

CRANFIELD UNIVERSITY

Mohanad Abdulameer Jawad

Evaluation of a novel biosensor for bioaerosol sampling and
modelling

School of Water, Energy and Environment
PhD

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Academic Year: 2018 - 2019

Supervisor: Dr Gill Drew
Associate Supervisor: Prof Sean Tyrrel
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the degree of PhD

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ABSTRACT

The identification and quantification of bioaerosols in the atmosphere is a significant subject of study. Bioaerosols emitted from composting sites are released directly to the atmosphere, which can be potentially harmful to human health. Although there are several studies undertaken to improve the risk assessment of bioaerosols from composting facilities, this subject still requires further research. Some studies focused on the direct detection of bioaerosol from the composting facilities, and the others focused on the dispersion modelling of bioaerosol. In both cases, the bioaerosol data detected in the direct measurement or the data used in the modelling were dependant on traditional sampling techniques. These techniques provide limited details about the bioaerosol emitted from the composting facilities. These traditional data provide spatial and temporal snapshots of bioaerosol emissions concentrations, and these concentrations represented particular components of bioaerosol. The SIBS (Spectral Intensity Bioaerosol Sensor) is a new sampling device and can provide significant data about bioaerosols, as it can sample continuously over long periods. These advantages made the SIBS able to provide an important picture of the variation of bioaerosol concentrations with time.

Several sampling experiments have been done in different environments to measure the bioaerosol concentrations using the SIBS alongside the filtration sampling technique (IOM/SKC). The collected data from the composting site showed the advantages and disadvantages of each sampling technique, and also increased the knowledge of bioaerosol emissions from composting facilities. These novel data were used to calculate emission rates of bioaerosol from agitation activities to improve the output of the bioaerosol dispersion modelling. The results achieved have presented new visions to the current understanding of the characterisation and dispersal of bioaerosols emitted from composting facilities and would be anticipated to make an important contribution to improve the risk assessment of bioaerosol from composting facilities to meet the increased regulatory requirements by Environment Agency.

Keywords:

SIBS, IOM/SKC, ADMS, composting

For my mother and father, Hiyam and Abdulameer

Thank you for all the unlimited sacrifices and support you have made
along the years

For my beloved wife, Alrabab

Thank you for your patience throughout the PhD

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1 Introduction

The large amounts of waste going to landfill are one of the most significant environmental challenges of our time. Several strategic stages are used to reduce the amount of waste directed to landfill. These strategies are referred to as the waste hierarchy (DEFRA, 2011a), which is shown in Figure 1-1.

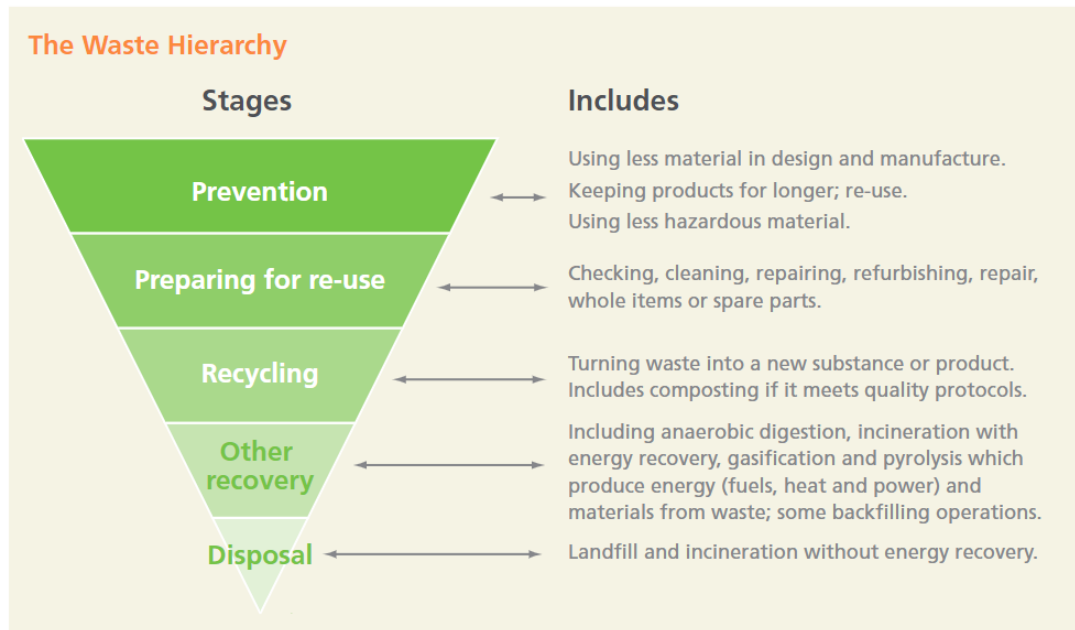


Figure 1-1 The waste hierarchy, highlighting the stages in place to reduce the amount of waste sent to landfill. Image duplicated from DEFRA (2011).

As shown in figure 1-1, composting plays a key contributor role to reducing the amount of waste directed to landfill, by transforming biodegradable waste to produce a useful material (Stagg et al., 2010). Composting is the process that transforms complex organic materials into more simple organic structures in aerobic conditions from biological activities of microorganisms (Swan et al., 2003). Due to European legislation, the quantities of material being composted are increasing (European landfill directive [1999/31/EC], Sykes, Jones and Wildsmith, 2007).

The essential factor in composting process is the presence of microorganisms, basically bacteria and fungi, which can become airborne. These microorganisms and their related cellular components are known as bioaerosols (ADAS and SWICEB, 2005). The concern about bioaerosol results from their potential for harmful effects that can be associated with exposure. Therefore, composting facilities are important sources of bioaerosols because they emitted from open windrow composting sites at high levels. Additionally, bioaerosols dispersed from these sources are not controlled or contained, and are not easy to capture (Taha et al., 2006).

Understanding of dispersal and composition of bioaerosol emissions emitted from composting facilities is still limited, despite over a decade of research about this subject (Pankhurst, 2010). Knowledge of bioaerosol dispersal is essential to inform the regulation and planning of composting facilities.

Environment Agency (2010; 2018) recommended that bioaerosol concentrations must stay within an acceptable level within 250 metres of the site boundary, or the nearest sensitive receptor, whichever is closer. The acceptable levels are currently set at 1000 and 500 Colony Forming Units per cubic metre (CFU/m³) for Total Bacteria and *Aspergillus fumigatus*, respectively.

Although there have been many studies done to improve the risk assessment of bioaerosols from composting facilities, further improvements are still required. Some studies focused on direct detection of bioaerosol from composting facilities, and others focused on dispersion modelling of bioaerosol from the same source. In both cases, the bioaerosol data detected in the direct measurement or the data used in the modelling were depended on traditional sampling techniques (Douglas, 2013; Douglas et al., 2017a; Drew et al., 2007b; Drew et al., 2005; Pankhurst, 2010; Taha and Pollard 2004a; Taha et al., 2006; Tamer Vestlund, 2009; Nasir and Tyrrel, 2017).

Currently, bioaerosol monitoring methods can provide insights into emissions in space and time (Environment Agency, 2018). These insights provide limited details about the bioaerosol emitted from the composting facilities. These kinds

of data are also used in the dispersion modelling of bioaerosols from composting facilities.

As there are insufficient details in these data, so the model output is also based on limited inputs. In particular, the current sampling techniques detect viable and culturable bioaerosols, however there is a large fraction of bioaerosols that are not enumerated in this way, being non-culturable, non-viable, or unable to grow according to the conditions selected for culture (Pankhurst, 2010; Swan et al., 2003).

These non-viable bioaerosols remain of concern with regards to human health. There are many available techniques that can detect these fractions of bioaerosols, including those based on direct counting and molecular analysis. However, these techniques cannot detect bioaerosol emissions in real-time, and thus also provide a snapshot of the emission in time and space.

The Spectral Intensity Bioaerosol Sensor (SIBS) is a novel sampling device and has only been used for sampling of bioaerosols from composting facilities in the context of two ongoing studies. The SIBS has the potential to provide significant data about bioaerosols, as it can sample continuously for a long time. These advantages allows the SIBS to provide an important picture of the variation of bioaerosol concentrations with time. This time may be a few minutes, hours or a whole day, where the traditional samplers such as filtration sampling technique (the IOM/SKC) can give data about the bioaerosols for a single sampling period. In addition, the SIBS can detect a wide range of bioaerosol component depending on the fluorescent particles detected.

Furthermore, this novel sampling technique (the SIBS) saves time and effort when collecting the data, whereas the traditional sampling technique needs a long time to process and produce the results. The traditional techniques are also exposed to certain factors that affect their accuracy, such as contamination during the sampling or laboratory processing, and losing viability of the organisms for many reasons.

Therefore, these advantage of the new data from the SIBS (real-time detection (fluctuations) and the detecting the viable and non-viable bioaerosol) can make an important improvement of risk assessments of bioaerosol from composting facilities. The general aim of this thesis is therefore:

To examine to what extent a new sensor (the SIBS) can be useful to apply dispersion models more effectively to model bioaerosol dispersal in the open windrow composting environment.

This improvement will enhance the knowledge on how bioaerosols are released and dispersed. Dispersion modelling plays an important role in the prediction of the exposure. Subsequently, this will develop the site risk assessment methods and inform the Environment Agency's policies, as well as help in the planning of new composting facilities in the future.

First Objective: Analyse bioaerosol concentrations measured using the SIBS alongside traditional sampling techniques.

The project initially undertook a series of sampling experiments for bioaerosol emissions at composting facilities and other environments by using the SIBS and traditional sampling techniques. The resulting database was subject to statistical analysis to compare the two methods. The objectives of this comparison are:

- 1- Quantifying and characterising the fluorescent particles from the composting facilities using the SIBS.
- 2- Analyse how SIBS data relates to data collected using traditional methods as the present knowledge is based on traditional methods. This new analysis will help to place the SIBS data within the context of previous findings.

Second Objective: Determine whether SIBS data can be used to improve bioaerosol dispersion modelling from composting facilities.

Dispersion modelling of bioaerosol emissions continues to be challenging, primarily due to incomplete input data. The new data measured by the SIBS can

improve modelling of bioaerosol emissions from composting sites. This objective, therefore, used the database that collected during the sampling campaigns to determine the most appropriate methods to incorporate the SIBS data into the ADMS dispersion model.

1.1 Thesis structure

The aim and objectives and the order in which they are addressed in this thesis are conceptualised in figure 1-2. Generally, the thesis is divided into two parts, which are associated with objective 1 (sampling) and objective 2 (modelling).

This thesis was composed of eight chapters. Chapter 1 reviewed the importance of the project, the gap in knowledge, as well as the aim and objectives of the project.

Chapter 2 started by reviewing the key literature associated with research, specifically, composting, bioaerosols, monitoring methods of bioaerosols and dispersion modelling of bioaerosol from composting facilities.

Chapter 3 discusses all sampling methods used, including sampling techniques, sampling protocol, samples processing and calculations. Sampling strategies were detailed, and how each sampling experiment was completed. Chapters 4 and 5 present the results of the sampling experiments associated with the first objective in this thesis. Chapters 6 and 7 present the modelling work associated with the second objective of the project.

Aim: To examine to what extent a new sensor (the SIBS) can be useful to apply dispersion models more effectively to model bioaerosol dispersal in the open windrow composting environment

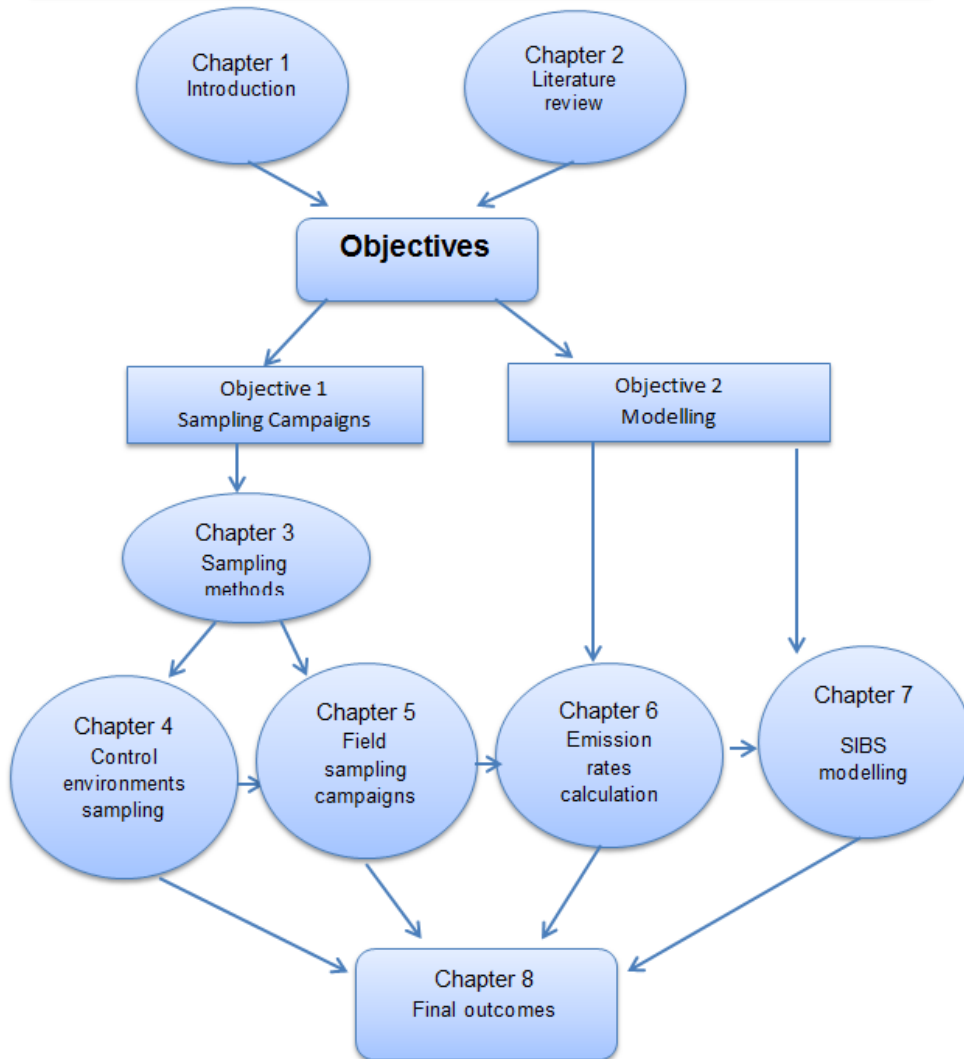


Figure 1-2 The general structure of the thesis, showing where each object is addressed throughout the thesis, and how the thesis objectives are correlated.

2 Literature review

2.1 Introduction

This chapter presents a critical analysis of current knowledge and literature on composting, bioaerosols, sampling and dispersion modelling of bioaerosols emitted from composting facilities.

2.2 Compost

2.2.1 Introduction

To maintain the fertility of soil and organic matter, farmers have practised composting for years. The composting process can be a treatment process for a wide range of organic substrates such as some municipal solid wastes, sewage sludge and agricultural and industrial by-products (Swan et al., 2002). Commercial composting operations are a key component of the UK waste management industry. It is anticipated that the total composting capacity in the UK will reach 5.9 million tonnes by 2012 (Pankhurst et al., 2011). This sustainable process aims to reduce the amounts of waste that discharged to landfill (Stagg et al., 2010).

Composting is the process that transforms complex organic materials into more simple organic structures in aerobic conditions from biological activities of microorganisms (Swan et al., 2003). This definition includes a wide range of activities, from piling manure at the bottom of a garden to advanced treatment processes for commercial wastes.

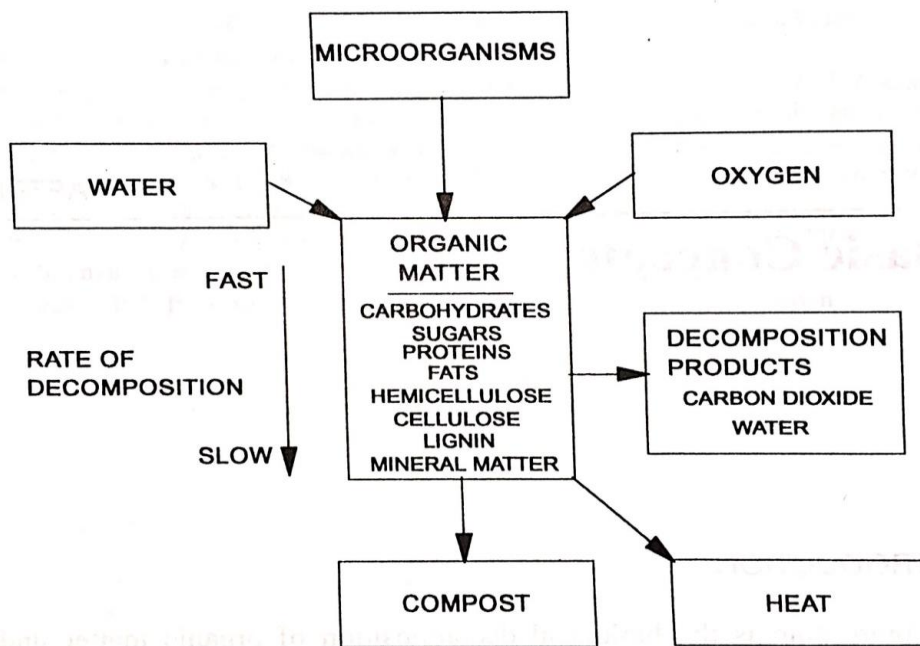


Figure 2-1 The basic composting process (Epstein, 1997).

