Although this manuscript provides interesting data and fills a lack of missing information from aerosols and bioaerosols from tropical cities, I would suggest major revisions (mainly in the introduction and in the discussion) before it is considered to be published:

First, I am wondering about the motivation behind the effort to link CCN concentrations to bioaerosol measurements. It is well known, that bioaerosols are generally low in number concentration around the globe (Hoose et al., 2010) and that most aerosol particles can act as CCN when a certain supersaturation is present. The authors make a statement about giant CCN in the introduction (line 78) but do not explain why bioaerosols from the city could be an important source of CCN. I encourage the authors to extent the introduction with a more detailed statement of bioaerosols as CCN.

Response: Agreed that bioaerosols contribute a small fraction (50 Tg yr^{-1}) of the total natural global emissions (~2900-13000 Tg yr^{-1}) (Hoose et al., 2010; Stocker et al., 2013). However, their mass and number concentrations are site specific and greatly vary depending upon the location and climatic condition (references therein Zhang et al., 2021). In terrestrial ecosystems, bioaerosols constitute a major fraction of total aerosol load. As far as urban and rural atmosphere are concern, bioaerosols of size greater than ~1 µm may account for around 30% (references therein Fröhlich-Nowoisky et al., 2016). There is evidence where bioaerosol may constitute a significant fraction

(5-50%) in the urban air (Jaenicke, 2005). The reference to giant CCN has now been expanded upon to explain that giant CCN play a special role in precipitation development because they can form larger droplets that more easily collide and coalesce to form raindrops. Hence, although small in number concentration they make up for in their size and capacity to contribute to early precipitation development.

Note: Most of the bioaerosols detected in the city of San Juan are originated in El Yunque rain forest and not in the city itself.

Second, the paper provides important data about bioaerosols in cities, measured close to emission sources. Tropical island, such as Puerto Rico are remote areas where a large fraction of aerosols comes from local emissions. Thus, it is very important to gain data about local, urban, aerosol compositions in order to make conclusions about human health, environmental and meteorological influence. Although, the authors make a few statements in the manuscript about the importance of their measurements, the paper would strongly benefit if the statements were more emphasized, and the knowledge gap would be identified more clearly in the introduction.

Response: The reviewer makes an important point and more background information has been added as well as why our measurement site in a location that is fairly representative of a typical Puerto Rican urban. We do disagree somewhat with the reviewer's comment about a large fraction of the aerosols coming from local emissions. Previous papers (Allan et al., 2008; Raga et al., 2016; Torres Delgado, 2021) have shown that because of the persistent flow of air coming from over the ocean, the largest fraction of aerosols are of marine origin.

Puerto Rico is characterized by tropical climate, urban land cover and use, moist soils, unique topography, and dense vegetation. These factors, associated with the easterly trade winds from the East, could influence the concentration of airborne particles, for examples, organic particles, viruses, bacteria, fungi, pollen, etc. (Velázquez-Lozada et al. 2006). Nevertheless, meteorological variables (high humidity and wind speed) are also the important factors, influencing the airborne particle population in the tropics, including rainy seasons. There are various sources of particulate matter degrading the air quality of Puerto Rico, i.e., from industrial activities, anthropogenic inputs, African temperatures dust storms and volcanic eruptions. The urban areas of Puerto Rico are considered developed with industrial growth, most of which is related to pharmaceutical and power generation plant. The power generation plants are responsible for releasing millions of pounds of air pollutant annually (Torro-Heredia et al., 2020). Data show that a large number of organic compounds (e.g., n-alkanes, esters, phthalates, siloxanes, and other) including plasticizer released into the atmosphere which could pose major health threat in this area (Torro-Heredia et al., 2020).

Third, the authors conclude that the influence of bioaerosols as CCN to be from minor importance. I was wondering if the paper would proliferate when highlighting the health effects which bioaerosol can have in more detail. Are those concentrations of fungal spores comparable to other cities? How is the concentration in rural and remote areas compared to this study (e.g. compare with El Yunque rainforest data if that is possible)? In addition to that it is well known that bioaerosols, especially fungal spores, are among the most active ice nucleating particles (see e.g. Kunert et al., 2019). Is it possible that those particles do not influence the budget of CCN drastically, but still have an influence on the INP budget? I would encourage the authors to include the role of bioaerosols as INPs in the discussion and to shortly state health effects. This can help transferring the results of the paper into a puzzle piece from the bigger picture of bioaerosols.

