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Relationship Between Biologically Fluorescent Aerosol and Local Meteorological Conditions

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Time-resolved characterization of biological aerosol is important both for understanding environmental processes that affect biological aerosols and for determining realistic test conditions for the evaluation of bioaerosol detection systems. Very little work has been done to develop an understanding of the temporal fluctuations in bioaerosol concentration. During an experiment from 1–10 November 2008 ambient biological aerosol and meteorological data were collected. A FLIR/ICx/S3I Instantaneous Bioaerosol Analysis and Collection sensor was used to count both the biological and nonbiological aerosol in two size bins. The data indicate that the ambient relative humidity affects the optically observable concentration of biological aerosol with higher relative humidity generally associated with higher biological aerosol concentrations. The short timescale over which these correlations exist implies an aerosol process, rather than a change in aerosol source.

INTRODUCTION

Advances in biological aerosol detection technology have made the collection of high temporal resolution information on ambient biological aerosol possible. Many new systems are capable of rapid measurements (up to 1 Hz) of biological aerosol concentration. Many of these sensors rely on laser-induced fluorescence (LIF) technology to provide information on fluorescence of individual aerosol particles, which is assumed to correlate to their biological origin. Other sensors have used single particle mass spectrometry (Steele et al. 2008) and laser-induced breakdown spectroscopy (LIBS; Lee et al. 2004) of multiple channels of fluorescence information (Sivaprakasam et al. 2004) to improve the characterization of the individual particles. None of these techniques has been proven to differentiate between different types of bacteria, and most of these techniques simply identify the fraction of the aerosol that is of putative biological origin.

Several past studies have surveyed the ambient biological background to provide a better understanding of the diversity, variability, and factors that control the biological aerosol population. Bacterial aerosol have been observed and studied in the ambient environment since the mid nineteenth century. One of the first studies to observe fluctuations in viable bacterial concentration was Lighthart and Shaffer (1995). This study used slit-to-agar samplers operated sequentially at 1-h intervals to determine the viable bacterial concentration with a temporal resolution of 2 min. The authors found diurnal variations in the bacterial concentration that were concluded to be associated with the local meteorology and climatology associated with land–sea breeze in the Willamette River Valley in coastal Oregon. Shaffer and Lighthart (1997) examined fluctuations in viable microbes at urban, rural, forested, and coastal sites in Oregon. The data also exhibited reproducible diurnal trending. Among the bacterial genera they were able to identify, *Bacillus* was the most prevalent and, in general, gram positive organisms accounted for greater than 70% of the collected organisms. Recently, a phylogenetics study of ambient urban biological aerosol diversity was performed in Austin and San Antonio, TX (Brodie et al. 2007). Using 16S rRNA (ribosomal ribonucleic acid)-targeted microarrays, this study was able to map the bacterial diversity in these two locations over 17 weeks. This study also monitored the mean meteorological and air quality conditions during sample collection, although the temporal resolution of this data set is low (1 week). Despite the low temporal resolution of the data, some organisms were found to be more sensitive to meteorological conditions than others, adding another component to the diurnal variations observed by Lighthart and Shaffer. They discussed the presence of specific taxa as evidence of aerosol sources, such as *Arcobacter* and *Heliobacter* as a potential indicator of fecal contamination from wastewater treatment facilities, but do not provide data that indicate these sources may have contributed

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to the sampled aerosol. Other recent research has used spectroscopic techniques, such as ultraviolet light-induced fluorescence (UV-LIF) to characterize individual aerosol particles as biological or nonbiological (Pinnick et al. 1995; Reyes et al. 1999). While this technique is useful for providing high temporal resolution information on the fluctuations in the fluorescent particle population, it is not possible with most instrumentation to discern what fraction of this aerosol represents bacteria, molds, viruses, or other materials that fluoresce at the chosen wavelength (typically 266, 365, or 405 nm). Using high-resolution spectrographs of fluorescence of a single aerosol particle at multiple wavelengths (Pinnick et al. 2004; Pan, Pinnick et al. 2007) provides some degree of discrimination; however, even the most sophisticated optical techniques cannot reliably discriminate beyond the difference between mold, humic acid, and bacteria. This is not to suggest that these techniques are without value, to the contrary. These techniques provide the only means to rapidly estimate the temporal variability of biological aerosols. Pöschl et al. (2010) and Huffman et al. (2012) used UV-LIF, aerosol mass spectrometry, and many other techniques to investigate the roles of biogenic rainforest aerosols, including biological aerosols, as cloud condensation nuclei. Gabey et al. also examined biological aerosol populations above and below the forest canopy. Their results also demonstrate a diurnal pattern to bioaerosol concentrations, similar to previous studies. Further, they observed transient increases biological aerosol below the canopy, which they attribute to fungal spore release, that correlates with increases in relative humidity (RH).