However, even though the practices of composting have been quite different as quantities increase, the biological principle remains the same whatever the amount being composted (Gilbert et al., 2001). The main purpose of establishing composting as an effective and environmentally useful waste treatment process and as a manufacturing process is to produce a useful and marketable output.

2.2.2 Types of composting facilities

Practically, there are four composting technologies (Swan et al., 2003), described below. Open windrow system is the most commonly used type of composting facilities in the UK, with about 90% of total composting facilities (Sykes et al., 2007). However, there is a growing trend toward the in-vessel system, as this method can compost the animal by-product, food and catering waste and green waste as well.

1. Windrow systems

This type of composting is considered the simplest system. Currently, open-air turned windrow facilities composed about 78% of waste (Smith and Pocock, 2006). Most feedstock that is processed in this composting type is green wastes, sometimes called botanical residues or yard trimmings. In this system, the compost needs sixteen weeks to complete. The composting process consists of three basic activities which are shredding, turning and screening. Initially, the waste is shredded to a uniform size.

Generally, the feedstock is formed into piles, which are called windrows; shaped as an extended triangular prism. The exact shape depends on the materials and equipment used. The pollutant emissions from open windrow system are not controlled, because the windrows are open to air. These windrows need to be regularly turned to improve oxygen content and regulate temperature and moisture content. Taha et al., (2006) mentioned that bioaerosol concentration increased during the agitation activities. The last stage is screening, where the material is filtered to separate the final product from any contaminants (Swan *et al.*, 2002; Sharma, 2010; Douglas, 2013).

2. Aerated static piles

The aeration technique is what distinguishes this system from other types composting systems. The aeration system may be positive aeration (blow air) or negative (suck air) through the composting materials. The concentrations of oxygen and temperature are linked to the rate of the aeration. The negative pressure system may contain a biofilter or scrubber to filter air before discharge to the environment, whereas the positive pressure system may have layers of mature compost to work as an in situ biofilter. These types of techniques are widely used in the United States to compost sewage sludge (Sharma, 2010; Swan et al., 2002).

3. In-vessel systems

This system differs from the open windrow and aeration static system that it is usually enclosed, and thus having a high level of process and emission control.

There are six different types of in-vessel system:

- Containers
- Tunnels
- Agitated bays
- Rotating drums
- Silos or tower systems
- Enclosed halls (Swan *et al.*, 2002; Sharma, 2010).

4. Vermicomposting

Vermicomposting is the process that depends on the species of earthworms to decompose organic wastes. It usually needs a long time at an average temperature of about 35 °C (Swan *et al.*, 2002).

2.3 Bioaerosols

National and local concerns have arisen about the impacts on public health resulted from exposure to potentially harmful bioaerosols (airborne microorganisms) emitted from compost facilities. The essential factor in composting process is the presence of microorganisms, basically bacteria and fungi. Bioaerosols are released as a consequence of compost agitation activities (shredding, turning and screening), but do also occur naturally in the environment and exposure to bioaerosols is not limited to composting facilities (

Recycled Organic Unit, 2007); Sanchez-Monedero, Stentiford and Mondini, 2003; Swan et al., 2003).

Bioaerosols can be defined as a group of aerosolised biological particles that are produced by living organisms which includes dust, bacteria, fungi, spores, pollen, protozoa as well as cell components and fragments of cells (Cox and Wathes, 1995). The highest percentage of bioaerosols is microorganisms and their cellular components such as bacteria, fungal spores, actinomycetes, endotoxins, mycotoxins and glucans (Pankhurst, 2010).

Bioaerosols can be in the single form or attached to other to form aggregates of bioaerosols in different size and from a different origin. Due to environmental stress factors such as desiccation, temperature and ultraviolet radiation, most airborne microorganisms except for gram-positive bacteria and fungal spores may be killed quickly. Therefore, living bioaerosols represent a small proportion of total bioaerosols that emitted from composting facilities (Recycled Organic Unit, 2007).

2.3.1 Bioaerosol components

The components and the species of bioaerosols at composting sites are summarised as follows:

1- Fungi

Fungi are eukaryotic organisms, and the filamentous growth as multicellular hyphae is the most important specification of the fungi (Prescott, Harley and Klein, 1996). Field fungi are mostly found in fresh green waste and living things such as *Cladosporium* spp., and *Alternaria* spp. (Swan et al., 2003).

Due to its ability to utilise recalcitrant substrates, they increase towards the end of the composting process. Many types of fungi can be found in composting such as *Penicillium* spp., *Eurotium* spp., and *Aspergillus* spp.. *Aspergillus* species are the most important bioaerosols that are released from composting facilities as they are known to be associated with health problem in humans.

Aspergillus fumigates is an essential organism in the composting process that can decompose cellulose and hemicellulose (Epstein, 1997).

Mycotoxins are poisonous secondary metabolites synthesised by certain types of fungi, and are considered non-culturable bioaerosols. Mycotoxins are mainly produced by *Aspergillus* and *Penicillium* species (Swan et al., 2003). Mycotoxins cause many toxic effects as well as death if human beings or animal consume polluted food. Mycotoxin can be produced in most plant products such as cereal, oilseed, rice and corn when they are contaminated. There are four types of mycotoxin can make health problems for a human which are aflatoxins, ochratoxins, zearalenone and trichothecenes (Sharma, 2010).

Another non-culturable bioaerosols component that can be emitted from the composting process is glucan. Glucans are polyglucose compounds present in the cell walls of fungi, some bacteria and plants. The main effect on human health by Glucans is the respiratory system because they can be a respiratory immunomodulatory agent (Swan et al., 2003).

2- Bacteria:

Bacteria are prokaryotic, single-celled organisms. They are present everywhere in the environment and have a basic function in the ecosystem (Prescott *et al.*, 1996). Mainly, bacteria can be classified into two major kinds which are Gram-positive bacteria and Gram-negative bacteria. This classification depends on the Gram stain to distinguish between two types of bacteria which differ in cell wall structure. Whereas gram-positive bacteria keep the gram-stain in their cell walls because they have teichoic acids which give the cell walls of gram-positive bacteria a negative charge. However, in Gram-negative bacteria, these acids are absent in the cell walls so that these bacteria cannot keep the gram stain in the cell walls (Sharma, 2010). Both types of bacteria can be found within bioaerosols that are emitted from the composting sites as thermophilic

and thermotolerant species which reproduce through the composting process (Pankhurst, 2010).

Gram-positive bacteria are usually present in the dust of animal origin and also are present in the dust of plant origins such as *Corynebacteria*, *Bacillus spp*, *Staphylococcus spp*, *Micrococcus spp* and *Streptococcus spp*. Whereas Gram-negative bacteria are present in the dust of plant origins such as *Pseudomonas spp.*, *Klebsiella spp*, *Pantoea agglomerans*, *Rahnella spp* and *Alcaligenes spp* (Swan et al., 2003). Actinomycetes are Gram-positive bacteria characterised as filamentous and spore-producing. Gram-positive bacteria are usually found in bioaerosols from animal origin, but actinomycetes are more commonly present in the soil and plant materials. Actinomycetes are essential organisms in the composting process due to their thermophilic nature and ability to break down cellulose and lignin (Swan et al., 2002; Swan et al., 2003).

Endotoxins are complex molecules present in the cell walls of Gram-negative bacteria that live on the plants and animals' surfaces. Endotoxin consists of protein, lipid as well as the main part which lipopolysaccharide (LPS) (Swan et al., 2003). Although all Gram-negative bacteria have LPS in the cell walls, LPS is not toxic unless it is released from the outer layer of the cell. The cell walls decay after bacteria death, and release the toxin, whereas some Gram-positive bacteria produce LPS during growth, and that will affect the host (Sharma, 2010).

2.3.2 Bioaerosol Health effects

Under prolonged or acute exposure conditions, bioaerosols have the potential to pose health risks to immune-compromised or vulnerable humans. Particularly, when high concentrations are emitted close to residences, schools, hospitals and other public facilities (Herr et al., 2003). Due to the microscopic size of bioaerosols, which vary from 0.02-100 microns in diameter, small bioaerosols components commonly less than 10 microns in size can reach

deeply into the respiratory system, and the hairs in a human nose cannot prevent these particles (Douglas, 2013). Bioaerosols may cause many types of health effects as summarised in table (2-1) as described by (Douglas, 2013).

Table 2-1 A summary of some of the health effects caused by bioaerosols as detailed in CORRAO et al. (2012), Domingo and Nadal (2008), Douwes et al. (2003), Harrison (2007), Hoppe et al. 2012; Lorenz (2004) Poulsen et al. (1995) Srikanth, Sudharsanam and Steinberg (2008), Swan et al. (2003), Ulla I. Ivens , Johnni Hansen , Niels O. Breum , Niels Ebbehøj , Morten Nielsen , Otto M. Poulsen , Helle Würtz (1997).

	Respiratory problems and diseases	Gastrointestinal problems	Skin problems
Health effects	Asthma Rhinitis Mucous membrane irritations Chronic bronchitis Tracheobronchitis Airflow obstructions Organic dust toxic syndrome Farmer's lung Sinusitis Aspergillosis	Diarrhoea Nausea	Skin rash Itching skin rash, Dermatitis, Dermatomycosis Pyoderma Eczema
Agents	Fungi Bacteria Actinomycetes Endotoxins Glucans Mycotoxins Peptidoglycans Microbial enzymes Plant Mammalian and invertebrate proteins	Gram-negative bacteria Endotoxin Fungi	Streptococci Enterobacteria Endotoxins

2.3.3 Introduction to monitoring methods

This section describes the monitoring methods of bioaerosols from composting facilities. In this project, filtration technique (IOM/SKC) was used as a traditional method and spectral intensity bioaerosol sensor (SIBS) as a novel technique. Therefore, only a brief description is provided for the other traditional sampling methods.

Many sampling methods, including those mentioned below, have been used to detect bioaerosols from composting facilities. The data resulted from these studies has been collected using different techniques, in different locations, under different conditions and enumerating different bioaerosol components (Douglas, 2013).

Impaction

The key impaction sampling method is the Andersen Sampler. The sampler contains Petri dishes with suitable culture media, which is later incubated in the laboratory. To collect culturable microorganisms, two or four single stages viable impactor Andersen samplers are used. A tripod or other suitable structure holds the Andersen sampler at a height between 1.5 and 1.8 m above the ground (Environment Agency, 2009).

A vacuum pump should be connected to the sampler by tubing of an appropriate length and internal diameter. Air is pumped through the sampler at constant flow rate of 28.3 l/min. The bioaerosols are sucked through the sampler directly onto the agar plates and therefore bioaerosols are impacted onto the solid agar surface in the Petri dishes loaded into the sampler. After sampling, the Petri dishes should be stored in a cool environment and then taken to laboratory for incubation. In addition, control Petri dishes are necessary for the sampling program (Environment Agency, 2009; Sharma, 2010). The Andersen sampler is recommended in the AfOR sampling protocol (2010; 2018) for measuring bioaerosols at composting facilities.

Impingement

The impingement method has many similarities with the impaction method except that they are collected into a liquid media (Environment Agency, 2009). The particle is removed from the air by inertial force. The most common type of impingement technique is AGI-30 all-glass imping sampler which has curved inlet pipe made to simulate the nasal passage, making the sampler beneficial for studying the respiratory infections potential of bioaerosols (Sharma, 2010). Glass impingers need to be sterilised before each use so that a single glass impinger cannot be used more than once per sampling campaign, which will increase overall cost (Environment Agency, 2009).

Deposition (Durham's gravitational sampler)

Gravity sampling, also known as settling plate or depositional sampling, is a semi-quantitative collection method. In this sampling method, airborne microorganisms are collected by gravitational settling onto an exposed agar plate or adhesive-coated solid surface as a glass slide. The simplest design for Durham's gravitational sampler was in the 1940s, which is still the most commonly used sampler even today in some places in the world. This simple device consists of two metal discs separated by three struts.

The microscope slide, coated with adhesive such as petroleum jelly, is placed on the horizontal support between two metal discs. The horizontal support for the slide is mounted in the centre of the lower disc. The upper disc acts as a rain shield. The sampler is used for qualitative or semi-quantitative analysis of the microflora of air (Sharma, 2010).

Filtration sampling technique (IOM/SKC)

The filtration sampling technique is recommended by the AfOR protocol (2010). This sampler is light, compact and easy to transport (Pankhurst, 2010). In this sampling type, a known air quantity pass through a measurement filter to collect suspended particles with a flow rate of 2 l/minute. Before sampling, filters, cassettes and heads should be sterilised.

After sampling, the filters are washed in a buffer solution to move the bioaerosols into solution that is then inoculated onto Petri dishes containing the culture media. These are then incubated at appropriate temperatures. Ideally, three filters (at least two) are used at every sampling point to collect culturable microorganisms (Environment Agency, 2009).

The height of the sampling head is 1.5 m above the ground. The sampling head is connected to the sampling apparatus using a pipe or hose. Sampling times should be at least 30 minutes on-site and 45 minutes off-site to collect microorganisms (Sharma, 2010). In addition to three filters, a minimum of two blanks are exposed from each sampling point (filters exposed without air pumps switched on). The resulting blank represents the number of colony forming units (CFU) entering the sampler simply by handling the filter during sampling (Environment Agency, 2009).

In this project, bioaerosols were collected using filtration technique (IOM/SKC) as a traditional sampling technique. The main reason this technique was used is that the filtration technique is recommended by the AfOR standardised protocol when measuring the concentration of bioaerosol at composting facilities (AfOR, 2010). This sampling technique can detect the high concentration of viable-culturable bioaerosols only; this concentration will be compared with the fluorescent particles concentration detected using the SIBS.

Furthermore, there were practical reasons for using this sampling technique such as the high portability and ease of handling with this technique, the sampling can be done for long or short periods, and sample replication is simple and relatively inexpensive. Additionally, this sampling technique can detect bacteria and fungi together as part of the same sample.

Another advantage of this sampling technique that it is suitable to use at high bioaerosol concentration such as close to the emission source, which means there is less risk of overloading.

This technique consists of the following kit:

- SKC Pump
- IOM cassettes
- IOM heads
- IOM filters (Polycarbonate (SKC) – 0.8 μm)
- Tube

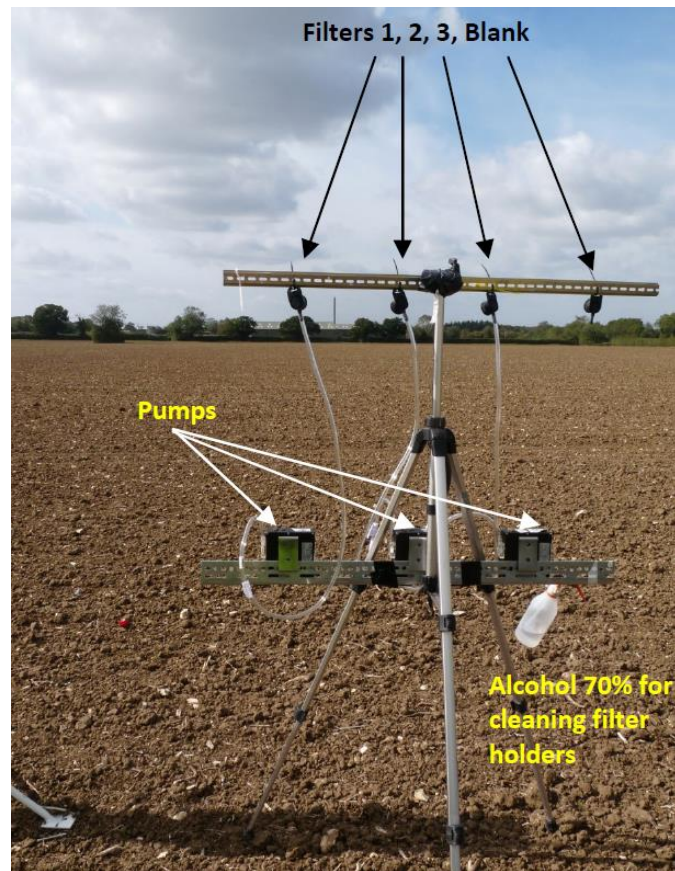


Figure 2-2 IOM/SKC Filter kit for bioaerosols sampling (R. Al-Ashaab and P. Douglas, 2011).

Spectral Intensity Bioaerosol Sensor (SIBS)

The Spectral Intensity Bioaerosol Sensor (SIBS) is the latest version of a series of devices to monitor biological particles, with the previous version known as Wide Integrated Bioaerosol Sensor (WIBS-4) (Healy et al., 2012). The

instrument is designed to detect bioaerosols by exploiting on-line detection of their autofluorescence (Kaye et al., 2004a).

The SIBS was developed by Droplet Measurement Technologies, USA. The device, originally developed by the University of Hertfordshire, is ideal for measuring mould, pollen, and fungi. Similarly to the WIBS-4, the SIBS provides highly sensitive measurements of mould and other bioaerosols, which include particle number concentrations; as well as particle size and particle shape (asymmetry) depending on spatial scattering analysis (Oconnor et al., 2014; SIBS, 2014; WIBS-4A, 2014).

The instrument uses UV xenon flashlamp sources to excite fluorescence in individual particles in two wavebands (280nm and 370nm) followed by measurement of fluorescence intensity in 16 wavelength bands. Unlike lasers, the UV xenon flashlamp sources allow for detection of common bioaerosol components such as tryptophan and nicotinamide adenine dinucleotide (NADH). These materials are usually present in the organisms' cells. Therefore, these fluorescent particles are assumed to be biological (SIBS, 2014; Z. Nasir and S. Tyrrel, 2017).