Response: It is evident that almost all of the fungal spores are released in the El Yunque rain forest (Lewis et al., 2019). Two of the air sampling sites, Pico Del Este (PDE) and Cabezas de San Juan (CSJ) are very similar and very low in the fungal spores (less 5,000 spores/m^3). At another sampling site in El Verde (located to the west within El Yunque National Forest), the concentrations increase to 72,000 spores/m^3 and are found to have a decreasing gradient of fungal spores towards the Metro Area. For the rest of Puerto Rico, the Central Mountain Range is the other source of fungal spores. Bioaerosols, especially fungal spores, are very good ice nucleating particles (INP); however, except for periods with tropical storms, cloud tops rarely grow higher than the freezing level. As per the reviewer suggestion, we will add the statement on the health aspects of bioaerosols

In summary, the paper provides an interesting insight to a pilot study of aerosol research in a tropical city. I hope that my feedback helps the authors to revise a manuscript with a clearer storyline. Furthermore, the manuscript would benefit from spelling and grammar checks. In terms of further field campaigns, I would encourage the authors to focus not only on CCN, but also on ice nucleation and/or health effects driven by urban bioaerosols.

Specific comments:

19 ... change to are capable of …

Response: Correction made in the revised manuscript

20 … What’s the difference between plant spores and pollen?

Response: Pollen is essential for sexual reproduction of flowering plants and plants that produce cones. Spores are microscopic propagative bodies, with single nucleus, whose primary function is plant dispersal and reproduction. Spores are produced by “lower” plants which include mosses, liverworts, clubmosses, horsetails and ferns.

27 … change to a population of 2,448,000 people …

Response: Correction made in the revised manuscript

68 … change to where vehicular and industrial emissions …

Response: Correction made in the revised manuscript

69 … The reader might be confused reading about local aerosols and then African dust in the next sentence. Maybe it is important to highlight the long-range transport before explaining African dust as CCN.

Response: A statement on long range transported aerosol is added in the revised manuscript

"Apart from this, clouds and rainwater in this region are influenced by long-range transported natural aerosols."

96 … What meteorological factors? Please elucidate more clearly.

Response: The meteorological factors (relative humidity and wind speed) were clarified in the revised manuscript

101 … This is an important statement why this time was chosen.

Response: Thanks to the reviewer for acknowledging the statement

134 . I was wondering how representative the location of the university is for the city center. Can you make a statement about the areas and the distance to the city center of San Juan in the text?

Response: A statement is added in the revised manuscript as per reviewer's suggestion.

Moreover, the measurement sites are located at the center of the San Juan city (199 km^2), a clear representative of typical urban atmosphere.

136 … How much rainfall, can you state a yearly average in brackets?

Response: The annual rainfall recorded in San Juan, Puerto Rico is 4.22±1.3 in. This value is added in the revised manuscript.

202 … Are Tryptophane and NADPH the only molecules exited by fluorescent light in that region. Are there other molecules that could potentially also show fluorescence in that area. If yes, please explain in more detail.

Response: There could be numerous molecules that can be excited by fluorescent light. For examples molecules such as proteins, large polymers, molecules having conjugated double bonds, heterocyclic aromatic compounds, particularly when nitrogenous substituents are present. Tryptophan is an amino acid that has the highest (~ 90%) fluorescence in the native protein. Nicotinamide Adenine Dinucleotide Phosphate (NAPDH) is one of the major contributors to the fluorescence signal when attached to the protein molecule and is produced widely in the metabolic cell. Other atmospheric relevant fluorophores are Riboflavin, Vitamin A, Vitamin D, Vitamin C, Vitamin B6 compounds, Cellulose, etc., which could be present in the FAPs but their emission and excitation matrix is yet to be conceptualized.

221 … I have seen that the fluorescence signals alone can be misinterpreted in some cases (see e.g. Savage et al., 2017). Why are you sure that those are specific for bioaerosols in your case?

Response: There could be a possible interference of the fluorescence signal but we have set a threshold to minimize fluorescence from non-bioaerosols, i.e. organic compounds that can fluoresce, but at an intensity that is typically much lower than bioaerosols. In addition, we observed a relatively strong correlation between the FAP signal and the fungal spores prevalent at the site. Also, the identified fungal spores have a systematic diel pattern, ubiquitous, and are observed to have a considerable population in the San Juan atmosphere based on this study and previously published literature.