In order to begin to further understand the factors that contribute to observed fluctuations in ambient biological aerosol, measurements of single-particle LIF and local meteorological conditions were made from 1–10 November 2008 on the campus of The Johns Hopkins University Applied Physics Laboratory in Laurel, MD. By examining the relationship between the local meteorology and rapid measurements of biological aerosol concentration, the processes that result in the observed changes in biological aerosol concentrations were examined.

METHODS AND INSTRUMENTATION

An Instantaneous Bioaerosol Analysis and Collection (IBAC; FLIR/ICx/S3I, Reiserstown, MD, USA) sensor was used to sample ambient aerosol with a 1 Hz resolution and provide number concentration of bioaerosol in two size bins (approximately 0.5–1.7 microns and 1.7 to approximately 10 microns) as well as a total bioaerosol concentration and total measured particle concentration in the same size categories. In this instrument, a particle is considered biological if its integrated fluorescence from 450 to 600 nm (excited at 405 nm) exceeds a preset threshold. Aside from detailed patent information (Silcott et al. 2006), the IBAC system has not been described in detail in the open literature. However, the use of 402–405 nm excitation sources as a low cost replacement for more expensive 355–365 nm laser sources has been described. Ho et al. (2001) studied a 402 nm laser diode as compared to 340–360 nm laser source. They reported that the performance of sensors using a 340–360 nm source could be replicated with the 402 nm diode. Others (Selbach and Kuhlmann 1999; Solovieva et al. 1999; Van Schaik et al. 1999) have shown that this change in excitation wavelength likely shifts the excited species in the biological material from nicotinamide adenine dinucleotide (NADH) to flavin molecules (such as riboflavin). Excitation–emission studies presented in Pan, Eversole et al. (2007), on a variety of biological materials that could be found in aerosol, demonstrated that most dried biological materials fluoresce when excited by light in the 405-nm range, including vegetative bacterial cells of both gram negative and gram positive varieties and pollens.

The IBAC was set up outdoors on a third floor terrace on the campus of The Johns Hopkins University Applied Physics Laboratory and allowed to sample autonomously. Temperature, RH, atmospheric pressure, wind direction, and wind speed were measured every 2 s using a Transportable Automated Meteorological Station (All Weather, Inc., Sacramento, CA, USA). These meteorological measurements were physically collocated with aerosol measurements.

Comparison between biological aerosol data and meteorological measurements required reducing the temporal fidelity of the aerosol data to match that of the meteorological data and identifying an appropriate interval over which to make a comparison. The 1-Hz biological data were reduced to 0.5 Hz simply by taking the average of the measured concentrations over the 2-s interval over which the meteorological data was measured. Identifying the temporal interval to perform the correlation analysis over was more challenging. Cursory inspection of the bioaerosol and meteorological data indicated that a relationship might exist between some of the variables. Such a relationship would be consistent with past studies (Lighthart and Shaffer 1995; Brodie et al. 2007; Gabey et al. 2010); however, a simple correlation between the datasets over the entire measurement period did not demonstrate this relationship. Along these lines, several schemes were attempted to find statistically significant correlations over the entire period, including allowing for lags in time between meteorological changes and aerosol fluctuations, but none of these attempts produced strong relationships. Fluctuations in aerosol concentration and therefore biological aerosol concentration over time could be due to many factors that are unrelated to the meteorology, such as changes in the source of the measured particles. Further, the meteorological variables could vary due to large-scale weather systems. After careful consideration, a diurnal period was used to evaluate these relationships. Both meteorological and biological aerosol data were divided into 12-h bins, from 12 AM to 12 PM, and subsequently from 12 PM to 12 AM local time. In this way, day-to-day variations in aerosol sources and the effects of large-scale weather patterns might be minimized, except in the cases where sudden changes occurred. Pearsons R values were used to examine the strength of the relationship between the bioaerosol and each meteorological variable over these 12-h periods. The R values were binned into three categories to simplify the description of the findings: (1) a strong value was assigned to R values