The main difference between WIBS and SIBS is the measurement technique of the emission fluorescence. The WIBS measures fluorescence in three emissions (λ_{em}) bands as follows: FL1: $\lambda_{ex} = 280 \text{ nm}$, $\lambda_{em} \sim 310\text{--}400 \text{ nm}$, FL2: $\lambda_{ex} = 280 \text{ nm}$, $\lambda_{em} \sim 420\text{--}650 \text{ nm}$, and FL3: $\lambda_{ex} = 370 \text{ nm}$, $\lambda_{em} \sim 420\text{--}650 \text{ nm}$. In the WIBS, the emissions are excited using two excitation wavelengths (280 nm and 370 nm).

The SIBS also uses UV xenon flashlamp sources to excite fluorescence in individual particles in two wavebands (280nm and 370nm), but the measurement of fluorescence intensity is in 16 wavelength bands from 288–735 nm, which is a unique feature of the SIBS (table 2-2). The xenon source is also far less expensive than a UV laser, making the SIBS a cost-effective alternative to other bioaerosol measurement instruments.

The fluorescence intensity for single particles is recorded by the individual channels. The biological aerosols can be discriminated from non-biological aerosols depending on the measured fluorescence data. Different analysis methods for this single data can provide more information about the nature of the bioaerosols (Toprak and Schnaiter, 2013a).

Figure 2-3 details the internal components of the SIBS. The SIBS employs a central optical chamber, around which are arranged the following components:

- A continuous-wave 785nm diode laser used in the detection of particles and the determination of particle size and shape
- A forward-scattering quadrant photomultiplier tube (PMT) used in the determination of particle size and shape
- Two pulsed xenon UV sources emitting at different wavebands
- A dual detector system comprised of an avalanche photodiode (APD) for particle detection and sizing, and a 16 channel spectrometer with individual photomultiplier channels for each optical channel. A dichroic mirror separates the light so that the scattered light, 785 nm, is reflected in the APD, and the fluorescence emission, 288-720 nm.

Table 2-2: Fluorescence Measurement Channels and Wavelength Ranges (SIBS, 2014).

Channel	Lower wavelength	Upper wavelength
1	298.2	316.4
2	316.4	344.8
3	344.9	362.5
4	377.5	401.5
5	401.5	429.7
6	430.2	457.5
7	456.7	485.6
8	486.0	514.0
9	514.1	542.0
10	542.0	569.8
11	569.9	597.6
12	597.6	625.2
13	625.3	652.8
14	652.8	680.2
15	680.3	707.5
16	707.5	734.7

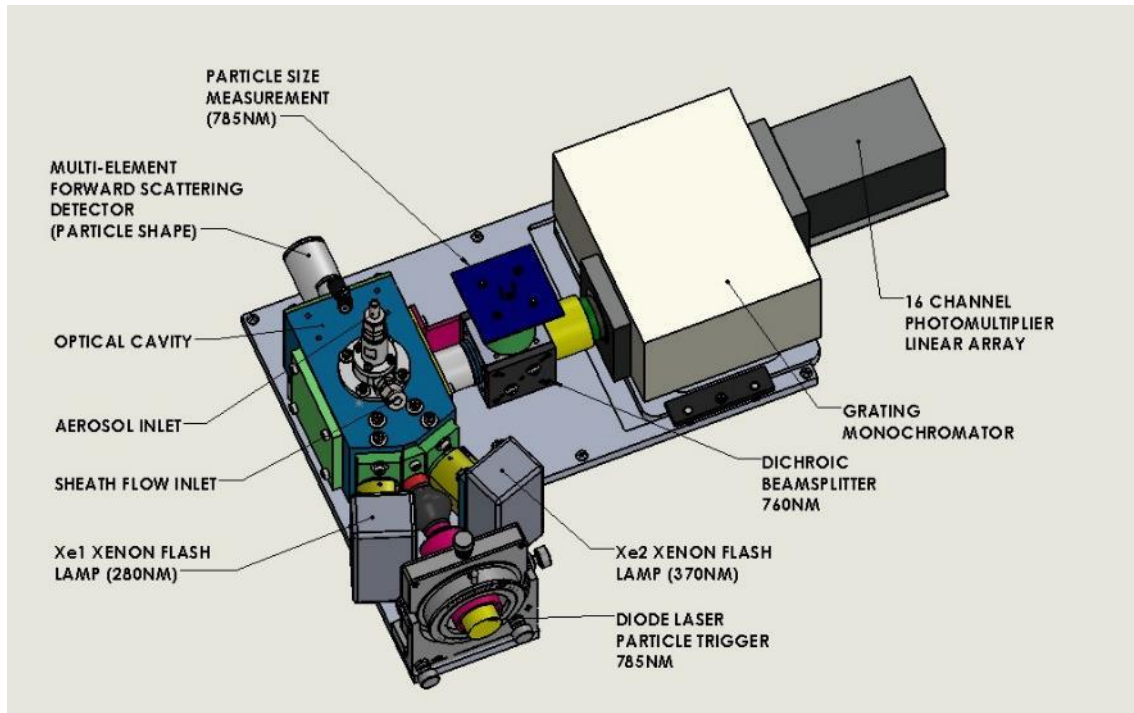


Figure 2-3: Diagram of the SIBS Optical Components(SIBS, 2014).

Thus, for each particle, a 2 x 16 excitation-emission matrix is recorded along with an estimate of particle size and particle shape. Table (2-3) summarises the advantaged and disadvantages of both sampling techniques used in this project where the advantages of both sampling techniques show the main reasons for choosing these techniques. In this study, the SIBS has a sample flow rate of 0.3 l/min and derives the equivalent optical diameter (EOD) and asphericity, in size range from 0.4–7 μm , along with the excitation-emission matrix of single particles.

Table 2-3 Advantages and disadvantages of the SIBS and filtration technique.

Sampling technique	Advantage	Disadvantage
SIBS	<ul style="list-style-type: none"> • Potential for continuous data for a long time. • Data available immediately • Detailed data such as particles size, shape and (number) concentration 	<ul style="list-style-type: none"> • Expensive • Needs electricity power during operation • Needs intense care during transportation and operation • Advanced data analysis techniques required
IOM/SKC	<ul style="list-style-type: none"> • Simple procedure • Low cost • Highly portable. • Less risk of overloading at high concentration (in comparison to direct impactor) • Different bioaerosol types sampled simultaneously 	<ul style="list-style-type: none"> • Snapshot data • Detects viable bioaerosol only • Contamination may affect the result • Needs preparing before the operation • Sampling processing is relatively long • High lower limit of detection (LLOD)

2.3.4 Analysis of current monitoring methods

Although the traditional sampling methods can detect the species of microorganisms emitted from the composting activities, they are still limited and not suitable for other purposes such as a real-time detection and detecting non-viable and non-culturable bioaerosols. As non-viable bioaerosols still have harmful effects, methods that can detect a wide range of bioaerosols are needed (Taha et al., 2007). Current data provides only spatial and temporal snapshots of bioaerosol emissions. Therefore, these data are still not sufficient to fully understand and model bioaerosol emission from composting facilities.

Previous studies have detected bioaerosols at different environments based on fluorescent particles using the WIBS-4 (Forde et al., 2018; Healy et al., 2012; Oconnor et al., 2015; Oconnor et al., 2014; Toprak and Schnaiter, 2013). Toprak and Schnaiter (2013) investigated the sensitivity of the WIBS to biological and non-biological aerosols and detected the biological particles in the ambient aerosol. Although, Toprak and Schnaiter (2013) showed a strong seasonal and diurnal variation in the concentrations of fluorescent particles, these concentrations represent the averages concentration only.

(O'Connor et al., 2013) collected data on the bioaerosol types released from hay and silage a combination of the WIBS-4 bioaerosol sensor and impaction/optical microscopy. (Oconnor et al., 2014) also used the WIBS to detect and quantify pollen released at a rural site in Ireland and an urbanised location in Germany.

In context with bioaerosol from composting sites, Connor, Daly and Sodeau (2015) used the WIBS to detect the fluorescent particles from a green waste composting site for several days and nights and during different seasons. They showed the variation of fluorescent particles concentration from the composting site and the relationship with the wind speed and direction. However, the agitation activities were not mentioned, so that the data showed only general trends.

Recently, the SIBS were also tested to detect bioaerosols at different environments (Nasir et al., 2018, 2019; Nasir and Tyrrel 2017). Nasir et al., (2018) used the SIBS along Aerojet Glass Cyclone, Eight-Stage Non-Viable Andersen Sampler and Six-Stage Viable Andersen Sampler to study the characterisation of bioaerosols emissions from compost in a controlled chamber. Nasir et al., (2018) provide information on comparisons of physico-chemical and biological characteristics of bioaerosols emissions measured using various techniques. (Nasir et al., 2019) detect the variability of the fluorescent particles concentrations at five contrasting outdoor environments which included agricultural site, agricultural farm, sewage treatment works, dairy farm and composting site. However, the SIBS data have not been used yet to model the dispersion of bioaerosols from composting facilities.

2.4 Air Dispersion Modelling

2.4.1 Introduction

The prediction of bioaerosol concentrations close to sensitive receptors and downwind of a composting facility will improve composting risk assessments. Although such estimates are currently provided by bioaerosol sampling, the use of air dispersion models has the potential to be a very useful and cost-effective way of exploring different bioaerosol control situations and assessing bioaerosol emissions in a composting site

Bioaerosol dispersion could be affected by the particle size, the emission rate, buoyancy effects, atmospheric effects and local topography (Drew et al., 2006). The dispersion can also be affected by turbulence in the atmospheric boundary layer, which is random by nature and therefore cannot be exactly predicted. Certain factors can introduce doubts in the model results such as errors in the input data, model physics, and numerical representation, and because of the effects of these uncertainties, the dispersion model cannot be perfect (Chang and Hanna, 2004).

Modelling of several bioaerosol components has been performed in many different environments. For instance, Ulleryl et al. (2012) have modelled the dispersion of *Legionella* species from cooling towers, and Wallensten et al. (2010) have modelled the dispersion of *Coxiella* from sheep farms.

Ulleryl et al. (2012) have studied the outbreak of Legionnaire's Disease which was caused by *Legionella pneumophila*, and Wallensten et al. (2010) have studied the outbreak of Q fever which was caused by *Coxiella burnetii*. The purpose of dispersion modelling within these studies was to apportion a source to a particular pollutant problem, not to accurately predict bioaerosol concentrations from a source at a particular location.

Therefore, using modelling in this way is not useful to estimate bioaerosols concentration downwind of an open windrow from composting facilities. Effective bioaerosols modelling from the open windrow composting process must be found to facilitate determination of the relationship between exposure levels and harmful health impacts. Despite the previous studies of bioaerosols dispersion modelling from open windrow composting sites, this subject still needs further improvements to be efficient to study and identify the relationship between exposure levels and the onset of negative health impacts.

2.4.2 Types of dispersion modelling

One of the essential elements of an effective dispersion modelling study is to select a suitable tool to match the scale of impact and complexity of a particular discharge. There are several kinds of air dispersion model including Box, Gaussian, Lagrangian, Eulerian, and Computational Fluid Dynamics (CFD). These models differ significantly in their capabilities and limitations (Holmes and Morawska 2006; Sharma et al., 2004).

The Gaussian models are considered to be well suited for pollutant dispersion with the ability to account for the random nature of atmosphere turbulence (Sharma et al., 2004). Gaussian models are favourite for researchers,

particularly when modelling odour emissions (Douglas, 2013), and have been the traditional and optimal selection of the models to simulate bioaerosol dispersion from composting sites (Drew et al., 2007). Although most Gaussian models are interested only in the diffusion and advection of the pollutants, more advanced Gaussian models include physical processes such as deposition and fast chemical reactions. (Holmes and Morawska, 2006).

Additionally, in very low wind speed, some Gaussian models cannot exactly evaluate pollutant concentrations (Qian and Venkatram, 2011). However, the modern Gaussian models, which use methods to describe diffusion and dispersion in the atmosphere, are now obtainable. So the effects of building, low wind speed and complex terrain can be modelled (Bluett et al., 2004).

Gaussian plume models assume that pollutant emissions normally distribute horizontally and vertically which produce a plume of polluted air that is approximately cone-shaped, with the apex of the cone towards the emission source (Beychok, 1994; Lines et al., 1997). The normal distribution of the plume is modified at greater distances due to the effects of turbulent reflection from the surface of the earth and at the boundary layer when the mixing height is low.

This is described mathematically in equation (2-1):

$$\chi_{(x,y,z,H)} = \frac{Q}{2\pi\sigma_y\sigma_zU} \exp\left[-\frac{y^2}{2\sigma_y^2}\right] \left\{ \exp\left[-\frac{(z-H)^2}{2\sigma_z^2}\right] + \exp\left[-\frac{(z+H)^2}{2\sigma_z^2}\right] \right\}$$

Equation 2-1

Where:

χ is the pollutant concentration at point (x, y, z, H) (g/m³)

x is the downwind distance from the source (m)

y is the lateral distance from the source (m)

z is the vertical distance above the ground (m)

H is the effective source height above the ground (m)

Q is the pollutant emission rate (g/s)

σ_z is the plume dispersion parameter in the vertical direction (m)

σ_y is the plume dispersion parameter in the horizontal direction (m)

U is the wind speed (m/s)

(Douglas, 2013)

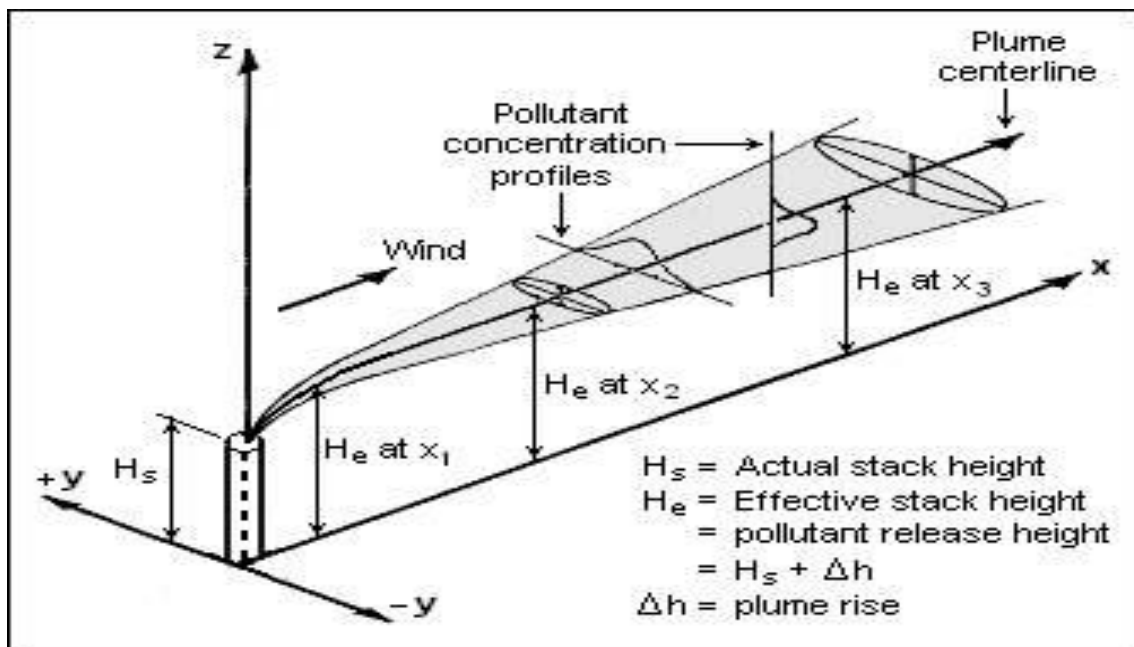


Figure 2-4 A typical visualisation of buoyant Gaussian pollutant dispersion plume from a point source (Beychok, 2007).

Figure 2-4 shows that the plume expands laterally and vertically due to the effects of turbulence when moves away from the source, the pollutants in the plume become more diluted. The rate of plume dispersal depends on many factors including source geometry, pollutant temperature, pollutant exit velocity and meteorological conditions.

There are many Gaussian models available, including ADMS, CALPUFF, AERMOD, ISC/ISCST3 and AUSPLUME, which are the most common (Douglas, 2013).

2.4.3 ADMS description

The Atmospheric Dispersion Modelling System (ADMS) used in this project is a new generation of dispersion model (CERC, 2016; Chang and Hanna, 2004; Riddle et al., 2004). Horizontally and vertically distribution of the plume concentration in ADMS is Gaussian during neutral and stable conditions, but during unstable conditions, the vertical distribution of plume concentration is non-Gaussian (Riddle et al., 2004).

ADMS does have some disadvantages. In particular, ADMS is unable to model the very low wind speed (below 0.6 m/s). In addition, ADMS may has significant errors in the calculation of boundary layer parameters when modelling emission near the pole or the equator (CERC, 2012).

2.4.4 Model input

2.4.4.1 Pollutant emission rate

The pollutant emission rate is a significant model input, which represents the amount and rate of emitted material from the source of the release (Barratt 2001). Emission rates that were used in previous studies differ by many orders of magnitude depending on the source type modelled (Douglas 2013).