251 … I have seen different forms of writing units in the manuscript (e.g. L min^{-1} or liters of air/min) please make sure that all the units are consistent in the manuscript.

Response: Thanks to the reviewer for notifying the inconsistency. It is corrected in the revised manuscript.

262 … change to … from a weather station that is located around 800 m away from …

Response: It is corrected in the revised manuscript.

264 … I am wondering how 100 m above ground level would correlate with your measurements at ground level? Do you assume the aerosol composition to be the same in those altitudes?

Response: This will depend upon how well mixed the boundary layer is. In this region, the boundary layer depth is 300-500 m and due to the solar heating it becomes well mixed by late morning. Although usually light, the local winds also contribute to maintaining a mixed layer.

Note: Using the Burkard we capture the fungal spores and pollens on the roof of the 10^{th} floor building which is about 60m above the ground level

281 … I was wondering if the number of CCN is always so low compared to CN. Is that also seen in other studies?

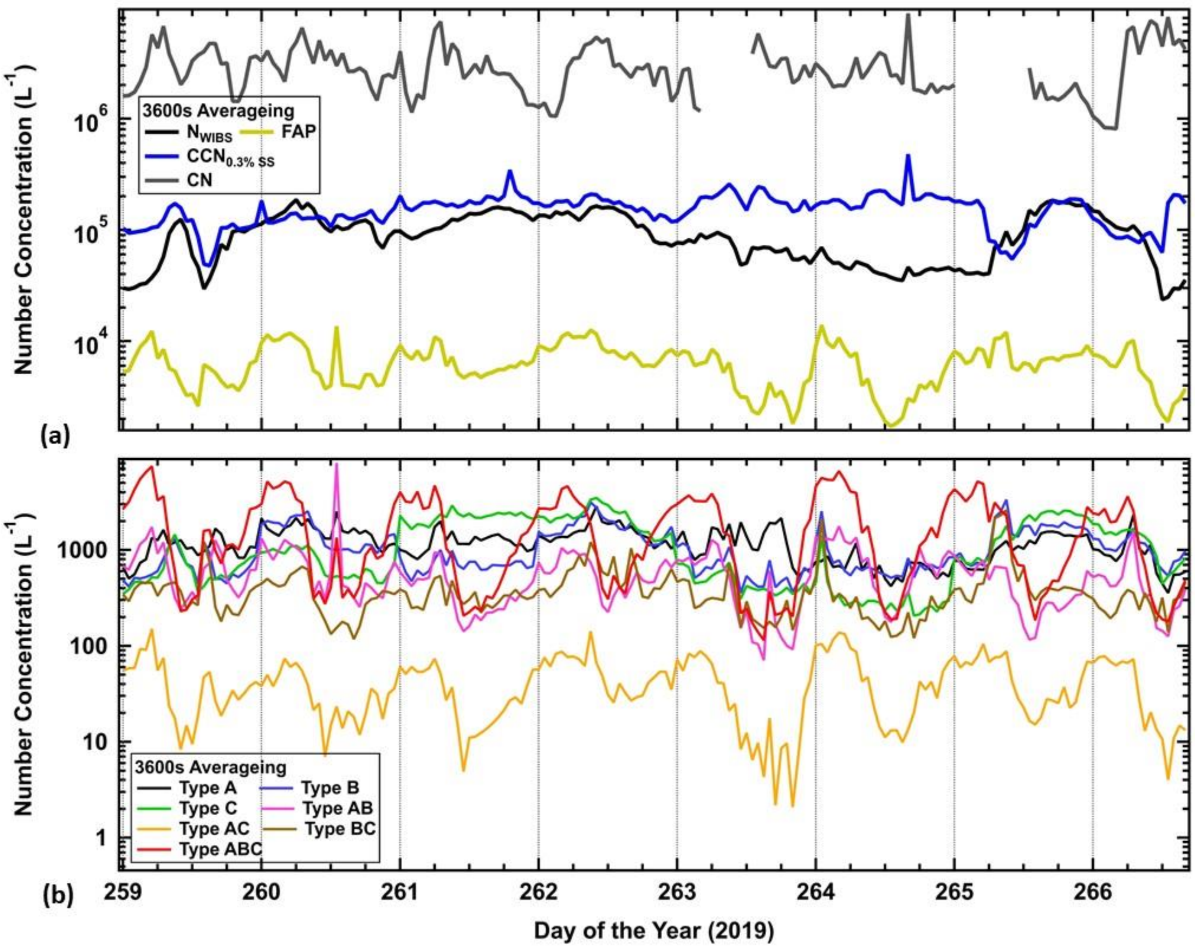
Response: Yes, Allan et al. (2008) reported similarly low numbers of CCN compared to CN in Puerto Rico.