exceeding ±0.7; (2) a medium value was assigned to *R* values between ±0.4 and ±0.7; and (3) a weak value was assigned to *R* values between ±0.1 and ±0.4. Pearsons *R* values between 0 and ±0.1 are considered to be uncorrelated. Although these categories are somewhat arbitrary, it is useful to bin the results as a visual aid in interpreting the data.

RESULTS

The ambient environment on the campus of The Johns Hopkins University Applied Physics Laboratory in Laurel, MD, was continuously measured from 1 November through 10 November 2008. This extended collection period allowed observations of diurnal patterns in the ambient aerosol as well as the effect of meteorological events, such as precipitation, that may cause fluctuations in particle sizes and concentrations of atmospheric aerosol.

Observations over the 2-week sampling period (Figure 1a) indicate significant fluctuations in the meteorological conditions during that time period. The diurnal patterns in temperature and RH are readily apparent in the time series. Total and biological aerosol concentrations, measured by the IBAC, are shown

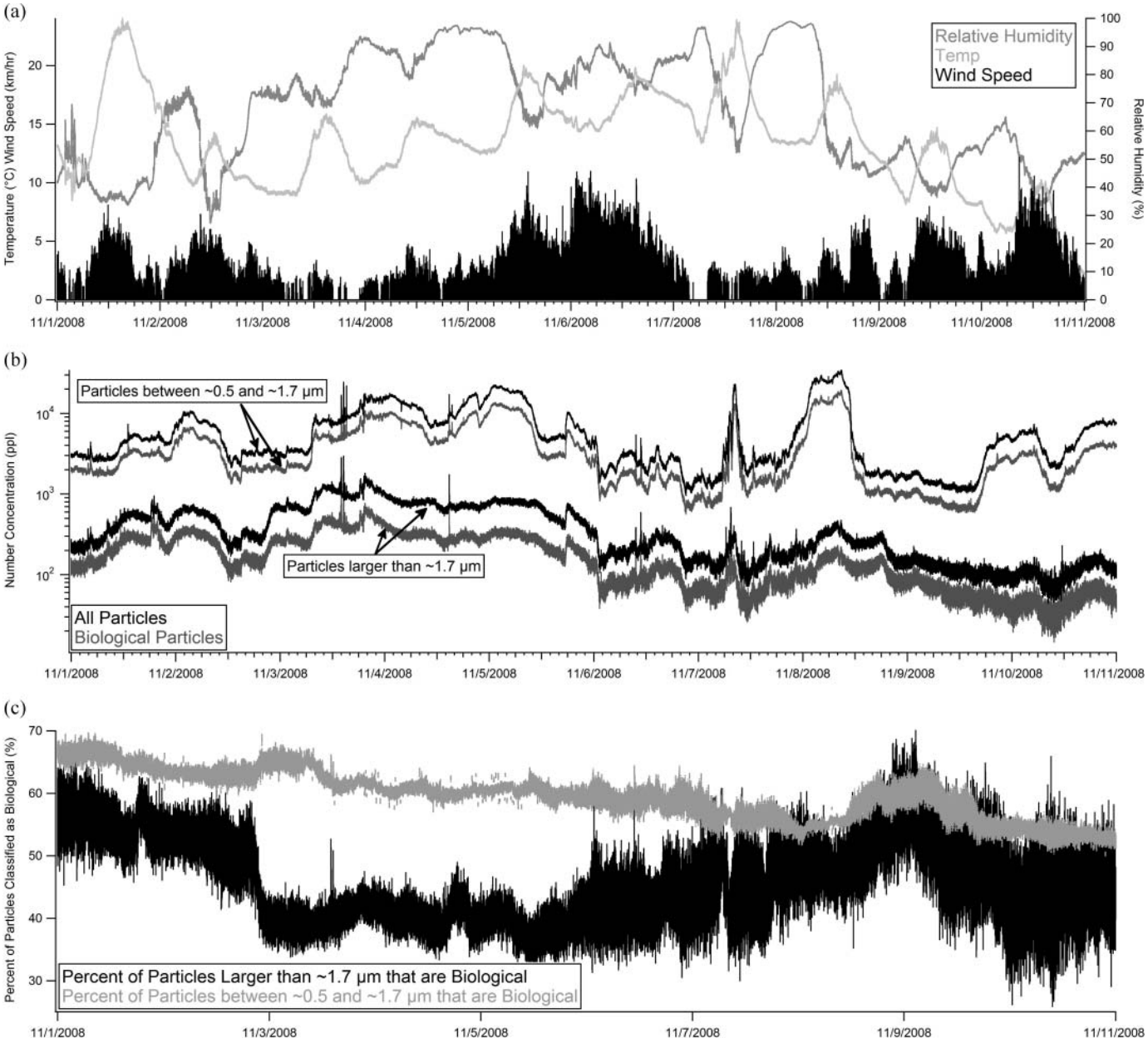

FIG. 1. Time series of: (a) ambient temperature, relative humidity, and wind speed over the 10-day sampling period, (b) IBAC measurements of total and biological particle concentrations in two size bins, and (c) the percent of particles classified as biological by the IBAC.

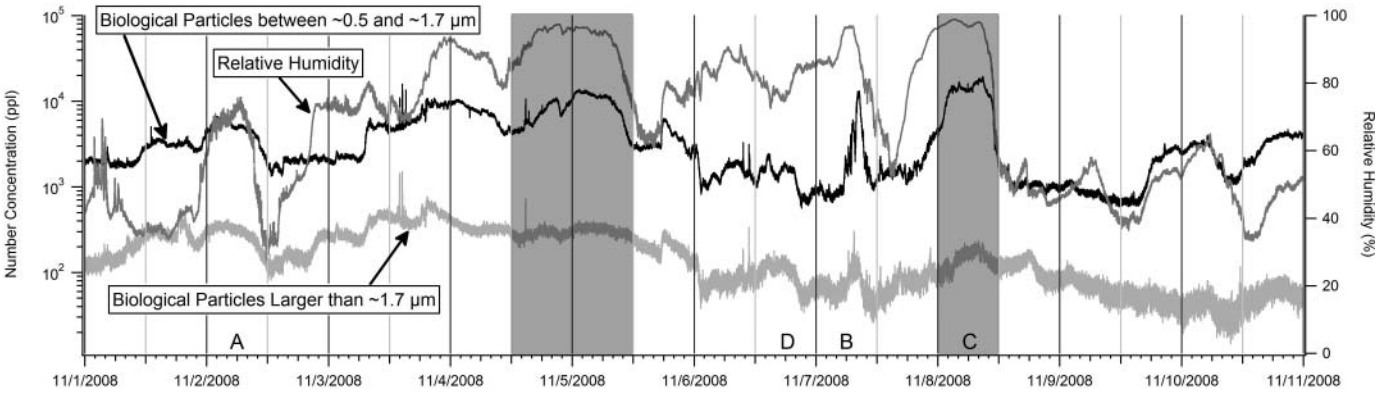

FIG. 2. Biological particle concentration for both the small (0.5– 1.7 µm) and large (1.7–10 µm) IBAC size bins and local ambient relative humidity as observed during the study period. Vertical lines represent the times over which correlation analysis with the meteorological variables was performed. The lettered regions represent the different example correlation plots from Figure 3 (A is 4A, B is 4B, C is 4C, and D is 4D). Shaded regions indicate periods during which accumulated precipitation was measured at the local weather station.