The emission rates that were estimated in previous studies used measurements were taken at various distances downwind of the emission sources, as summarised below:

- 0.3 metres (Douglas 2013).
- 0 – 250 metres (Ji Ping Shi Hanah Hodson, 2012)
- As near as possible and 30 metres (Drew et al., 2007)
- 2-10 metres (Drew et al., 2007)
- 5-10 metres (Taha et al. 2006)
- 10 metres (Taha and Pollard 2004)
- 15 metres (Tamer Vestlund, 2009)
- 10-100 metres (Wheeler et al. 2001)
- 150 metres (Danneberg et al., 1997)
- Not detailed (Millner *et al.*, 1980)

There are two basic methods have been used to calculate emission rates from composting activities: calculation and back-calculation (back-extrapolation). Emission rate estimated using the calculation method are based on the measurements taken directly from the emission source to a maximum of 2 meters from the source, as the emission plume dilutes after release.

Pollutants concentration is inversely proportional to the distance from the source (Beychok, 1994). Direct measurements were possible to measure the emissions from static windrows using a portable tunnel (Drew et al., 2007; M. Taha, 2005; Taha et al., 2007, 2006; Tamer Vestlund, 2009). However, this is not possible with the agitation activities (Taha et al., 2007, 2006).

Previously, emission rate calculation has been done by rearranging model equations as used by Millner et al.,(1980) and Danneberg *et al.* (1997), depending on the model of Pasquill and the Giebels' formula respectively. The equation used by Taha et al., (2007; 2005; 2006) and Drew et al., (2007) to calculate fugitive bioaerosol emissions in a wind tunnel adapted from Shi and

Hodson (2010), was modified by Douglas (2013) to calculate bioaerosol emission rates from agitation activities.

Although Douglas (2013) measured the bioaerosol concentration at the source (0.3 meters from the source) using a novel sampling strategy, this concentration represents the viable bioaerosol only as the sampler was the traditional, culture based method (IOM/SKC). Due to the importance of the non-viable bioaerosols, it is necessary to use a method that can detect a wide range of bioaerosols (Taha et al., 2007).

On the contrary, the back-calculation method is based on the emission measurements taken 2 meters or more downwind of the composting activities (Danneberg et al., 1997; Environment agency, 2001; Taha et al., 2006, 2007; Drew et al., 2007). The back-calculation method includes adapting the emission rate model parameter until the modelled outputs resemble some measured concentration data (Environment Agency, 2001; Danneberg et al., 1997; Taha et al., 2006, 2007; Tamer Vestlund, 2009).

Emission rates were calculated using back-calculation by Shi and Hodson (2010) for point source scenarios for different sites, utilising datasets collected from different distances. Point sources were used because there were no details with the data about the nature of the composting activities. Therefore, point source was assumed for all emission sources.

Whatever the method used to calculate the emission rate, whether the calculation or the back calculation, the bioaerosol measurements are the essential factor to estimate the emission rates. The distance between the emission source and the sampler determines what type of calculation methods should be used (Douglas, 2013). Emission rate estimations from back-calculation can be completed in very short timescales, and inexpensively, whereas the method used by Douglas (2013) requires additional sampling and time as well.

It was not possible yet to measure the fluorescent particles at the source using the SIBS as done by Douglas (2013) using the filtration sampling technique. That was because of the dangerous conditions such as the movement of the heavy machinery around the compost windrows, high noise level and the high level of the emissions which make it difficult to place the SIBS close to the source and monitor it during the sampling time.

2.4.4.2 Source geometry

Source geometry is one of the most important factors that can affect the rate of plume dispersion (Beychok, 1994). It is not easy to estimate the dimensions of emission release at open windrow composting facilities throughout agitation activities as emission are not controlled or contained (Douglas, 2013; Millner, Bassett and Marsh, 1980; Taha et al., 2006).

Due to the dangers that were discussed above, direct measurement to the agitation area is not possible, so that personal observations were used to estimate and justify the source dimensions (Douglas, 2013; Millner et al., 1980; Taha et al., 2006).

ADMS model provides several types of source types:

a- Point source:

A point source is circular with a specific height, diameter and the coordinates of its centre (CERC, 2012). It is used to represent pollutant stack as the dimensions of the stack are easy to measure or well defined already (Drew et al., 2007).

To model the bioaerosol concentration from composting facilities, point source was used to represent different composting activities including turning, screening and shredding (Danneberg et al., 1997; Drew et al., 2007b; Millner et al., 1980; Taha et al., 2006, 2007).

b- Area source:

This source kind has a specific width, length, height and coordinates of three or more vertices (CERC, 2012). An area source is used to

represent the emission dispersion upon a horizontal plane at a particular height. The emission dispersion is supposed to be homogenous through the area and mostly used to model pollutants dispersion from sewage tanks or landfill, where the dimensions of the emission source can be measured.

c- Line source:

This kind of source is used to represent a homogenised release over a straight line at the same source height. The line has a specific width, height and coordinates (CERC, 2012). The model is designed to simulate the pollutant dispersion from mobile source to support the assessment of human exposures in near-roadway environments where a significant portion of the population spends time (Snyder et al., 2013).

d- Volume source:

This type is used to represent fugitive emissions from buildings and has been used with building at in-vessel facilities (CERC, 2016), where emission dimensions were based on building dimensions (Drew et al., 2007a). This type cannot be used in open windrow composting facilities because emissions are not confined to buildings and released in the open air.

e- Jet source:

This type represents the releasing in both vertical and horizontal component which is incompatible with open windrow composting sites (Douglas, 2013; CERC, 2016).

2.4.4.3 Pollutant exit velocity

Knowledge of the exit velocity of bioaerosol emissions from the agitation activities in the composting facilities is still limited. Pollutant exit velocities, such as 0.19, 0.2 and 0.5- 1.7 metres per second, were used as an exit velocity in the bioaerosol dispersion modelling by Drew et al., (2005), Taha et al., (2007) and Tamer Vestlund (2009) respectively. These pollutant exit velocities were

used without any clarification how these figures were calculated, measured or estimated.

Many studies did not state what exit velocity was used in the bioaerosol dispersion modelling from composting facilities (Douglas 2013). Miller et al., (1980) used the model of Pasquill to estimate emission rates by equation rearrangement, but this model needs an ambient wind speed and not a pollutant exit velocity. Douglas (2013) estimated the pollutant exit velocity by using the camcorder to measure the movement of the pollutant plume releasing during agitation activities. After observing the video, the following equation was used to estimate a pollutant exit velocity:

$$V=DT$$

Equation 2-2

Where:

V is the pollutant exit velocity in metres per second

D is the distance travelled by the pollutant plume in metres

T is the time taken by the pollutant plume to travel distance D in seconds.

The result shows that the estimated pollutant exit velocity ranged between 0.3 to 3 metres per second which were lower than the measured ambient wind speed (Douglas, 2013).

2.4.4.4 Meteorological inputs

One of the most significant inputs into any air dispersion modelling is meteorological data, as meteorology is a primary factor affecting the dilution of the pollutants in the atmosphere. There are two meteorological elements control the ground-level concentrations of contaminants which are wind speed and direction (for transport), and turbulence and mixing height of the lower boundary layer (for dispersion) (Bluett et al., 2004).

The portable weather stations are easy and safe methods to measure the meteorological parameters. In theory, accurate meteorological data must be available in all modelling studies. Only a few studies have stated what meteorological conditions were used in bioaerosol modelling (Douglas, 2013).

Ambient wind speeds range from 0.1 to 8.9 metres per second are the most commonly reported model inputs in the most studies of bioaerosols modelling (Drew et al., 2007a; Millner et al., 1980; Taha and Pollard, 2004; Taha et al., 2006). Oke (1992) mentioned that when wind speed increases from 2 to 6 metres per second, the dispersion will be quicker and pollutant concentration downwind will be lower. This is because there will be a larger volume of air passing the emission source per unit time.

Many studies did not report the modelled ambient temperature. Taha et al., (2006) and Drew et al., (2005) have reported the ambient temperature of 16.3 to 19.3 and 17.5 °C respectively. The magnitude of plume rise and dispersion are affected by the ambient temperature (Douglas, 2013).

2.5 Knowledge gap

Although there have been many studies done to improve the risk assessment of bioaerosols from composting facilities (Douglas, 2013; Douglas et al., 2016, 2017; Drew et al., 2007; Nasir et al., 2019; Taha and Pollard, 2004; Taha et al., 2006; Nasir and Tyrrel, 2017), further improvement is still required. Some studies focused on the direct detection of bioaerosol from the composting facilities, and others focused on dispersion modelling of bioaerosol from the same source. In both cases, the bioaerosol data detected in the direct measurement or the data used in the modelling were depended on traditional sampling techniques.

These techniques provide limited details about the bioaerosol emitted from the composting facilities. This is because the traditional data provide spatial and temporal snapshots of bioaerosol emissions concentrations, and these concentrations represented only particular components of bioaerosol. As

mentioned above, these sampling techniques can detect the viable-culturable bioaerosols only, however non-viable bioaerosols may still have harmful effects.

The SIBS has not been used to sample bioaerosols from composting facilities, and has not been compared with the traditional sampling methods. The analytical comparison shows how each sampler represents the bioaerosols concentrations, and the sensitivity of each sampler to the bioaerosols. These data may be useful not only as direct monitoring of bioaerosols but in dispersion modelling studies. However, the SIBS data is not currently used for bioaerosol dispersion modelling. Therefore, the using the SIBS data for dispersion modelling of bioaerosol emissions from open windrow composting facilities will facilitate and improve this process.

3 Bioaerosol sampling methods

3.1 Introduction

This chapter describes the general methods that were used in this project to sample bioaerosol concentrations from different environments. In addition to the sample collection, processing and results calculations, this description included two sampling techniques which are the IOM/SKC sampler as a traditional technique and the SIBS as a novel technique.

Bioaerosol sampling has been undertaken in three different environments. The most important environment was the composting facility as the aim of this thesis is to improve the sampling and dispersion modelling of bioaerosol from composting facilities. Before the sampling campaigns at the composting site, two sampling experiments were completed in an enclosed chamber and in a mouldy room.

The purpose of these experiments is to understand how the sampling techniques measure bioaerosols concentration in the air under the same conditions, as well as how each technique compares to the other.

3.2 Experimental methods

3.2.1 Sampling protocol

Pre-sampling laboratory practices

Before any sampling campaign, the filtration sampling technique kit, particularly the filter cassette, filter head and head, were sterilised in the autoclave at 121 °C for 15 minutes. The pumps flow rate was calibrated, according to Universal Sample Pump (2012), by using the small adjusting screw (FLOW ADJ) on the front of the pumps. After switching on the pump, this adjusting screw was turning using a small screwdriver either clockwise or anticlockwise which should increase or decrease the flow calibrator until the float is on the red line at 2.2 l/minute on the Rotameter as shown in figure 3-1.

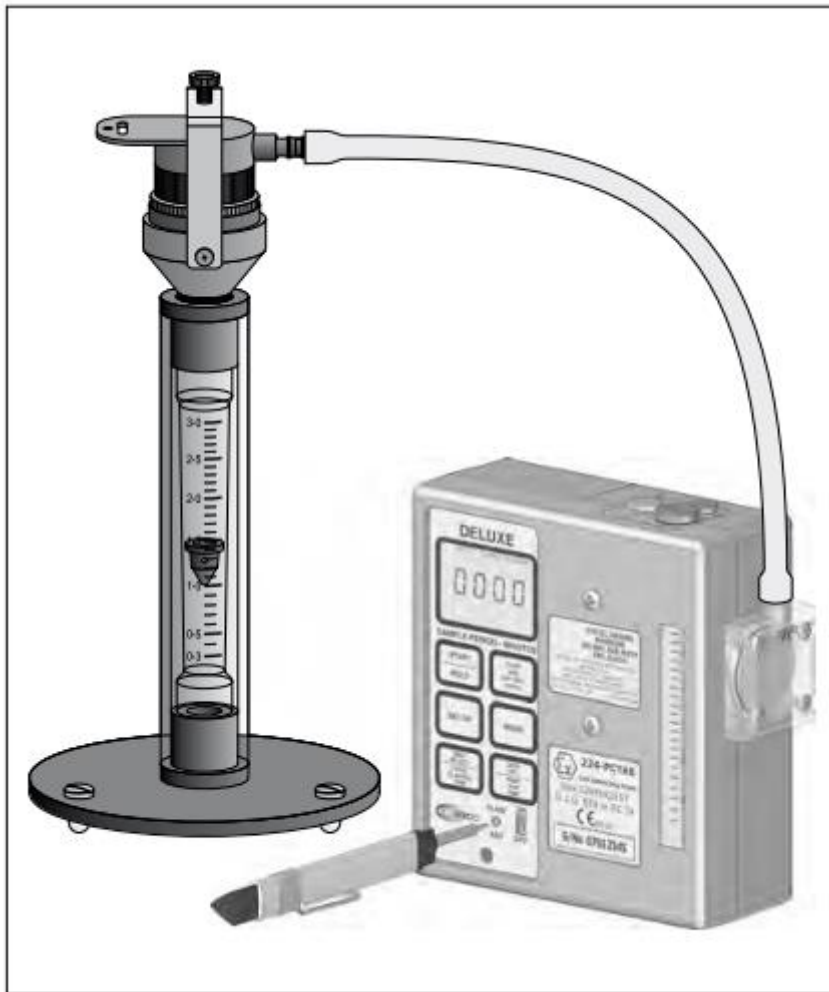


Figure 3-1: Flow rate calibration of SKC pump as mentioned in Universal Sample Pump, (2012).

Filters were loaded according to the manufacturer's instructions. The shinier side of the filter is more suitable for bioaerosol sample collection which contains a proper surface for microscope analysis (filters were handled with tweezers only) (AfOR, 2010; Environment Agency, 2018).

The buffer solution was prepared by adding 1 gram NaCl, and 3 drops Tween 80 per 1 litre of distilled water. After well mixing, the resulted solution was sterilised by autoclave at 121 °C for 15 minutes. 10 ml sterilised buffer solution was transferred into each Calvin tube under aseptic conditions to use them for the sampling process.

The microorganisms sampled in this study were bacteria and fungi as the aim of the sampling using the filtration technique is to measure the viable-culturable bioaerosol. Malt extract agar was used to culture the fungal samples. Preparing this culture media was by dissolving the powder in distilled water (50 gm per 1000 ml). To prevent bacterial growth 9.62 mg penicillin G-toxic and 42.33 mg streptomycin sulphate-toxic were added to every 1000 ml after autoclaving and leave the media liquid to cool down because this toxics material can be affected by high temperature.

For bacterial bioaerosol samples, nutrient agar was used. This culture media was prepared by dissolving the powder in distilled water (28 g per 1000 ml of distilled water). Cyclohexamide-toxic was added to prevent the fungal growth (11.9 mg per 1000 ml) (R. Al-Ashaab and P. Douglas, 2011).

Finally, after the dissolving, treating and sterilising, 15-20 ml of each agar was poured into separate Petri dishes and left to solidify in a laminar-flow safety cabinet for 30 minutes with half open lids. Then, the agar plates were kept in the original plastic bag and stored at 4 °C for use after sampling.

Quality control

Laboratory work was done according to standard techniques to minimise microbiological contamination. This involved sterilisation of all sampling kit (IOM/SKC heads, cassettes and filters) and laboratory equipment (buffer solution, pipette tips). All these items were autoclaved at 121 °C for 15 min (TL AC-EV, ProirClave, UK) to ensure sterilisation, which is according to standard autoclaving procedures.

70% ethanol solution was used to sterilise any surface used during laboratory work. All kit was handled within a laminar-flow cabinet (SC 18-R, Labcaire system, UK) to keep them sterilised and avoid any cross-contamination from the laboratory environment and between other samples during the work.

Additionally, hands were always sterilised with 70% ethanol solution during the laboratory and sampling work. Furthermore, blank samples were periodically

taken to ensure sample quality. The purpose of blank samples was to monitor any background contamination from the sampling environment. The contamination might result from the sampling kit or hands when changing the filters if they were not well sterilised.

Sampling campaigns

Three kinds of sampling experiments were done in this project including three sampling visits to the Ramsey composting site; two sampling repeats in the mouldy room and sampling in Porton down chamber.

Sampling at Ramsey composting site

AWO Recycling Limited, known as Ramsey, is located in Cambridgeshire near Ramsey Heights. The site is considered to be a rural location. The residents of Ramsey Heights, Upwood and Ramsey, are the nearest sensitive receptors about 1-2 kilometres South East, South and east of the site. The site is small and only processes green garden waste. The operation time on this site is between 0730 and 1730 hours Monday to Friday, and 0800 and 1200 hours on a Saturday. The site is closed on a Sunday (AWO Recycling Services, 2008).

Three sampling visits have been done at the same composting site at different times. The repetitions of bioaerosols sampling provides diverse bioaerosol samples from the same composting site, but in a different time, which means under different weather conditions and with different composting activities. The sampling visits were done in March, May and October 2016 respectively.

The exact sampling location depended on two important factors; wind direction and the nature of the place around the composting site. During all the sampling visits, the sampling locations were as close as possible to the downwind boundary of the composting site because the pollutants in the plume become more diluted as the plume expands laterally and vertically due to the effects of turbulence (Beychok, 2007). The three sampling locations for the three sampling visits were in three different places (illustrated in the chapter 5). These

particular locations were downwind of the site, about 40-50 m from the site boundary; southeast, northwest and southwest respectively.

