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A comparison of on-line and off-line bioaerosol measurements at a biowaste site

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ABSTRACT

An air measurement campaign was carried out at a green-waste composting site in the South of Ireland during Spring 2016. The aim was to quantify and identify the levels of Primary Biological Aerosol Particles (PBAP) that were present using the traditional off-line, impaction/optical microscopy method alongside an on-line, spectroscopic approach termed WIBS (Wideband Integrated Bioaerosol Sensor), which can provide number concentrations, sizes and "shapes" of airborne PBAP in real-time by use of Light Induced Fluorescence (LIF). The results from the two techniques were compared in order to validate the use of the spectroscopic method for determining the releases of the wide-range of PBAP present there as a function of site activity and meteorological conditions. The seven-day monitoring period undertaken was much longer than any real-time studies that have been previously performed and allowed due comparison between weekday (working) activities at the site and weekend (closed) releases. The time-span also allowed relationships between site activities like turning, agitation or waste delivery and the WIBS data to be determined in a quantitative manner. This information cannot be obtained with the Andersen Sampling methods generally employed at green-waste management sites. Furthermore, few specific bioaerosol types other than *Aspergillus fumigatus*, are identified using the traditional protocols employed for site licensing purposes. Here though the co-location of WIBS with the impaction instrument made it possible to identify the real-time release behaviour of a specific plant pathogenic spore, *Ustilago maydis*, present after green-waste deliveries were made by a local distillery.

1. Introduction

The air quality determined for indoor or outdoor ambient or occupational environments is significantly affected by the levels of particulate matter present. The health risks associated with particulate matter (PM_{10}, $PM_{2.5}$ and PM_1), mainly generated by combustion processes and from non-exhaust vehicle emissions, are well known (Pöschl, 2005, Cohen et al., 2005). Although the effects on humans and plant life of corresponding biological particulate matter (termed Primary Biological Aerosol Particles, PBAP, or bioaerosols) have been studied for many years (Pinnick et al., 1995, Després et al., 2012, Poehlker et al., 2012), the real-time monitoring of this type of airborne material at known "hot-spot" locations such as composting sites has lagged behind. Hence there are few studies published that quantify the number and type of fungal spores being released at places where individuals (particularly those who are immuno-compromised) are present or where susceptible crops may be affected on a routine basis. Green-waste management sites are a good example of such outdoor spaces that are often found in rural areas close to crops, served by staff and visited by customers. There are licensing guidelines set by national and regional environmental agencies for commercial composting operations. These primarily focus on people inhabiting the surrounding areas and for site workers themselves. On Irish compost sites the monitoring of a limited number of harmful biological particles like *Aspergillus fumigatus*, are performed off-site just once a year for a short time period with the results returned some days later to the local management (Williams et al., 2013, O'Connor et al., 2015). Therefore no indication of minute-to-minute or hour-to-hour changes in bioaerosol releases are made even when the normal agitation processes associated with the

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composting process occur or when deliveries are made. The English Environment Agency guidance specifies that to be deemed acceptable, monitoring should be carried out at times of high site activity *e.g.* turning, shredding, screening.

One of the most important aims of the waste industry is to maximise the benefit that can be recovered from global waste. Composting is a method of waste management that is based on the biological degradation and stabilisation of organic matter performed under aerobic conditions (Sanchez-Monedero et al., 2005, Wéry, 2014, Avery et al., 2012, Beffa et al., 1998, Hryhorczuk et al., 2001). However, it has now become an important industrial activity, which impacts on waste management targets. Therefore, a substantial increase, throughout the world, has become apparent for the use of local composting facilities to manage and utilize green- and food-waste (Hryhorczuk et al., 2001, Wéry, 2014, Recer et al., 2001).

Many of the microbial processes involved in the degradation and transformation processes from green-waste to fertilizer are now understood and airborne bioaerosols are a natural, unavoidable outcome of the processing. Hence there can be major releases of sometimes pathogenic and irritant bioaerosols at composting sites especially during activities involving the vigorous movement of material such as shredding, compost pile turning and compost screening (Wéry, 2014, Taha et al., 2006, Sanchez-Monedero et al., 2005, Hryhorczuk et al., 2001, Millner et al., 1980, Taha et al., 2005, Gillum and Levetin, 2008, Sanchez-Monedero and Stentiford, 2003, Pankhurst et al., 2011). The increased emissions of bioaerosols are currently causing concerns related to potential, occupational health impacts on the workers, visitors to commercial composting facilities and nearby residents (Sanchez-Monedero et al., 2005, Wéry, 2014, Van der Werf, 1996). Furthermore, certain crops in the site vicinity have the potential to be harmed. Of course, the human health aspect is the reason why licensing authorities have put in place (although only in some countries) regimes where monitoring techniques involving the impaction of viable microorganisms onto a culture *e.g.* Andersen Sampling must be used to measure levels of mesophilic bacteria and the fungal spore *Aspergillus fumigatus* (Williams et al., 2013, Pankhurst et al., 2011).