in Figure 1b. A visual examination of this data indicates that fluctuations in biological and total particle concentrations have similar fluctuations over the time period that was measured. It is important to note that despite the close relationship between biological particle concentrations and total particle concentration, the relationship is not identical. Calculations of percent of the total particle concentration that was classified as biological (Figure 1c) indicate that the percent of biological particles fluctuates over the 10 days of observations. In general, the percentage of small biological particles declines from almost 70% to just below 60%, and the percentage of large biological particles fluctuates from greater than 60% to less than 40% over the observation period. The high percentage of biological particles indicates that factors affecting the biological particles may dominate the trends in the data, but variability in the percentage indicates that biological and nonbiological particles do not behave identically during this period.

When comparing the meteorological data to the aerosol data, several interesting comparisons can be made. One readily apparent observation is that increases in the concentration of small biological aerosol, measured by the IBAC, seem to correspond to increases in the RH (Figure 2).

The correlation analysis, performed at 12-h intervals, indicates several relationships between both large and small biological aerosol concentrations and meteorological variables. The strongest set of these relationships appears to be between RH and the small biological aerosol, as indicated by inspection. Statistically strong relationships ($R > 0.7$) were very prevalent in both AM and PM time periods (Table 1), but more so in the PM periods. Further, relationships between temperature (dominantly anticorrelated) and barometric pressure also seem to be prevalent during the same period. The fact that both ambient RH and ambient temperature would be oppositely related is not surprising due to the dependence of the saturation vapor pressure, and therefore the RH, on the ambient temperature, particularly within a 12-h time frame.

A visual inspection of the correlation indicates several important factors that are not immediately obvious when comparing *R* values. Figure 3 shows example correlations between the concentration of large biological aerosol and RH. The medium and weak correlations shown (Figure 3b and c, respectively) have very similar trends to strong correlations, but have a significant increase in the spread of data near 100% RH. In general, 12-h periods where strong correlations were observed do not have RH values approaching 100%, and so it may be that the relationship between RH and these particles changes near 100% RH.

Rain was observed on several days during the study. Rainy conditions were first observed during the 12-h period beginning at noon on 11/4/2008 and ending around noon the following day. Accumulation of precipitation was light and intermittent during that period (as reported by the local weather station), but cloudy and foggy conditions accompanied by high RH persisted during that time (Figure 2). During the AM period of 11/8/2008, similar conditions also occurred. Consistent with the dominant trends in the data, the high RH measurements during these times corresponded to higher biological particle concentrations (Figure 2 and Table 1).

In general, higher RH correlates with higher concentrations of observable biological aerosol, but in a few cases an anticorrelation is observed. Figure 3d shows an example of this anticorrelation. An important observation is that the independent variable (RH) varied only a very small amount (less that 10%) over the entire 12-h period. Except in a very few cases, no, or only weak to medium, relationships are observed between the local wind direction and speed and either the large or small biological aerosol concentration (Table 1). This may indicate that changes in the local sources of biological particles did not significantly impact the biological aerosol concentration, or that the observed changes in the wind speed and direction had minimal impact on the aerosolization of biological particles.

Relationships between the total aerosol concentration and the meteorological data indicate relationships very similar to

TABLE 1
Pearson's R values for correlations between large (A) and small (B) biological aerosol concentrations and large (C) and small (D) total aerosol concentrations and measured meteorological variables