The sampling time in every sampling visit was four hours continuously. The time for each IOM/SKC sample was 30 min, which means there were eight sampling periods per sampling campaign. Longer sampling times should be avoided as this can cause micro-organisms to dry out and lose viability as well as to prevent the filters from being overloaded (AfOR, 2010). In those four hours, the SIBS worked continuously, but the IOM/SKC filters were changed every 30 min. For each sampling period (30 minutes), three IOM/SKC were run concurrently; therefore, the total number of filters was 24 filters plus two filters (one blank and one handling blank).

Mouldy room sampling

Two sampling repeats were done in the mouldy room which is located in building 178, Cranfield campus. This room is old, and unused room, and obvious mould is visible on the walls and ceilings as a result of the high level of humidity (Figure 3-2). Both repeats were done using the same procedure, same sampling time and same sampling techniques but on two different days. These sampling experiments have been done as a calibrating experiment for the SIBS as it was a novel biosensor with limited available results. The sampling methods for the IOM/SKC technique of these two sampling repeats were exactly as used in the sampling visits to the Ramsey composting sites. This included sampling time, sampling technique, preparation before sampling, samples processing and result calculation.



Figure 3-2 Mould room which is the environment of the first sampling experiments in this project. The two samplers were placed in the table and worked for four hours continuously.

Porton Down sampling

In this experiment, one IOM/SKC sampler was installed along with the SIBS inside a confined chamber. As the sampling was not continuous, there was no need to use two tripods for IOM/SKC samplers. The dimensions of this chamber were about 3 X 4 metres and 3 metres in height. In order to simulate bioaerosol emission from a composting site, the Glass Duran bottle was used to aerosolise compost materials as a bioaerosols source.

There were 14 sampling periods over three days. Each sampling period was 30 minutes with three filters connected to the pumps and one filter as blank. Five sampling periods were as a control sampling, where the chamber was supplied with ventilation fans to remove the air and all possible emissions from the room. In this way, the sampler sensitivity to the emissions was checked and this

ensured that emissions detected in the particular aerosolised sampling period were not from other sampling periods.

The control sampling was done by running the ventilation fans, switching off the Glass Duran bottle and then switching on the samplers. In contrast, the aerosol sampling was done by switching off the ventilation fans, switching on the Glass Duran bottle and then switching on the samplers. A small fan has been used to move the air of the room to homogenise the emissions through all parts of the room including the samplers.

Sample processing

The processing of the IOM/SKC samples included preparing the sample solutions, inoculation, incubation and colony enumeration. All IOM/SKC samples have been processed within 24 hours of sampling to minimise loss of viability of the microorganisms. All work was in aseptic conditions, depending on the method stated in the standard protocol (AfOR, 2010) as shown below:

- IOM/SKC filters were removed from the cassettes using tweezers sterilised in a flame. The filter was placed into 10 millilitres of sterile buffer solution and shaken for 15 minutes using a shaking machine.
- As the filters were used for only 30 minutes sampling, there was no need for dilution for all samples except some bacterial samples from Porton Down (samples: 2, 4, 9, 10, 12 and 14). The dilution of these samples was 10^{-1} , which was prepared by transferring 1 millilitre of the suspension to 9 millilitres of saline solution. After dilution, the suspension was shaken for 1 minute.
- 100 microliters of each sample were cultured onto four prepared media plates. L-shape spreaders were used to spread the suspension evenly over the surface of the agar.
- Both kinds of bioaerosols samples (bacteria and fungi) were incubated using two temperatures. Nutrient agar plates were incubated at 25°C and

37°C; two plates for each temperature. Malt extract agar plates were incubated at 25°C and 40°C; again, two plates for each temperature.

The plates were incubation for 2-4 days, and checked for growth daily. Plates were counted immediately when growth was visible (Figure 3-3). According to standard laboratory protocols, 37°C and 40°C are the incubation temperature for bacteria and fungi respectively. In addition, 25°C was used for both bacteria and fungi to increase the chance of growth chance of bioaerosol les suited to standard temperatures.

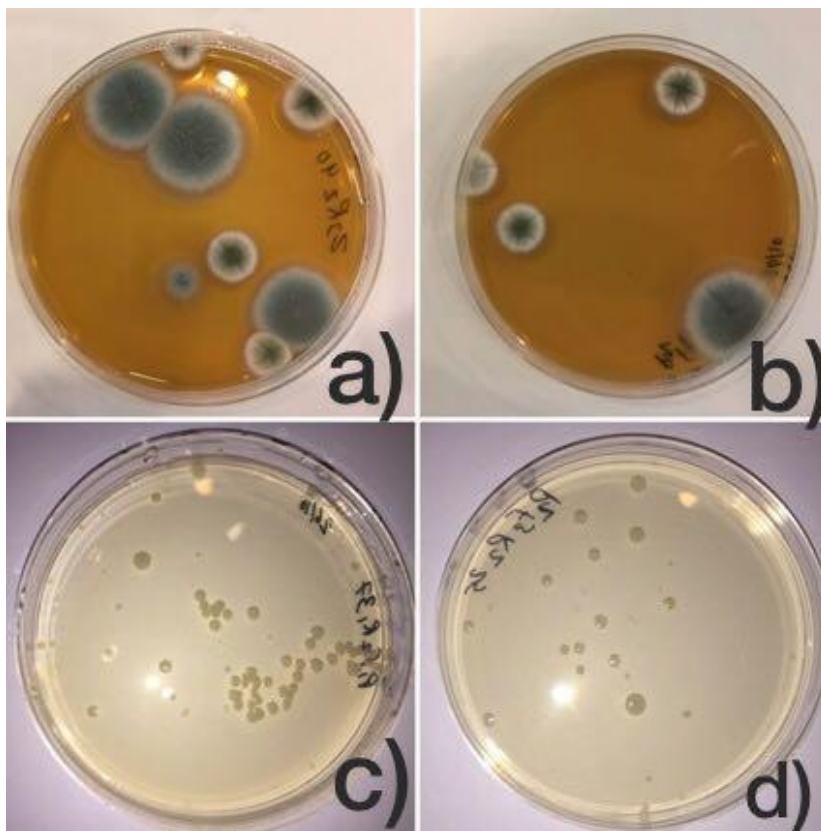


Figure 3-3: Examples of microbial growth of third visit samples: a- Fungal growth in 40°C (Malt extract agar), b- Fungal growth in 25°C (Malt extract agar), c- Bacterial growth in 37°C (Nutrient Agar), d- Bacterial growth in 25°C (Nutrient Agar).

3.3 Calculation of results

3.3.1 IOM/SKC results

Equation 3-1 was used to calculate the bioaerosol concentration present in the air at the time of sampling.

$$C = \frac{\left(\frac{n}{I}\right)V}{(FD)/1000} \quad \text{Equation 3-1}$$

Where:

C is the bioaerosol concentration in air (CFU/m³)

n is the average number of colonies retained across replicates.

I is the volume of inoculum plated (ml). This was equal to 0.1 throughout the project

V is the volume of the buffer solution in the original vial (ml). This was equal to 10 throughout this project

F is the sampling flow rate (L/min). This was equal to 2 throughout the project

D is the sampling duration (minutes). This was equal to 30 through the project

As samples were replicated on-site using three filters for each sample and in the laboratory using two plates for each incubation temperature, mean averages of the concentrations calculated for each replicate were reported and used throughout the statistical analysis. Total bioaerosols were calculated for each sampling period using the highest concentration of each species of bioaerosols. Regardless of the incubation temperature, the highest bacterial concentration was added to the highest fungal concentration to produce a total concentration of viable bioaerosol detected using the filtration technique. While this does not represent a true total viable bioaerosol, this was done to provide the closest

approximation to the bioaerosol detected by the SIBS to facilitate comparison between the two methods.

As with all methods, the traditional sampling method used as part of this project is subject to limitations. The lower limit of detection (LLOD) is one of the main limitations of the filtration sampling technique. A LLOD can be defined as the lowest concentration of bioaerosol that can be detected by the sampling technique, thus any apparent 'zero' values may be between 0 and the LLOD minus 1. Any zero values can therefore be considered as the lower limit of detection (Douglas, 2013).

Pankhurst (2010) calculated the LLOD by assuming the growth of one colony averaged across all replicates. This assumption can be applied in Equation 3-2 using the mean average number of colonies. Therefore, the LLOD in this study has been calculated as below:

$$\frac{\left(\frac{0.5}{0.1}\right)10}{(2 \times 30)/1000} = 833.33 \text{ CFU/m}^3$$

Equation 3-2

Where

0.5 is average of one colony across two plates retained

0.1 is the volume of inoculum

10 accounts for sample volume

2 is the flow rate

30 is the sampling time

This concentration does not represent the lowest concentration detected using the IOM/SKC because the lowest concentration is calculated using three filters,

whereas the LLOD is calculated using one filter only. The LLOD calculated in this study is presented in Table 3-1 with the LLODs from previous studies.

Table 3-1: The low limit of detection calculated in this study and in previous studies including the differences in the plate replicates, sampling time and flow rates.

Studies	No. of replicate	Sampling time	Flow rate	Resulted LLOD
This study	2	30	2	833
Douglas (2013)	3	2	2	1388
Douglas (2013)	6	2	2	695
Pankhurst (2010)	2	30	2.2	757

3.3.2 SIBS data processing and calculation

The steps required to process the SIBS data are summarised in Figure 3-4. The SIBS stores single particle data directly during sampling. These data are imported into a data analysis toolkit for offline data processing. The single particle data files were analysed by choosing an averaging interval of 60 s from 0.5–0.7 µm. As the flash lamps of the SIBS needs time to recharge, no fluorescent measurements can be taken while the lamps are recharging. Therefore, SIBS data includes three kinds of particles, namely total particles (particles interred the sampler), excited particles (flashed particles) and fluorescent particles. This means there are particles that may be fluorescent, but they were not excited because of the recharge time for the flash lamps.

To calculate the correct fluorescent concentration, equation 3-3 was used:

$$\text{Fluorescent concentration (cm}^{-3}\text{)} = (F/E) T \quad \text{Equation 3-3}$$

Where:

T= Total particles (cm⁻³)

E= Excited particles (cm⁻³)

F= Fluorescent particles (cm⁻³) (Nasir et al., 2019).

To compare the SIBS data with the IOM/SKC data, 30 min averages of fluorescent particles were calculated. Although the SIBS data needs some calculation steps using the toolkit and then using Excel, SIBS data processing can be done in very short time in comparison with the time needed when processing IOM/SKC samples where 3-4 days are needed to obtain the final results. This advantage of the SIBS makes the sampling process easy and fast when sampling bioaerosol emissions from any environment.

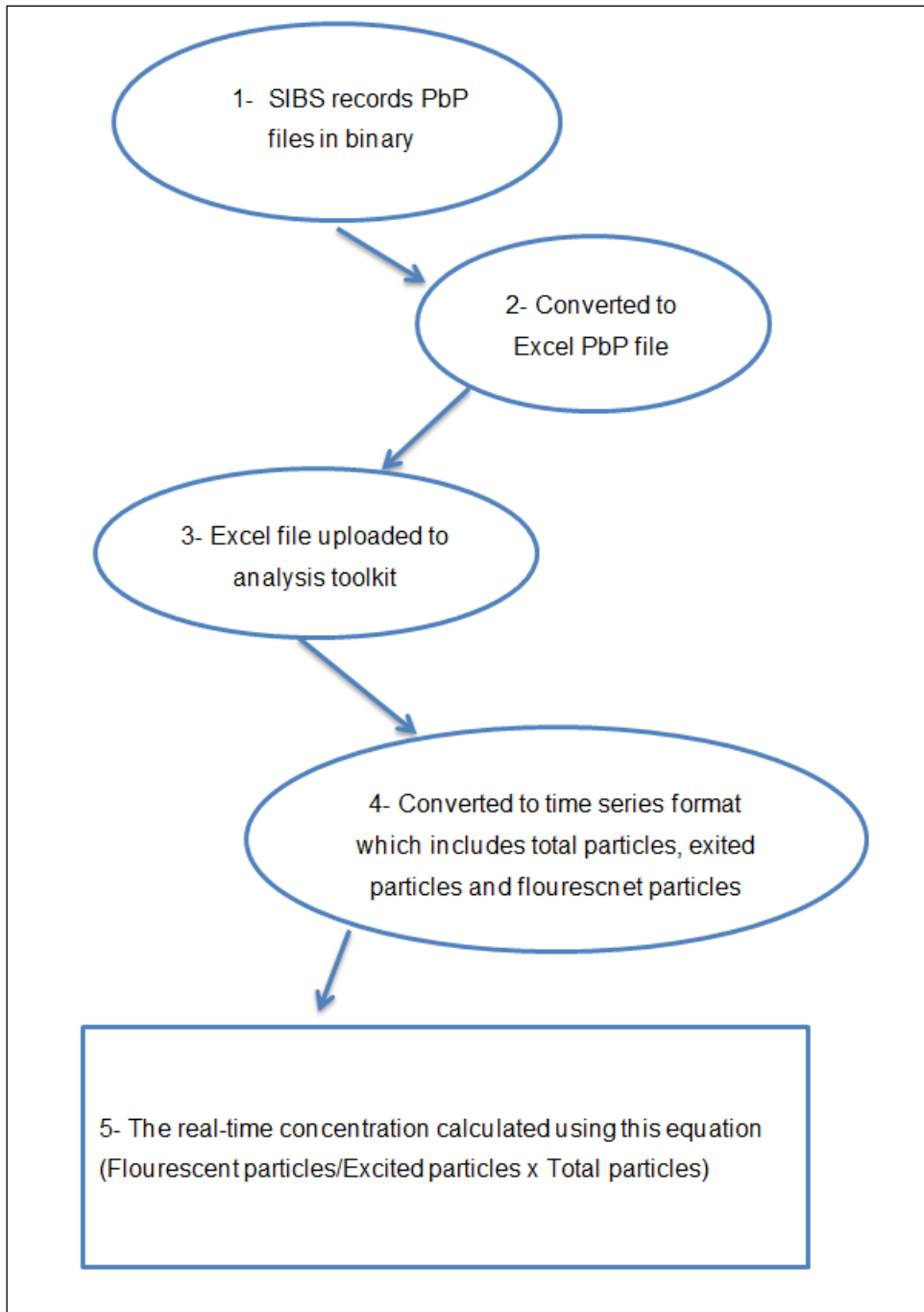


Figure 3-4 SIBS data calculation process.

4 Control environment sampling

4.1 Introduction

This chapter provides the results of bioaerosol sampling campaigns in the controlled environments using the novel sampling technique (the SIBS) alongside the traditional sampling technique (filtration technique). These, two sampling experiments were done in two different environments. The purpose of these sampling experiments was to collect data about bioaerosol concentrations in the same environment using the SIBS and the filtration sampling technique side by side. The objectives of this comparison was to:

- 1- Quantify and characterise the fluorescent particles detected using the SIBS.
- 2- Analyse how SIBS data relates to data collected using traditional methods as the present knowledge is based on traditional methods. This places the SIBS data within the context of previous findings.

The first sampling experiment was undertaken in the mouldy room; building 178- Cranfield campus. This room is old, and the unused room contains a lot of growth of microorganisms on the walls and ceilings as a result of the high humidity level (figure 4-1).

This room can be considered as a controlled environment, as it is closed and not affected by weather conditions. In addition, the amount of the microorganisms was static during the sampling time. This experiment was repeated twice. Both repeats were done using the same procedure, same sampling time and same sampling techniques but on two different days as detailed in chapter 3.



Figure 4-1: Two pictures for mouldy room explaining the growth of microorganisms on the walls and ceilings. This table shown in the left picture was used to install the two bioaerosol samplers.

The second experiment was done in the Porton Down the chamber. These samples were processed and used in the same way as other samples as detailed in Chapter 3 and below.

4.2 Method

This section describes the sampling method for each sampling experiments including sampling environment, sampling strategy and sampling time. The pre-sampling and post-sampling works have been described in details in chapter 3.

4.2.1 Mould room sampling experiments

To minimise time lost during changing of the filters, two IOM/SKC samplers were installed to facilitate the sampling process. Once the first sampler stopped after 30 minutes of sampling, the second sampler was ready and started

immediately. In this way, bioaerosol samples were taken using the filtration technique concurrently with the SIBS.

For both sampling techniques, the total sampling time was four hours. Therefore, the sampling time for the IOM/SKC sampler was divided into eight sampling periods, 30 minutes for each period. These eight periods have been combined to form four hours sampling as shown in the results below.

The culture media and incubation temperatures have been described in Chapter 3. However, due to a technical error in the 40°C incubator, all 40°C plates of both mouldy room sampling experiments were lost. The incubator temperature was increased by an unknown person on the second incubation day, which caused dehydration of the culture media and fusion of some Petri dishes. There was not enough time to repeat these two sampling practices as the SIBS was scheduled for Ramsey composting sampling campaigns and maintenance. Therefore, the results show the bacterial growth at 37°C and 25°C and fungal growth at 25°C only.

4.2.2 Porton down chamber experiment

In this sampling experiment, only one IOM/SKC tripod was used as the sampling periods were not continuous and there was a break between every two sampling periods (figure 4-2). The Glass Duran bottle was used to aerosolise compost materials as a bioaerosols source (figure 4-3).