Typical mass concentrations for fungal spores of $\sim$1 μg m^{-3} are found in continental boundary layer air with estimated global emissions being $\sim$50 Tg/yr (Poehlker et al., 2012, Elbert et al., 2007). Fungal spores are the most abundant of the PBAP to be found in many local environments (Després et al., 2012, Elbert et al., 2007, Womiloju et al., 2003) and occur wherever decaying vegetation is present as it provides a food/energy source. The average size of these spores sits in the range between 2 and 10 μm ($PM_{2.5}$ and PM_{10}) (Simon-Nobbe et al., 2007) and therefore potentially have adverse health effects on humans. For example their presence has long been associated with asthma and other reactive airway diseases such as allergic bronchopulmonary mycoses, rhinitis, allergic sinusitis, hypersensitivity pneumonitis and allergic aspergillosis (Simon-Nobbe et al., 2007, Chaudhary and Marr, 2011). Deterioration in the pulmonary function of people with chronic asthma and cystic fibrosis and pathogenicity in immune-compromised people *e.g.* patients undergoing chemotherapies for cancer are of particular concern to the medical community.

The current regulation set by the English Environment Agency (and adopted by the Irish EPA) is for compost sites to demonstrate "acceptable levels" of mesophilic bacteria (10^3 CFU m^{-3}) and the fungal spore *Aspergillus fumigatus* (500 CFU m^{-3}) (Williams et al., 2013, Pankhurst et al., 2011). The benchmarks have been set as a $\times$10 factor relative to outdoor ambient levels. *Aspergillus fumigatus* is monitored as a total marker for fungal spore emissions on compost sites. It is a known human pathogen (Simon-Nobbe et al., 2007, Horner et al., 1995, Fischer et al., 1998, Abba, 2004) with compost heaps being an environmental source of the spore due not only to the self-heating process but also to the intrinsic thermo-tolerance of the spore (Vincken and Roels, 1984, Gillum and Levetin, 2008, Van der Werf, 1996, Millner et al., 1977). Hence composting plant operations must be located 250 m from any other property in the vicinity (Sanchez-Monedero et al., 2005).

Andersen sampling is the current method of choice used for determining the concentrations of mesophilic bacteria and *Aspergillus fumigatus* at compost sites although IOM or CEN filters followed by culture in the laboratory may also be used. It is a traditional off-line technique for monitoring bioaerosols and relies on pumped air impaction onto an agar gel. The collected samples are then returned to the laboratory for further cultivation (Després et al., 2012). The technique has two major limitations: (i) it only measures the spores and bacteria that are viable for its specific media and (ii) it only collects a small volume of air, off-site, for a short period of time (Hryhorczuk et al., 2001). The reason for making collections over 2–10 min spans is because, as the micro-organisms are impacted directly onto the agar surface, there is a danger of the plate becoming overloaded in environments that give rise to high concentrations. Compost sites are a good example of this possibility and so sampling is never performed on-site when using Andersen sampling, rather 100–250 m upwind and downwind from the source in Ireland (Cartwright et al., 2009, Eduarda and Heederik, 1998, Williams et al., 2013).

Andersen sampling does not provide a comprehensive list of fungal spores that may be emitted from a green-waste site and in order to undertake such a counting and identification regime an impaction/optical microscopy approach was used in this campaign. The equipment was set up, on site and adjacent to a real-time monitor for biological/fluorescent particulates. This novel approach gave a direct comparison between the traditional aerobiological method that provided unequivocal information about the number and identity of many fungal spores in local air with an instrumental technique, based on intrinsic fluorescence and optical scatter measurements of airborne particulates, termed the Wide-band Integrated Bioaerosol Sensor (WIBS). The model used on this campaign (WIBS-4A) was manufactured by Droplet Measurement Technologies.

The WIBS instrument has now been utilized in many field campaigns directed to the measurement of bioaerosols. It has been deployed in a number of locations around the world from tropical rainforests (Gabey et al., 2010) to urban/rural and coastal sites to measure outdoor ambient air (Healy et al., 2014, Crawford et al., 2014). However, it has only been deployed and reported in the literature once before in an outdoor green-waste occupational setting, albeit for a very short period of time (1–2 days) (O'Connor et al., 2015).

In the seven-day, outdoor, on-site campaign reported here online continual measurements of fluorescent and non-fluorescent particles released from a green-waste site have been quantified using the WIBS method. The data obtained are then compared to those obtained using a Hirst-type, impaction device (followed by analysis using optical microscopy). The monitoring period undertaken was much longer than any real-time studies that have been previously performed and allowed due comparison between weekday (working) activities at the site and weekend (closed) releases. The time-span also allowed relationships between site activities like turning, agitation or waste delivery and the WIBS data to be determined in a quantitative manner. This information cannot be obtained with the Andersen Sampling methods generally employed at green-waste management sites. Furthermore, few specific bioaerosol types other than *Aspergillus fumigatus*, are identified using the traditional protocols employed for site licensing purposes. Here though the co-location of WIBS with the impaction

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