	A					B				
	00:00:00 – 11:59:59					00:00:00 – 11:59:59				
Date	RH	Temp	Pressure	Wind S	Wind D	RH	Temp	Pressure	Wind S	Wind D
1-Nov	−0.673	0.865	−0.198	0.017	0.488	−0.269	0.629	0.204	0.006	0.232
2-Nov	0.868	0.281	−0.820	−0.340	0.432	0.837	0.249	−0.767		−0.400
3-Nov	0.034	0.814	−0.209	0.000	0.015	0.312	0.651	−0.014	0.004	−0.002
4-Nov	0.437	−0.451	0.734	0.087	0.320	0.919	−0.959	0.741	−0.112	0.208
5-Nov	0.722	−0.732	0.584	−0.263	0.185	0.980	−0.976	0.846	−0.404	0.182
6-Nov	−0.218	0.275	0.515	−0.172	0.025	0.112	−0.043	0.634	−0.181	0.082
7-Nov	0.583	−0.564	0.418	−0.014	−0.025	0.205	−0.159	0.564	0.003	0.166
8-Nov	0.215	−0.192	−0.596	0.005	−0.249	0.661	−0.586	−0.231	−0.010	−0.278
9-Nov	0.310	−0.216	−0.806	−0.516	−0.471	0.268	−0.153	−0.887	−0.546	−0.517
10-Nov	0.433	−0.372	−0.430	−0.003	−0.269	0.818	−0.630	−0.896	−0.004	−0.385
	12:00:00 – 23:59:59					12:00:00 – 23:59:59				
1-Nov	−0.104	0.206	−0.158	−0.015	−0.033	0.294	0.073	0.043	0.037	0.001
2-Nov	0.848	−0.724	0.833	0.006	0.008	0.695	−0.781	0.680	0.001	−0.030
3-Nov	0.337	−0.324	0.065	0.006	−0.039	0.869	−0.849	−0.438	0.010	−0.137
4-Nov	0.418	−0.326	−0.090	−0.006	−0.037	0.835	−0.883	−0.521	−0.005	−0.051
5-Nov	0.101	0.231	0.575	0.132	0.020	0.502	−0.340	0.045	−0.004	0.233
6-Nov	−0.858	0.786	−0.528	0.374	0.319	−0.806	0.724	−0.540	0.410	0.285
7-Nov	0.568	−0.577	−0.722	0.006	0.184	0.708	−0.706	−0.783	0.006	0.110
8-Nov	0.561	0.659	−0.712	−0.016	−0.104	0.657	0.579	−0.314	−0.002	−0.196
9-Nov	−0.437	0.460	−0.515	0.183	0.142	0.964	−0.952	0.977	−0.417	−0.311
10-Nov	0.528	−0.464	0.367	−0.185	0.011	0.953	−0.921	0.878	−0.432	−0.192

	C					D				
	00:00:00 – 11:59:59					00:00:00 – 11:59:59				
Date	RH	Temp	Pressure	Wind S	Wind D	RH	Temp	Pressure	Wind S	Wind D
1-Nov	−0.689	0.879	−0.203	0.018	0.496	−0.252	0.622	0.219	0.005	0.226
2-Nov	0.878	0.274	−0.823	−0.342	−0.433	0.838	0.243	−0.763	−0.291	−0.398
3-Nov	0.047	0.812	−0.196	0.000	0.015	0.299	0.666	−0.035	0.004	0.000
4-Nov	0.351	−0.346	0.713	0.120	0.328	0.918	−0.960	0.732	−0.114	0.204
5-Nov	0.575	−0.569	0.509	−0.149	0.143	0.981	−0.976	0.851	−0.404	0.179
6-Nov	−0.255	0.281	0.561	−0.193	0.029	0.135	−0.041	0.640	−0.183	0.084
7-Nov	0.514	−0.472	0.504	−0.011	0.025	0.205	−0.160	0.565	0.003	0.166
8-Nov	0.225	−0.188	−0.604	0.002	−0.252	0.675	−0.601	−0.220	−0.010	−0.279
9-Nov	0.300	−0.212	−0.754	−0.490	−0.439	0.193	−0.075	−0.859	−0.515	−0.502
10-Nov	0.557	−0.453	−0.606	−0.003	−0.327	0.819	−0.632	−0.893	−0.004	−0.384
	12:00:00 – 23:59:59					12:00:00 – 23:59:59				
1-Nov	−0.154	0.312	−0.205	0.042	0.011	0.360	0.009	0.033	0.003	−0.026
2-Nov	0.835	−0.694	0.824	0.004	0.006	0.639	−0.750	0.619	0.002	−0.032
3-Nov	0.289	−0.281	0.152	0.003	−0.034	0.872	−0.851	−0.462	0.010	−0.139
4-Nov	0.272	−0.279	−0.244	−0.010	−0.035	0.839	−0.879	−0.510	−0.005	−0.052
5-Nov	0.030	0.329	0.678	0.163	−0.014	0.506	−0.355	0.025	−0.009	0.238
6-Nov	−0.869	0.825	−0.619	0.402	0.326	−0.811	0.721	−0.524	0.409	0.285
7-Nov	0.420	−0.434	−0.672	0.008	0.173	0.710	−0.708	−0.782	0.006	0.110
8-Nov	0.665	0.782	−0.814	−0.015	−0.158	0.661	0.589	−0.323	−0.001	−0.203
9-Nov	−0.422	0.451	−0.485	0.180	0.131	0.966	−0.955	0.980	−0.416	−0.309
10-Nov	0.561	−0.485	0.381	−0.190	0.019	0.958	−0.929	0.888	−0.439	−0.199