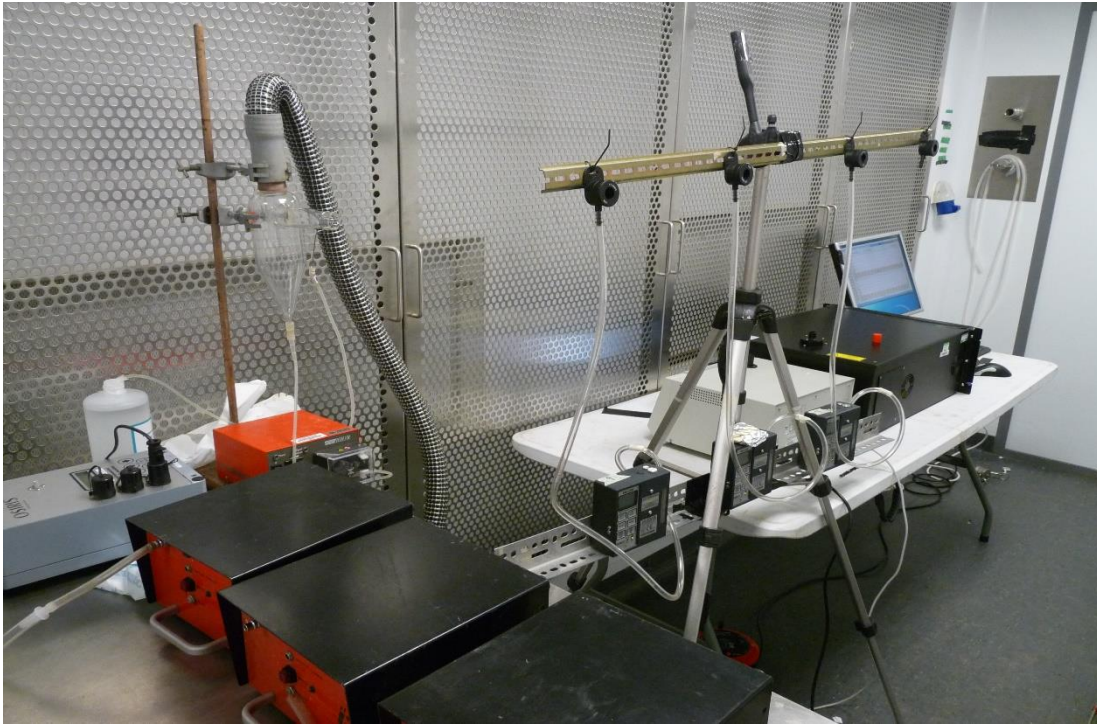


Figure 4-2: The sampling chamber including the SIBS and the filtration sampling technique with three IOM/SKC filters connected to the pumps plus one filter as blank.

The Porton Down chamber experiment included 14 sampling periods over three days. Each sampling period (30 minutes) was done using three filters connected to the pumps plus one filter as blank. Five sampling periods were considered control samples, where the chamber was supplied with ventilation fans to remove the air and all possible emissions from the room. In this way, the sampler sensitivity to the emissions was checked and to make sure that the emissions detected in the particular aerosolised sampling period are not from the other sampling periods.



Figure 4-3: The Glass Duran bottle and the mixing fan.

As the IOM/SKC filter needed to be transported back to the laboratory, each filter was inserted into 10 ml buffer solution after sampling and then kept in a fridge to maintain the viable microorganisms. The laboratory work (as described in chapter 3) was completed as soon as possible.

4.3 Results

4.3.1 Mould room samples

Figures (4-4 and 4-5) show the concentrations of bioaerosol emissions in the room during the sampling time of two repeats of the sampling experiment. The SIBS data are represented in two ways, the real-time data and the 30 minute average data. The real-time data, which is the red line, represents the concentration of fluorescent particles in minutes. The blue line represents the 30 minutes average concentrations, which were originally calculated from the 1-minute concentration. The purpose of using the 30 minute concentration is to

compare the fluorescent particles concentrations detected using the SIBS with bioaerosol concentration detected using the filtration technique.

The bioaerosols data, detected using the filtration technique, are represented by the green bars. Total bioaerosol concentrations have been calculated by collecting the highest concentration of bacteria and fungi; therefore, it is not a real total bioaerosol. Total bioaerosol should include all bioaerosol components including viable and non-viable. However, filtration sampling technique can only detect viable-culturable bioaerosol including bacteria and fungi which can grow in the selective media used in this project. Therefore, the total bioaerosol in this project represents viable-culturable bioaerosol only. Using total bioaerosol concentration in this way was for illustrative purposes, to compare the bioaerosol data collected using the filtration technique with the fluorescent particles data collected using the SIBS.

The 1 min average data of SIBS show the fluctuation in the bioaerosol concentration during the sampling time, which is the significant advantage of the SIBS sampler. The figures show the variation of emission concentration detected using the SIBS during the sampling time, which is not detected by the filtration sampling technique. Analysis of the real time data shows that the highest concentrations were always at the beginning of each 30 minutes. In this time there was always movement in the room due to the changing of the IOM/SKC filters every 30 minutes. This highlights the sensitivity of the SIBS, and how these emissions data directly relate to a source of emission. This feature is very useful when detecting the bioaerosol from composting facilities.

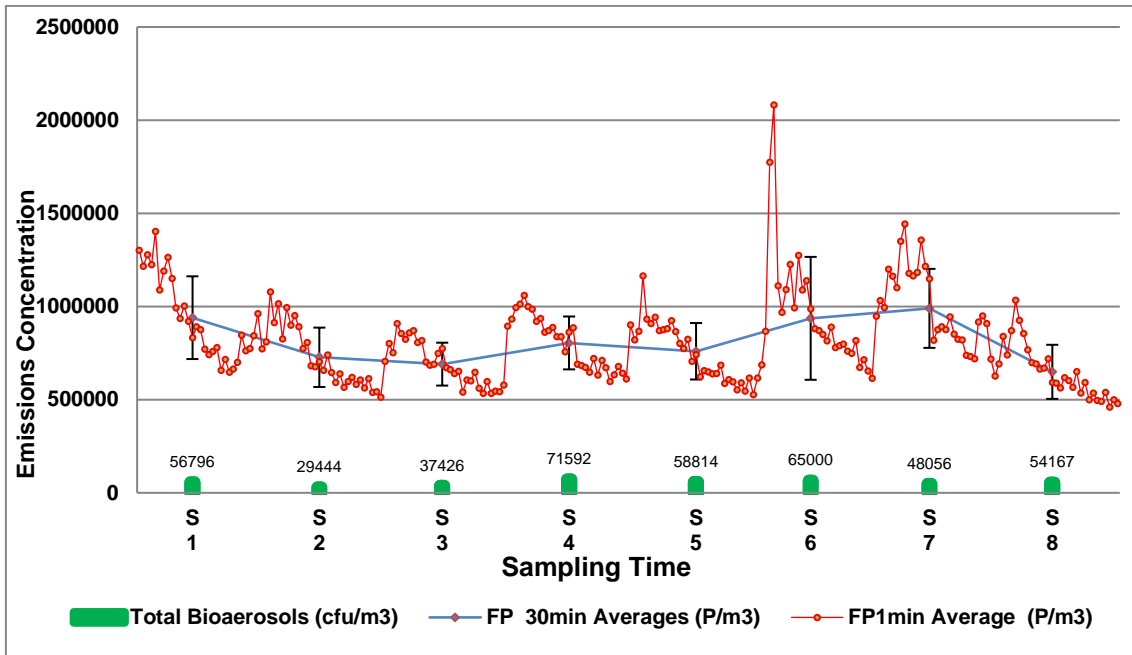


Figure 4-4: Fluorescent Particles and Total Bioaerosols concentrations from the Mouldy room/Building 178 measured using the SIBS and the IOM/SKC during the first sampling campaign in Feb-2016. The fluorescent particles data are presented in two ways; 1 min average (Red line) and 30 min average (blue line). Total bioaerosols (green bars) are the highest bacteria and the fungi at 25°C. There were 1 or 2 min gaps between the sampling durations for IOM/SKC filters changing (every 30 min). These gaps have been removed from the SIBS data as these gaps are not represented in the IOM/SKC data.

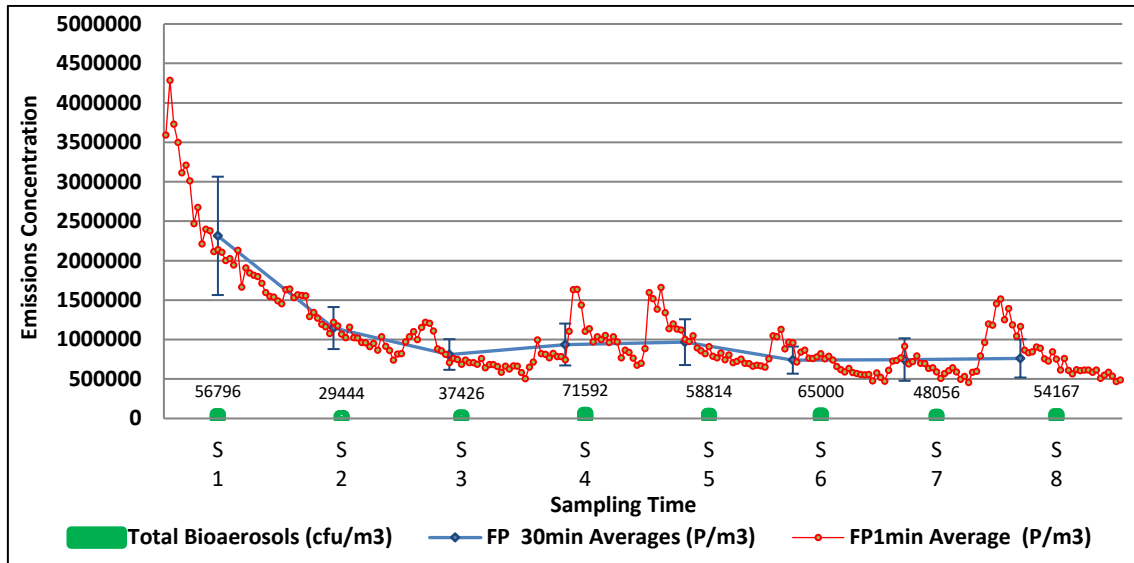


Figure 4-5: Fluorescent Particles and Total Bioaerosols concentrations from the Mouldy room/Building 178 measured using the SIBS and the IOM/SKC during the second sampling repeat in Feb-2016. The fluorescent particles data are presented in two ways; 1 min average (Red line) and 30 min average (blue line). Total bioaerosols (green bars) are the highest bacteria and the fungi at 25°C. There were 1 or 2 min gaps between the sampling durations for IOM/SKC filters changing (every 30 min). These gaps have been removed from the SIBS data as these gaps are not represented in the IOM/SKC data.

Generally, there is a considerable variation between the data of both samplers as well as between the IOM/SKC data itself. IOM/SKC data show that there is a big difference between the concentrations of fungi and bacteria (figure 4-6). Although the 40°C plates were lost, the growth of 25°C shows that most bioaerosols in this room were fungal with lower bacteria concentrations. In both sampling repeats, there were many nutrient plates with zero growth. On the contrary, there was always growth on all malt-extract plates (table 4-2).

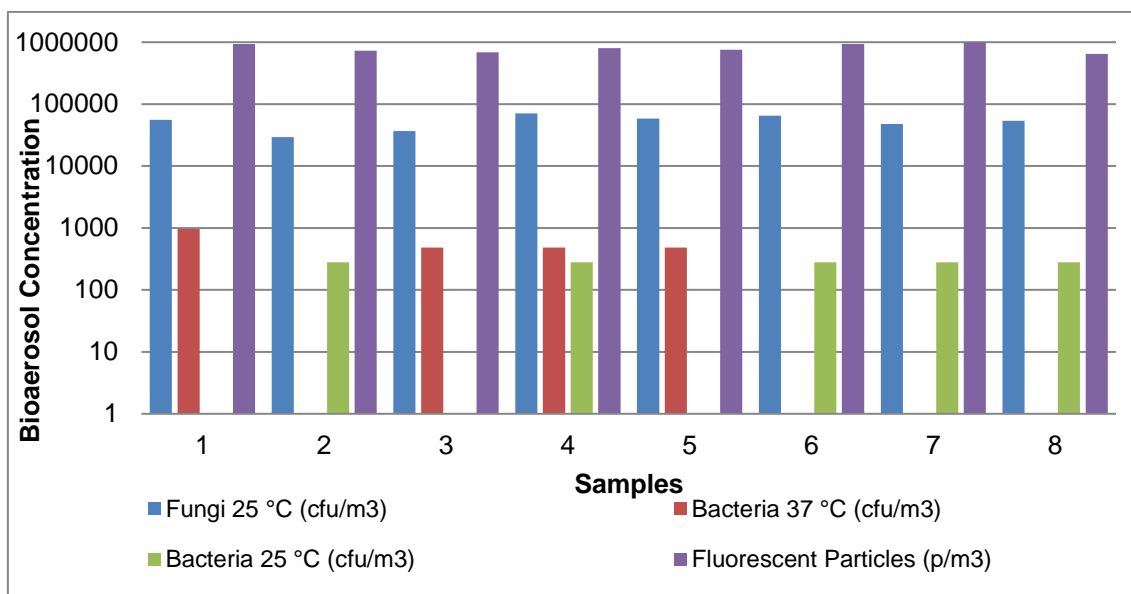


Figure 4-6: Bioaerosol concentration at the mouldy room detected during the first sampling repeat using the SIBS and filtration sampling technique. Fungi concentration was always higher than bacteria concentration in all samples during both sampling repeats.

In the first two hours of sampling time during the first sampling visit, the concentrations of fluorescent particles from the SIBS (461500, 357875, 349250 and 423917 p/m³) were lower than the concentration of bioaerosols in the second half of sampling time which were 422500, 525417, 582625 and 413208 p/m³.

There were some similarities in the concentration trends between SIBS and fungi data from the IOM/SKC, but with large differences in the level of the concentration. The fungal bioaerosol concentrations in the first two sampling hours were also lower than the second two sampling hours.

When comparing the concentration of fluorescent particles (SIBS data) and the bioaerosol concentration (IOM/SKC data), it is worth mentioning, that there are several orders of magnitude difference between IOM/SKC data and SIBS data. This difference is because the SIBS data represents the fluorescent particles concentration which is likely to include viable and non-viable bioaerosols, where the IOM/SKC data represents the viable-culturable bioaerosols only.

However, this scenario was different in the second sampling repeat. In this sampling repeat, bioaerosol movement was agitated by moving objects on the ground or by scraping the walls using a piece of iron. Therefore, all concentration averages of the second sampling visit were higher than those of first sampling visit (table 4-2).

Table 4-1: The statistics of the two sampling visits to the mouldy room/Building 178. SIBS data (FP/m³) including 4 hours sampling average, standard deviation, median, maximum and minimum of 1 min average, and maximum and minimum of 30 min average.

Sampling	4 hours Average	Standard Deviation	Median	Average Time	Maximum	Minimum
First Visit	812190	226923	774859	1 min Average	2082137	459549
				30 min Average	989884	649291
Second Visit	1052991	604367	845733	1 min Average	4286037	243456
				30 min Average	2314444	455147

Table 4-2: The statistics of the two sampling visits in the Mouldy room/Building 178. IOM/SKC data (CFU/m³) including 4 hour sampling average, an average of 30 min sampling, standard deviation, median and maximum and minimum of 30 min sampling.

Sampling Repeat	Kind of bioaerosol sample	4 hours Sample	Standard Deviation	Median	Maximum	Minimum
1st Visit	Bacteria25°C	174	144	278	278	0
	Bacteria 37°C	301	358	241	962	0
	Fungi 25°C	52222	13892	54861	71111	29167
	Total Bioaerosols	52662	13940	55481	71592	29444
2nd Visit	Bacteria25°C	833	727	556	2500	278
	Bacteria 37°C	1563	741	1528	2778	556
	Fungi 25°C	55313	21135	56528	82222	24167
	Total Bioaerosols	57049	21624	58750	83889	25556

4.3.2 Porton Down samples

Table (4-3) shows the conditions and information about the Porton Down sampling experiment. The experiment included 14 samples from three sampling days. First five samples were on the first day. These five samples included three aerosol samples and two control samples. No temperature or humidity recorded on this day.

In the second sampling day, seven samples were taken, two samples were as control samples, and five samples were aerosol samples. The temperatures of these samples were between 22.3 to 24.4 °C, where the humidity was between 46.3 to 58.9 %. There were no significant differences between the temperature and humidity through the sampling duration because the sampling environment was controlled.

Table 4-3: Information and Measurements of Porton samples including (Temperature, Humidity, Compost and kind of samples).

Samples	Sample type	Temperature (°C)	Humidity %	Age of compost	Compost Mass
S 1	Control	NA	NA	Old	
S 2	Aerosol	NA	NA	Old	100mg/m ₃
S 3	Control	NA	NA	Old	
S 4	Aerosol	NA	NA	Old	1000mg/m ³
S 5	Aerosol	NA	NA	Old	300 mg/m ³
S 6	Control	22.3	52.9	Fresh	
S 7	Control	23.7	59	Fresh	
S 8	Aerosol	24.3	46.3	Fresh	200 mg/m ³
S 9	Aerosol	23.3	46.6	Fresh	1300 mg/m ³
S 10	Aerosol	22.4	58.9	Fresh	1300 mg/m ³
S 11	Aerosol	23.2	59.6	Fresh	300 mg/m ³
S 12	Aerosol	23.7	51.3	Fresh	3000 mg/m ³
S 13	Control	22.7	68	Fresh	
S 14	Aerosol	23.5	58.5	Fresh	1200 mg/m ³

The last sampling day was for the last two samples only, one control sample and one aerosol sample. The temperatures were 22.7 and 23.5 °C, and the humidity was 68 and 58.5 respectively. All measurement readings have been taken in the first 10 minutes of each sampling period.