285 … This statement assumes that the WIBS and CPC would count the same number concentration for all aerosols above 500 nm. Can that be seen in your data or confirmed with literature?

Response: Yes, this statement can be confirmed by previously published literature (Healy et al., 2012). However, the counting of the particles by the instruments may differ a little because of the different sampling flow rates used for WIBS (0.3 L min^{-1}) and CPC (1 L min^{-1}) in this study.

Figure 2 … I have a hard time seeing the daily trend for CCN and CN. Would it be better observable if one would include vertical lines whenever a day ends in the diagram as a grid?

Response: As per the suggestion, vertical line as a grid will be added to the figure 2 in the revised manuscript.



318 … very interesting finding

Response: Thanks

Figure 4 … the letters (a) and (b) are poorly visible

Response: The letters (a) and (b) were placed outside of the figure 4 in the revised submission.

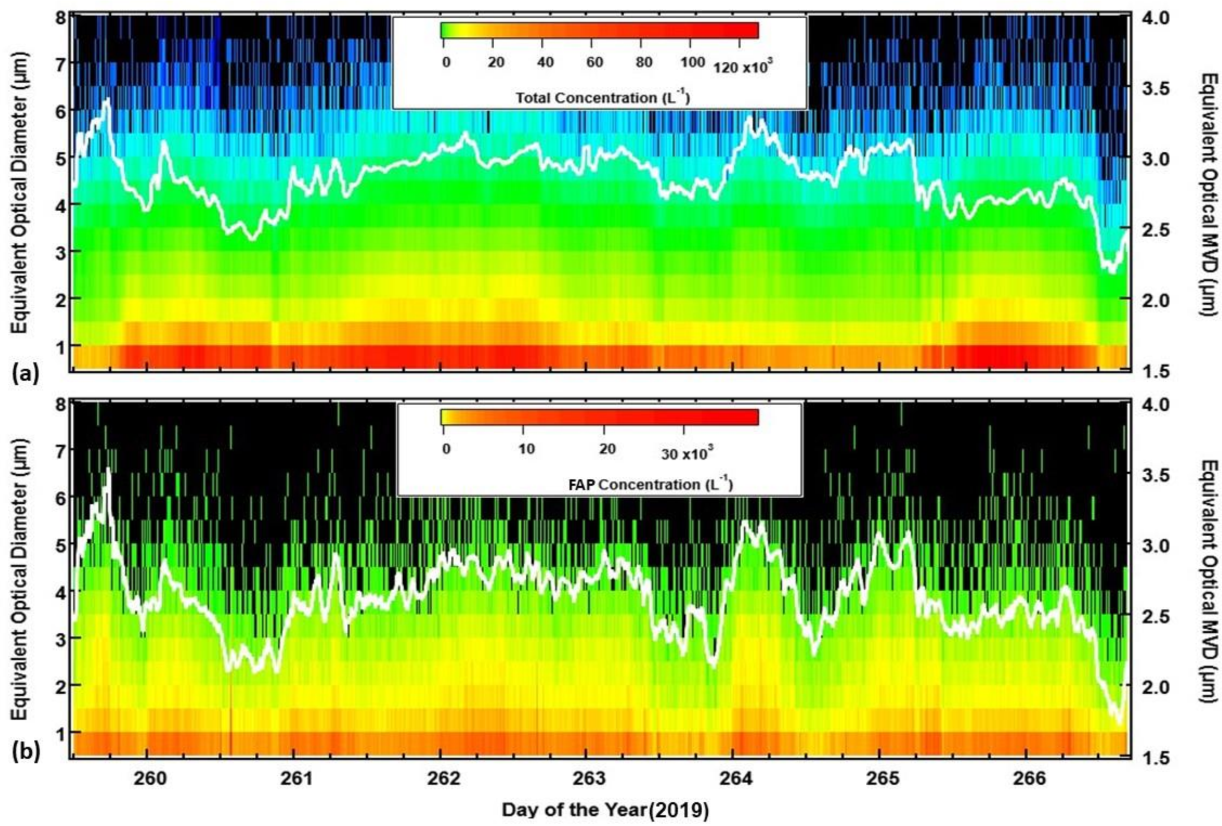
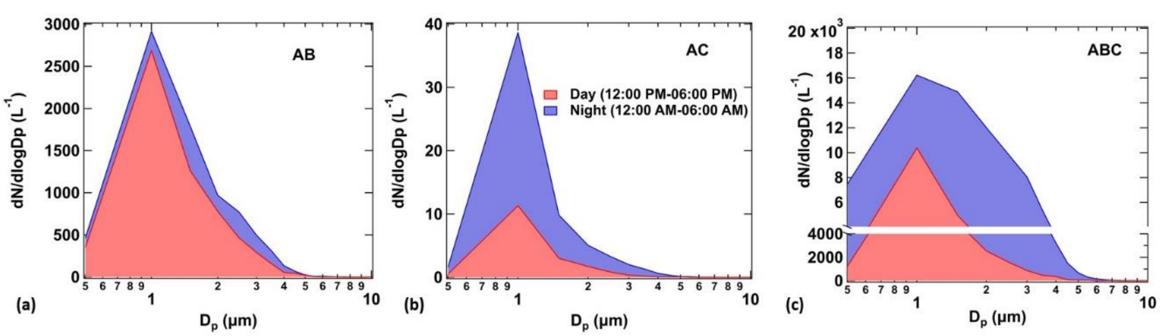