Note: Values are coded (black with white text: $R > 0.7$; gray with white text: $0.4 < R < 0.699$; black with italic white text: $R < -0.7$; gray with italic white text: $-0.699 < R < -0.4$) to indicate the relative strength of the correlation, as described in the text. Hashed date boxes indicate periods during which accumulated precipitation was measured at the local weather station.

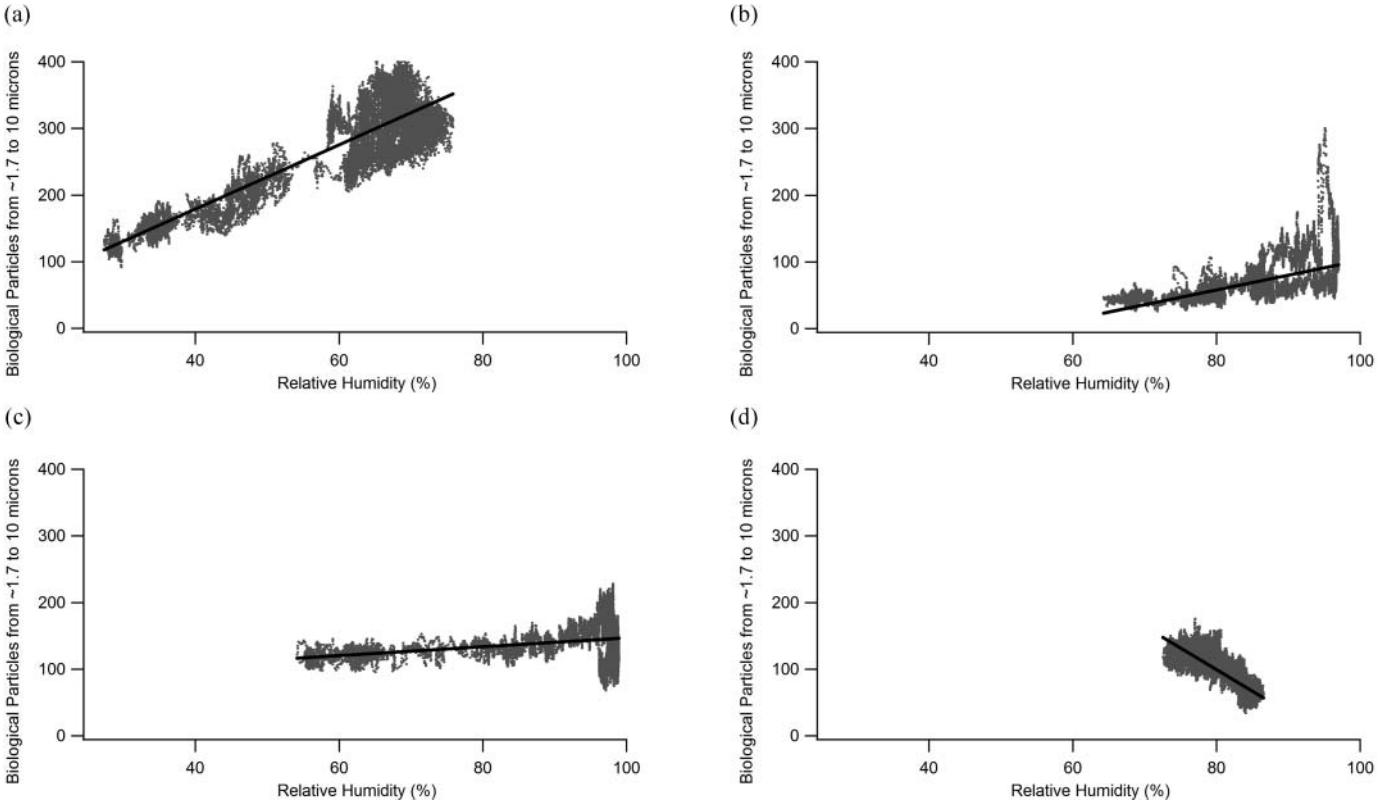

FIG. 3. Correlations between relative humidity and large biological particle aerosol concentrations: (a) is from the AM period of 11/2 showing a typical strong correlation; (b) is from the AM period of 11/7 showing a medium correlation with significant variability above 90% RH; (c) is from the AM period of 11/8 showing a weak correlation with significant variability near 100% RH; (d) is from the PM period of 11/6 showing a strong anticorrelation where the RH changed very little during the 12-h period.

those seen in the biological particle relationships (Table 1C and D). As discussed previously, the high percentage of biological particles observed during this period (Figure 1c) may account for the similar trends.

Rubel (1997) made measurements of the hygroscopic properties of *Bacillus atrophaeous* (*Bg*; formerly *globigii*). They found that these spores grew in the presence of even low RH, compared with dry spores. The RH observed in this study ranged from less than 30% to almost 100%, over the course of the time period measured. These RH conditions could result in the growth of biological aerosol if the results of Rubel can be applied broadly to all biological particles. This particle growth could account for the changes in the biological aerosol concentration as smaller biological aerosol uptake water and grow to a size observable in the small-size bin of the IBAC and some fraction of small biological aerosol previously observed in the small-size bin of the IBAC grow large enough to be placed in the large-size bin.

SUMMARY AND DISCUSSION

Observations of biological aerosol, derived from UV-LIF measurements of single particles excited at 405 nm and select meteorological variables (temperature, RH, barometric pressure, wind speed, and wind direction) were made from 1–10 November 2008. Analyses of these data demonstrate several relationships between the local meteorology and the biological aerosol. When data were binned into 12-h blocks, several relationships between the biological aerosol data and the meteorology became apparent. Generally