As shown in figure (4-7), the SIBS data has been presented as the average for each sampling period. There was no need to show the real-time data in this sampling experiment because the samples were not continuous. The IOM/SKC data included five forms of data; concentration of bacteria grow in two different temperatures (25 & 37 °C), and concentration of fungi also grow in two different

temperatures (25 & 40 °C). In addition, the total bioaerosol concentration which was calculated using the highest concentration of both bioaerosol types as explained before.

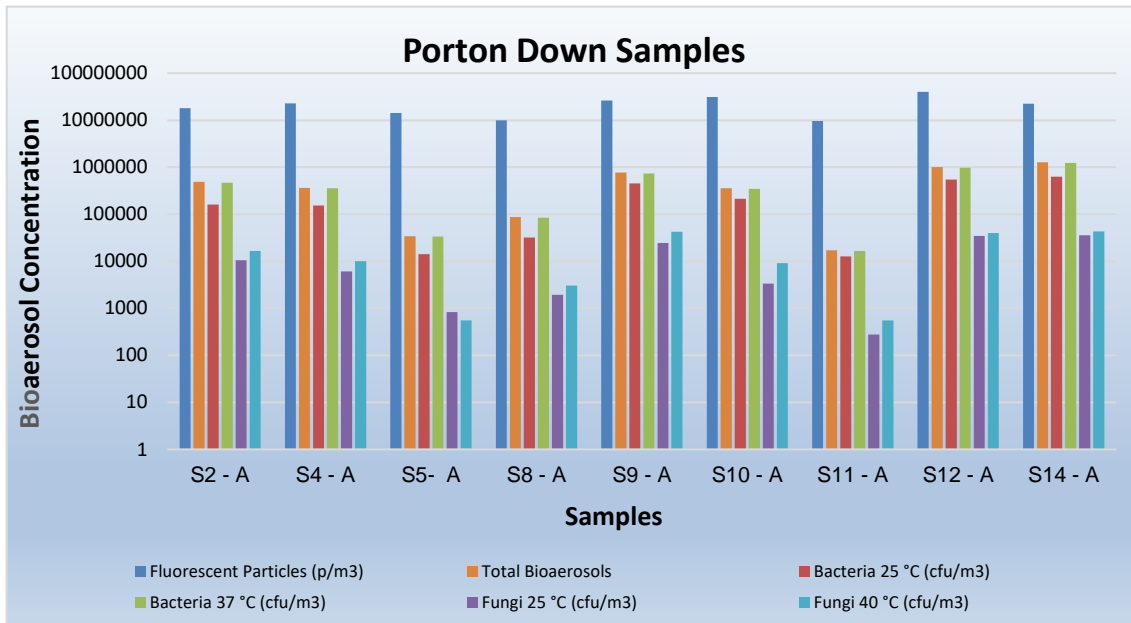


Figure 4-7: Fluorescent particles (SIBS) and bioaerosols (IOM/SKC) concentration during the aerosolised samples in the Porton Down chamber. Total bioaerosols are the highest bacteria and the highest fungi regardless of incubation temperature.

Conversely with the mould room bioaerosol samples, in all samples, the concentrations of bacteria were higher than the concentration of fungi (figure 4-7). This means that the bacterial content in compost material was higher than the fungal content. In all aerosol samples, the highest concentrations of bacteria and fungi for both temperatures (37 and 25°C for bacteria and 40 and 25°C for fungi) were in sample 14, and the lowest concentrations for both bacteria and fungi and both temperatures for each type were also in the same sample which was sample 11. The highest concentration of bacteria was in sample 14 which was 1233333 CFU/m³ for bacteria grow in 37°C, and 625000 CFU/m³ for bacteria grow in 25°C, but the lowest bacteria concentration was in sample 11

was 625000 CFU/m³ for bacteria grow in 37°C, and 12778 CFU/m³ for bacteria grow in 25 °C.

The general fungal growth of IOM/SKC samplers was low at both temperatures (40°C and 25°C) if compared to the bacterial growth. The concentrations of fungi that grew at 40°C were mostly higher than the concentration of fungi that grew at 25°C in all samples except sample five, which were 833 CFU/m³ for fungi at 25°C and 556 CFU/m³ for fungi at 40°C.

The highest concentrations of fungi were in sample 14, which was 43333 CFU/m³ for fungi at 40°C and 35556 CFU/m³ for fungi at 25°C. While the lowest concentrations were in sample 11 which were 556 CFU /m³ for fungi at 40°C, and 278 CFU /m³ for fungi at 25°C.

4.4 Discussion

4.4.1 Mouldy room

Direct comparison between IOM/SKC data and SIBS data is not possible as the IOM/SKC sampler measures the concentrations of culturable bioaerosols only, and gives the concentrations of bioaerosols as (CFU/m³), whereas the SIBS measures fluorescent particles concentrations as (particle/m³). Therefore, this comparison focuses on the general trends of the measured concentrations for both samplers, and how each sampler represents the concentrations of bioaerosols from the same source, at the same time, for same environment and same conditions as well.

As the sampling has undertaken in a closed environment, the only factor that can effect the bioaerosols concentrations during sampling is air movement. The air movement resulted from our activities during the sampling process such as monitoring the sampling devices and changing the filters of IOM/SKC sampler.

Table 4-4: The correlation coefficient between FP/m³ (SIBS data) and CFU/m³ (IOM/SKC data) during the two sampling repeats in the Mouldy room/Building 178. The SIBS data include the 30 min averages of every sampling repeat. The IOM/SKC data include four readings; Bacteria in two temperatures (25°C & 37°C), Fungi 25°C and total bioaerosols. Total bioaerosols are sum of the highest bacteria and the highest fungi regardless of incubation temperature.

SIBS with:	1st visit	2nd visit
Bacteria 25°C	0.097	-0.064
Bacteria 37°C	0.116	-0.621
Fungi 25°C	0.344	-0.339
Bioaerosols	0.343	-0.347

The correlation coefficient between the SIBS data and the IOM/SKC data in the table (4-4) indicates that there was no relationship between the fluorescent particles concentrations and viable bioaerosol concentrations. This may suggest that the ratio of viable bioaerosol in the sampling environment was very low, as the SIBS can detect all biological aerosolised particles, regardless of whether they are viable or not.

Table 4-5: Peak/mean ratios for SIBS data during the two sampling repeats in the Mouldy room/Building 178-Cranfield campus.

Samples	Peak/mean ratios	
	1 st sampling repeat	2 nd sampling repeat
1	1.49	1.85
2	1.48	1.43
3	1.32	1.50
4	1.32	1.75
5	1.53	1.72
6	2.22	1.53
7	1.46	2.03
8	1.59	1.83

Table (4-5) shows the peak/mean ratios of fluorescent particles concentration detected during the two sampling visits in the mouldy room using the SIBS. These ratios indicate the difference between the peak concentration and the mean concentration and shows the nature of bioaerosol dispersion. The snapshot data does not show the highest or lowest concentration of bioaerosol during the sampling time. The fluctuations of the concentrations detected using the SIBS shows these details and the concentrations can reach several orders above the mean concentration as in sample 6 in the first visit and sample 7 in the second visit.

The ability of SIBS to show peak emissions and fluctuations will be very useful to detect the variability of bioaerosol concentration dispersed from the composting sites during agitation activities, and to analyse the effects of the

agitation activities and weather conditions on the concentration and dispersion of bioaerosols.

4.4.2 Porton Down

Regardless of the difference in actual concentration of fluorescent particles (SIBS data) and bioaerosols (IOM/SKC data), similar general trends were identified. There was a correlation between both sampling techniques as shown in the table (4-6). Although the concentration of fluorescent particles was the highest from sample 12 and not from sample 14 as with the IOM/SKC data. Whereas the lowest concentration of both techniques was lowest from sample 11.

Several factors during sampling affected the concentration of emissions. The first factor was the mass of the compost material that was aerosolised during sampling using the Glass Duran bottle. For example, in the first three samples, i.e. S2, S4 and S5, the bottle of the Glass Duran was filled with 25 gm of old compost product; therefore the concentration was reduced with time.

Where the highest concentration of this three sampling durations was S2, then S4 and the lowest was S5 as the compost has been used, taking into account that the first and third samples were control samples. Another example is S12, which was higher than S11 as the container had been topped up with the compost after S11, and therefore more compost was aerosolised during S12 than during S11.

Another factor was the use of two piped air supplies in the container, which was used first time in sample 9. Therefore the concentrations of bioaerosol in the first sampling day (S-2, S-4 and S-5) were lower than the other samples such as S-9 and S-14. So that, using only one pipe air supply significantly affects the concentration.

Additionally, the distribution of compost material inside the bottle had an important effect on the aerosolising process. For example, the concentration in sample-8 was relatively low. The compost material was not well distributed inside the bottle as shown in figure (4-8), so the pipe could not suck the compost. Therefore less compost was aerosolised into the chamber, and lower concentrations were detected.



Figure 4-8: The Glass Durham bottle when the compost material was not close to the pipe (the right picture) and when the compost was close to the pipe (the left picture), and therefore the emissions concentration was affected by this factor.

Figure (4-9) shows the bioaerosol emission concentration during the five control samples. These data show the ability of the SIBS to detect the emission in the very clean environment as well as the contaminated environment. The lowest concentration of bioaerosol detected using the filtration technique was 278 CFU/m³ during the control samples where there was only one colony in 6 cultured plates. This detected concentration is the lower limit of detection (LLOD) of the IOM/SKC technique.

However, there was no fungal growth in all control samples, and no growth for both bacteria and fungi in sample one (table 4-6). Even though, the SIBS could detect fluorescent particles during these control samples, which indicates the high sensitivity of the SIBS.

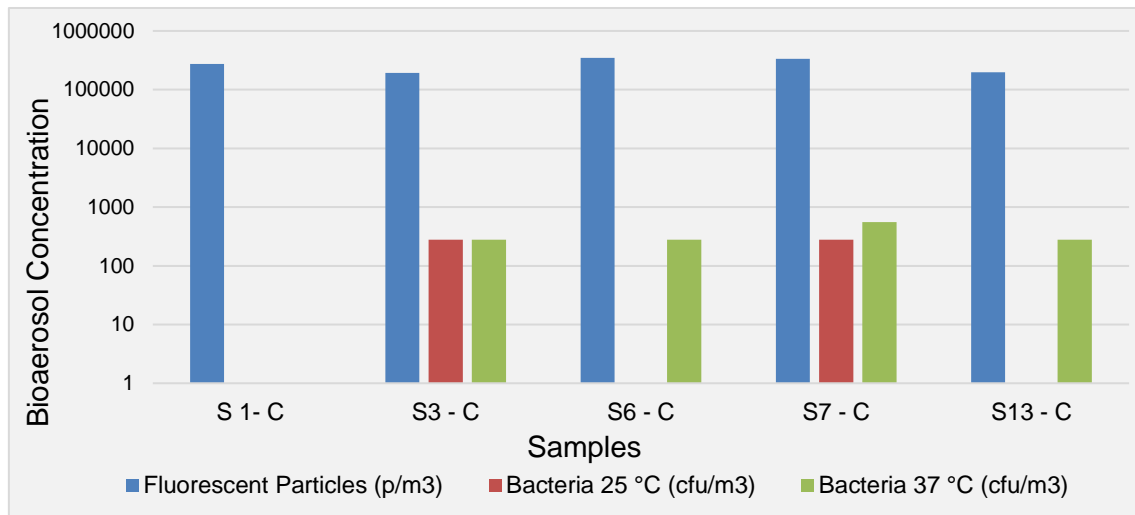


Figure 4-9: Fluorescent particles (SIBS) and bioaerosols (IOM/SKC) concentration during the control samples in the Porton down chamber. In all control samples, there was no fungal growth.

Figure (4-10) shows the concentrations of fluorescent particles during all samples including the aerosol and control samples. Not only are there clear differences in the concentration between the aerosol and control samples, there are also clear differences in the standard deviations of each concentration. The concentrations detected during the aerosol samples have a standard deviation higher than the concentrations detected during the control samples. This means the fluctuations during the aerosol samples were higher than the fluctuations during the control samples.

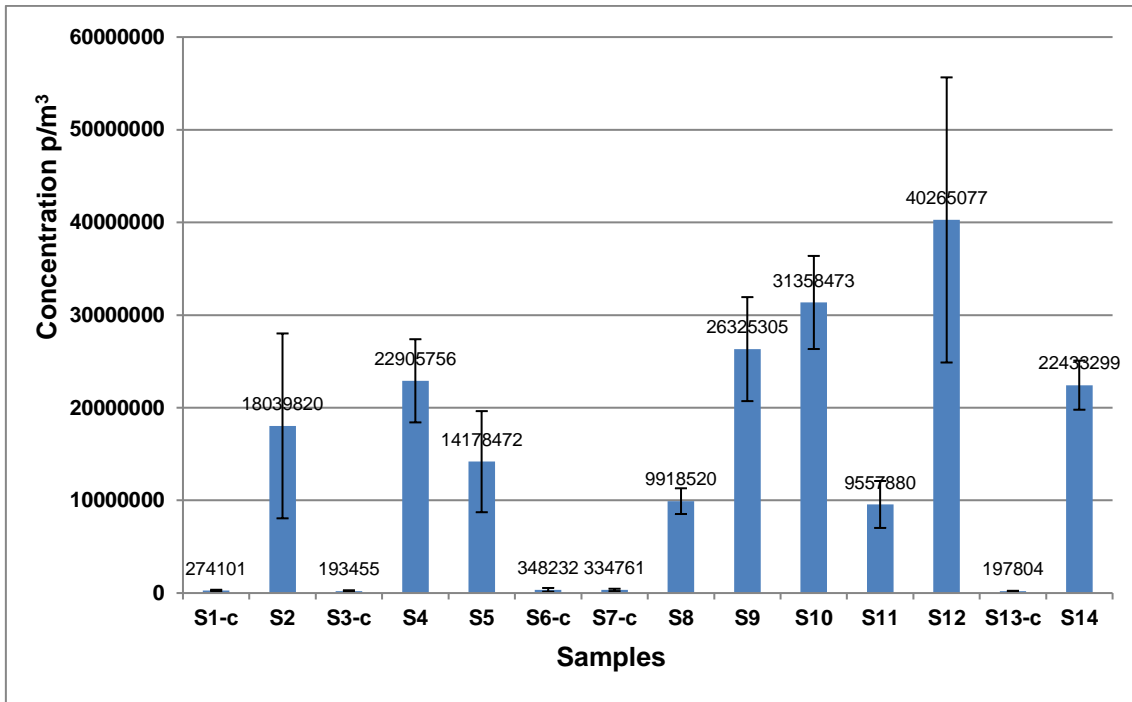


Figure 4-10: Fluorescent particles concentrations detected using the SIBS in the Porton down chamber. The samples included nine aerosol samples and five controls samples.

Previously, chamber bioaerosol sampling has been done using the WIBS but for another purpose such as characterising purposes. Healy *et al.*, (2012) used the WIBS to distinguish between natural airborne samples such as the fungal spores and detect and discriminate the toxic fungal spore, *Aspergillus fumigatus*, from others in real-time. Similarly, Hernandez *et al.*, (2016) also used the WIBS to investigate the bioaerosols classes using bacteria, fungi and pollen samples, which were aerosolised in an controlled environment chamber. Both Healy *et al.*, (2012) and Hernandez *et al.*, (2016) stated that this technique could be used as a very useful analytical tool distinguishing rapidly between the bioaerosol components. The results presented in this study are the first to present the SIBS output in a controlled environment and in comparison to a traditional technique.

Table (4-6) provides statistical data for both SIBS and IOM/SKC data for all Porton Down samples including aerosolised and control samples. Although there was a positive correlation coefficient between SIBS data and IOM/SKC data, there was an important difference between fluorescent particles concentration (SIBS) and bioaerosols concentration (IOM/SKC). The difference between both types of data was about two orders of magnitude, which was expected as the SIBS detects the fluorescent particles, whereas the IOM/SKC detects the viable-culturable bioaerosol only.

However, a key disadvantage of the SIBS is that it detects all fluorescent particle, and there are non-biological particles that may be fluorescent. It is therefore necessary to determine what emissions the SIBS does detect.

On the other hand, although the IOM/SKC detects the viable-culturable bioaerosol only, it is clear that this method is detected true bioaerosols. Table (4-6) also shows the high sensitivity of the SIBS and the high lower limit of detection of the IOM/SKC. The SIBS could detect the emissions during the control sampling period whereas the IOM/SKC could not, and most plates of the control samples had no grow in both types of growth media.

Table 4-3: The Statistics of SIBS and IOM/SKC data; including correlation coefficient, averages, medians, maximum and minimum. This table summarised all results of the Porton down sampling experiment which were detailed in the previous figures in the result section.