Figure 5 … I am confused by numbers on the y-axis in Figure5(c)

Response: The y-axis in Figure 5 is corrected in the revised manuscript.



409 … The findings about the relative humidity and the fungal spore production are interesting and one of the main findings in this work

Response: Thanks

444 … I have a hard time in understanding this paragraph and the conclusions of it. 1. Why is ABC fluorescence related to Basidio & Ascospores? Are those spores simply bigger than the other

fungal spores and therefore the fluorescence signal extents the threshold for ABC? 2. Why is the concentration of the spores in WIBS so much higher than from the Burkart trap (Figure 7)? 3. Did you also count bacteria, bacteria agglomerates, and pollen with the Burkart trap samples? If no, why not? If yes, how did they influence the measurements of the fungal spores in the WIBS?

Response: (1) Previous studies (Quintero et al., 2010; Rivera-Mariani et al., 2020) reported that the most common fungal genera detected were the Basidiospores and Ascospores in the San Juan atmosphere. Which is also confirmed in this study. The size of the Basidiospores and Ascospores (10-20 um) is usually larger than Aspergillus, Penicillium, and Cladosporium spores. They are release at the early morning hours due to dew point and increase in relative humidity at these hours. Furthermore, we observed a systematic diel pattern in the number concentrations of these fungal spores which is strongly correlated to diel pattern of FAPs detected in ABC channel of WIBS. (2) The difference in number concentrations because both the instruments work on different detection principle and different sample flow rate. Furthermore, the Burkart trap was not located in the same site (18°24'6.4"N, 66°03'6.5"W, 6 m a.m.s.l.) where the WIBS was operational but was installed at a different location (18°23’48” N, 66°4’30” W, 60 m a.m.s.l) in San Juan. (3) Due to their small size the bacteria are not usually counted with the Burkard. Only pollen and fungal spores are enumerated with this air sampler. Pollens are release during late morning and noon. They much lower in concentrations compared to the fungal spores.

473 … I would be careful with this statement since you are assuming that the measurement techniques cover all particles in the area above 500 nm. Also, this would include that in terms of number concentration the study only looks at 2% of the particles closely. This further implies that bioaerosols are a very low fraction of the total aerosols, yet they are important for health and climate. Maybe the authors can state that more clearly and explain that coarse mode aerosol is typically smaller in number concentration, yet high in mass concentration.

Response: The suggestion made by the reviewer has been included in the revised manuscript.

504 … This is a main message of the paper and should be extended with one or two more sentences of what particles matter for CCN.

Response: The additional statement is added in the revised manuscript.

This suggests that if FAP are good CCN, they do not contribute significantly to the overall CCN population. It is important to note that contributions to the overall CCN populations depend on particle size, chemical composition, and number concentrations.

Figure 8 … It is hard to see the trend in the graph with the current color scale. Would it be helpful to consider a logarithmic color scale or other colors?

Response: The figure is modified with different color scale in the revised manuscript.

603 … As far as I know Basidiomycetes and Ascomycetes are not fungal species but more of a phylum.

Response: Reviewer is thanks for notifying this. The term Basidiomycetes and Ascomycetes changes to Basidiospores and Ascospores in the revised manuscript.

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