speaking, increase in RH is correlated to increases in the biological aerosol concentration for both large and small particles. As might be expected, this relationship is also apparent in an anticorrelation with temperature. This relationship between biological aerosol concentration and RH may be explained by the uptake of water by biological particles, due to their hygroscopicity. Further, there are few observable relationships between the measured wind variables and biological aerosol concentrations. This indicates that the changes in wind speed and direction were not significant enough to affect the local sources of biological particles, or that the changes in the source did not dramatically affect the biological aerosol concentration.

Taken together, the findings of this study imply the conclusion that aerosol processes, such as water uptake, dominated the changes in biological aerosol concentration that would be observed over timescales up to hours, in this dataset. This is in contrast to past studies (Lighthart and Shaffer 1995; Brodie

et al. 2007), whose correlations were observed over longer periods (hours to days), that suggested changes to the source of biological particles, with changes in the meteorology, might be responsible for the observed relationships. The timescale over which correlations are observed in this dataset is very short (2 s), which does not leave time for changes to the source to have an effect on particle concentrations. The strong relationship between RH and biological aerosol concentration and the comparatively weak relationship between those aerosol and the measured wind variables over the same time frame indicates that water uptake by the biological particles, making them more observable by the IBAC, was likely responsible for the change in concentration rather than a change in the local sources of biological particles. Likewise, it is understandable that the uptake of water would have a greater effect on the small particle category than on the large category. The addition of particle volume from liquid water will cause a greater diameter change for the smaller particles than the larger ones. This finding warrants future study of both the hygroscopicity of biological particles and how other aerosol growth processes affect the properties of atmospheric biological aerosol since these properties may directly impact the role those biological particles ultimately play in other atmospheric processes.

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