Samples	The correlation coefficient between the fluorescent particles (P/m ³) and the bioaerosol (CFU/m ³)	Average		Median		Maximum		Minimum	
		Aerosolised samples	Control samples	Aerosolised samples	Control samples	Aerosolised samples	Control samples	Aerosolised samples	Control samples
Fluorescent Particles/m ³ (SIBS)		2.17 x 10 ⁷	2.70 x 10 ⁵	2.24 x 10 ⁷	2.74 x 10 ⁵	4.03 x 10 ⁷	3.48 x 10 ⁵	9.56 x 10 ⁶	1.93 x 10 ⁵
Total Bioaerosols (cfu/m3)	0.784	4.90 x 10 ⁵	2.22 x 10 ²	3.66 x 10 ⁵	2.78 x 10 ²	1.28 x 10 ⁶	5.56 x 10 ²	1.72 x 10 ⁴	0
Bacteria 25 °C (cfu/m3)	0.801	2.45 x 10 ⁵	1.11 x 10 ²	1.61 x 10 ⁵	0	6.25 x 10 ⁵	2.78 x 10 ²	1.28 x 10 ⁴	0
Bacteria 37 °C (cfu/m3)	0.784	4.71 x 10 ⁵	2.78 x 10 ²	3.56 x 10 ⁵	2.78 x 10 ²	1.23 x 10 ⁶	5.56 x 10 ²	1.67 x 10 ⁴	0
Fungi 25 °C (cfu/m3)	0.731	1.31 x 10 ⁴	0	6.11 x 10 ³	0	3.56 x 10 ⁴	0	2.78 x 10 ²	0
Fungi 40 °C (cfu/m3)	0.758	1.84 x 10 ⁴	0	1.00 x 10 ⁴	0	4.33 x 10 ⁴	0	5.56 x 10 ²	0

4.5 Conclusion

Both sampling experiments were undertaken using the new biosensor (the SIBS) alongside the traditional sampling technique (IOM/SKC) in controlled environments for the first time. Both experiments showed how to use these sampling techniques, especially the SIBS as a novel technique. The results show how the SIBS could be used to monitor the bioaerosol emission from the composting facilities as the SIBS can detect the real-time dispersion of emissions continuously for long time periods.

These SIBS data is expected to show the relationship between bioaerosol concentration and agitation activities, as this ability was demonstrated during the mouldy room experiment. Real-time detection will demonstrate how the weather conditions affect the dispersion, and the variability in the bioaerosol emission concentration with the time.

The sensitivity of the SIBS will also contribute to discovering the impact of agitation activities including detecting the peaks and the lowest concentrations and the timings of these.

4.5.1 Key findings

In summary, the key findings from this chapter are:

- 1- The SIBS is more sensitive to low concentrations, where the filtration sampling technique has a high lower limit of detection. This is an important advantage of the SIBS, which enables the SIBS to detect bioaerosol emissions at different environments and different levels of emission concentrations.
- 2- The real-time detection provided new insights into bioaerosol concentration fluctuations with time, which was not available with the traditional data.

- 3- The concentration of fluorescent particles concentration (SIBS data) was always higher than bioaerosol concentration (IOM/SKC data) including fungi and bacteria. This indicates the present of non-viable bioaerosol that should be taken into account when studying bioaerosols because they still have harmful effects.

5 Field Sampling Campaigns

5.1 Introduction

Bioaerosols emitted from composting sites are released directly to the atmosphere, which can be potentially harmful to human health (Douwes et al., 2003). The knowledge about dispersal of bioaerosols from composting sites still needs further improvements to improve bioaerosol risk assessments and planning of the future composting facilities, to reduce the public exposure to bioaerosols. There are many techniques used to detect bioaerosols concentrations (as described in chapter 2). Although the traditional sampling methods can detect the species of microorganisms emitted from the source, it is still limited and not suitable for other purposes such as a real-time detection and detecting non-viable and non-culturable bioaerosols.

The filtration sampling method, as a traditional method, provides a snapshot of bioaerosols concentration emitted as a result of the compost agitation activities, but without details about how they disperse. Using snapshot sampling and methods that may reduce culturability of bioaerosol may produce an underestimation of bioaerosol concentrations. In other words, traditional sampling techniques can detect the total number of emitted bioaerosol during the sampling time and not the real time concentrations. In the calculation, the sampling time and the flow rate of pumped air are taken into account to calculate the average concentration of viable and culturable bioaerosols during the sampling time, which may be 30 or 45 minutes.

The Spectral Intensity Bioaerosol Sensor (SIBS) is a new version of WIBS-4, which can measure aerosols individually (Healy et al., 2012). The instrument is designed to detect bioaerosols by exploiting on-line detection of their autofluorescence (Kaye et al., 2004b).

This chapter aims to address several gaps in knowledge, including quantification of bioaerosol concentrations downwind from composting sites, analysis of the effect of operational and environmental influences on emission

and downwind concentrations, and compare the novel sampling method with the traditional sampling method.

Three sampling visits have been undertaken at Ramsey composting during March, May and October 2016. The purpose of these sampling campaigns was to collect data about bioaerosol concentrations at composting sites using the SIBS alongside the IOM/SKC. These data have been to compare the SIBS and the IOM/SKC.

The objectives of this comparison are:

- 1- Quantifying and characterising the fluorescent particles from the composting facilities using the SIBS.
- 2- Analyse how SIBS data relates to data collected using traditional methods as the present knowledge is based on traditional methods. This new analysis will help to place the SIBS data within the context of previous findings.

5.2 Field work-sampling

Three sampling visits have been done at the same composting site (Ramsey composting site), which was detailed in chapter 3, at different times. The repetitions of bioaerosols sampling provides opportunities to capture diversified bioaerosol samples from the same composting site, but at different times, which means under different weather conditions and with different composting activities (Figure 5-1).

The sampling location depended on two important factors; wind direction and the nature of the place around the composting site. During all the sampling visits, the sampling locations were as close as possible to the downwind boundary of the composting site because the pollutants in the plume become more diluted as the plume expands laterally and vertically due to the effects of turbulence (Beychok, 2007).

The three sampling locations for the three sampling visits were in three different places. As shown in the figure (5-2), the sampling locations were southeast, northwest and southwest respectively. These particular locations were downwind of the site, about 40-50 m from the site boundary.



Figure 5-1: Ramsey Composting site during the shredding activity. The agitation activities during the sampling visits were turning and screening, turning only, and shredding and screening during the first, second and third sampling visit respectively.

The sampling time in every sampling visit was four hours continuously. In those four hours, the SIBS worked continuously, but the IOM/SKC filters were changed every 30 min.

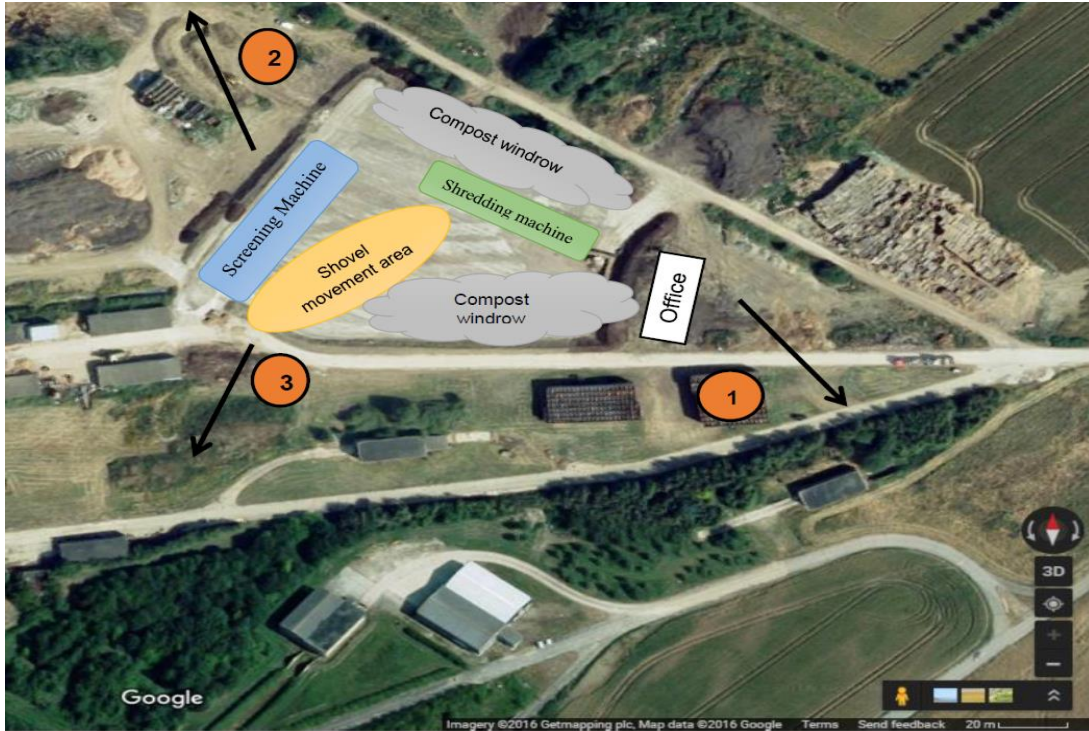


Figure 5-2: Ramsey Composting Site with the explanations of the sampling visits including the sampling locations, wind directions and composting operations.

- ● : Sampling locations on the three visits.
- —→ : The arrow represents the wind direction in each sampling visit.

The SIBS can detect the concentration of emissions in real-time, which means the SIBS detect the average fluorescent particles per second (figure 5-3). The SIBS data is presented below as both one minute average and the 30 min averages, to reflect the IOM/SKC sampling times. For the IOM/SKC sampling technique, there are four original dataset, namely bacteria at 37 °C, bacteria at 25 °C, fungi at 40 °C and fungi at 25 °C. In addition, for the IOM/SKC, we used the total number of bioaerosols by adding the highest concentration of bacterial growth plus the highest concentration of fungal growth for each sample, as explained in chapter 3.

5.3 Results

Wind speed and direction are the most important meteorological conditions that influence the airborne transfer of bioaerosols (Sanchez-Monedero, Stentiford and Mondini, 2003). The meteorological data from the three sampling visits is presented in Table 5-1.

Table 5-1: Weather conditions and Composting operations during the three sampling visits. The weather conditions include; wind speed and direction, temperature, humidity and cloud cover. The composting operations include; screening and turning during the first sampling visit, turning during the second sampling visit, and shredding and screening during the third sampling visit. S1, S2, S3, etc., indicates the samples number during each sampling visit, every sample means 30 min sampling. Therefore the total sampling time was 4 hours for every sampling visits.

Visits	Samples	Weather Conditions					Composting Operations
		Wind Direction	Wind Speed	Temp °C	Humidity	Cloud cover	
First Sampling Visit	S1	Northwest	3	6.2	73%	0%	Screening &Turning
	S2	Northwest	3	7	70%	0%	Screening &Turning
	S3	Northwest	3	7.2	60%	0%	No activity
	S4	Northwest	4	8	60%	10%	No activity
	S5	Northwest	5	9	56%	30%	Screening &Turning
	S6	North	4	9	55%	30%	Screening &Turning
	S7	North	3	8.6	57%	40%	Screening &Turning
	S8	North	4	8	63%	100%	Screening &Turning
Second Sampling Visit	S1	Southeast	1.4	17	54.9%	0%	Turning
	S2	Southeast	2	17	51%	0%	Turning
	S3	Southeast	2.2	17.6	46.4%	0%	Turning
	S4	Southeast	3	18.4	46%	0%	Turning
	S5	Southeast	3	18.3	47.5%	0%	No activity
	S6	Southeast	1.5	19.4	40.9%	0%	No activity
	S7	South-Southeast	1.7	20	45.4%	0%	Turning
	S8	South-Southeast	1.5	20.9	42.2%	0%	Turning
Third Sampling Visit	S1	Northeast	0	12	70%	90%	Screening
	S2	Northeast	1	14	73%	95%	Screening
	S3	Northwest	1	15	57%	95%	Screening
	S4	Northwest	0.8	15	58%	90%	Screening
	S5	Northwest	1	16	65%	90%	Screening
	S6	Northwest	1	17	62%	90%	No activities
	S7	Northwest	1	15	69%	90%	Screening
	S8	North	1.3	17	56%	80%	Screening & Turning

Figures (5-3, 5-4 and 4-5) show the concentrations of bioaerosol emissions for the three sampling visits to the Ramsey composting site. The 1 min average data of SIBS show the fluctuation in the bioaerosol concentration during the sampling time, which is the significant advantage of the SIBS sampler. The figures show the variation of emission concentration during the sampling time, which is not detected by the IOM/SKC technique.

Table 5-1 shows that in all three sampling campaigns, the sampling started when the sampler was downwind of the emission source as the location of the sampler was selected depending on the wind direction. After sampling started, the wind direction started changing gradually, until the sampler was no longer directly downwind of the emission source, which affected the emission concentration (see figures below). As would be expected, when the sampler was downwind of the emission source, the emission concentration was higher than when wind changed direction because the sampler was no longer directly downwind. The greatest change in wind direction was in the first and third visits in that order. The change in wind direction during the second sampling visit was smaller in comparison with the other two visits; therefore, the differences in the bioaerosol concentrations were smaller in the samples from this sampling visit.

In all three sampling visits, it was not easy to move the sampling locations with the winds because it is difficult to move the samplers after installing and the difficulties of finding a new location suitable with the new direction of the wind. Additionally, it was a good chance to check the effect of the wind direction on the bioaerosol dispersion.

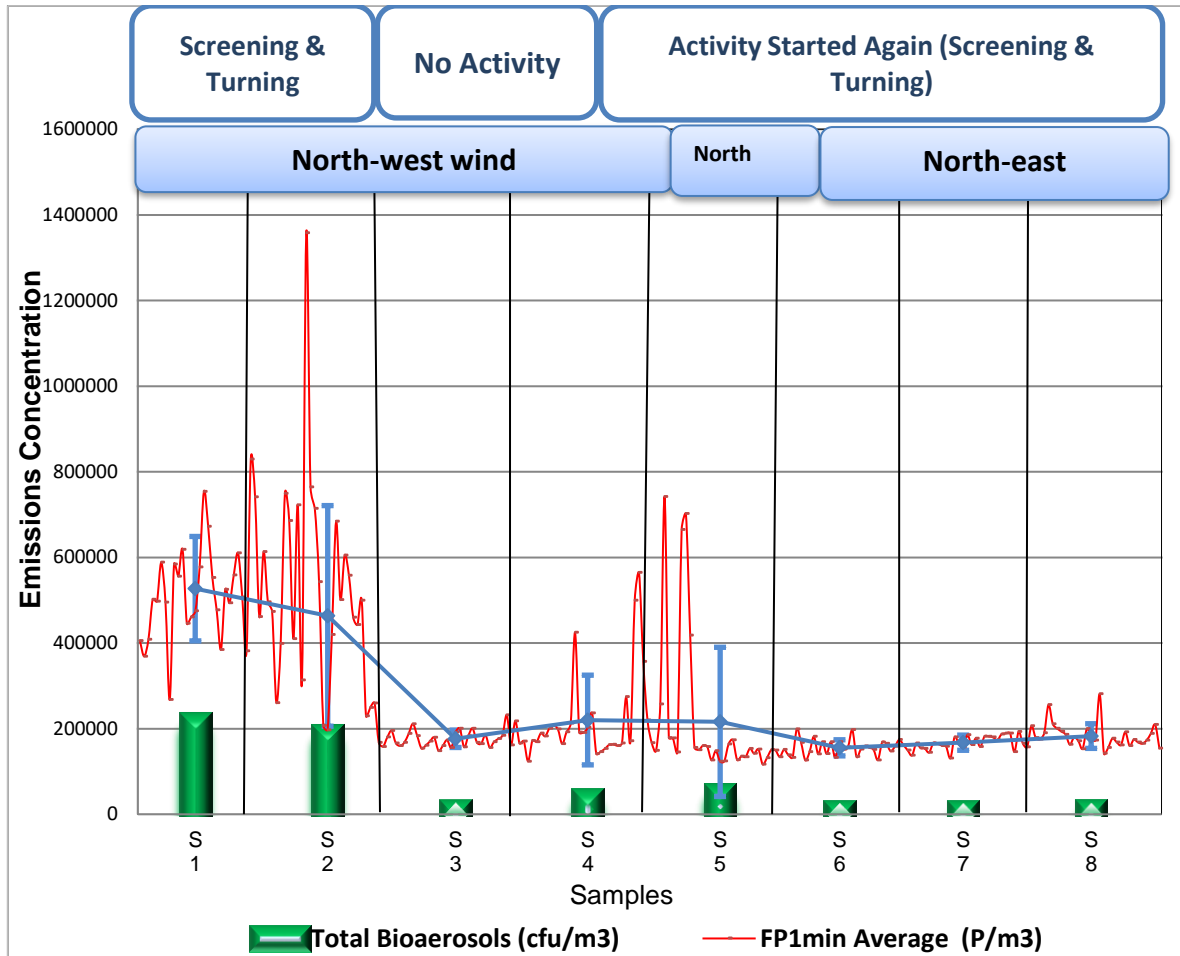


Figure 5-3: Fluorescent Particles and Total Bioaerosols concentrations from Ramsey composting site measured using the SIBS and the IOM/SKC during the first sampling visit in March-2016. The fluorescent particles data are presented as 1 min average (Red line) and 30 min averages (blue line). Total bioaerosols (green bars) represent the combined value of the highest bacteria and the highest fungi regardless of incubation temperature.

The highest emission concentrations of all three visits were recorded in the first sampling hour of the first visit (Figure 5-3) because screening and turning were being carried out, the sampling location was directly downwind, and the wind speed was between 3-4 metre/sec. In the second hour of sampling, the composting activities stopped for lunch, which caused a significant decrease in the emission concentrations.

After the lunch break, the composting activities started working again, and the concentration started increasing but did not reach the peaks of the samples before the break. The change in the wind direction was most likely the reason for this difference before and after the break.

During the second sampling visit, the change in wind direction was much less than during the first visit (Table 5-1). Therefore, the differences in the concentrations between the samples of the second visit were lower than the differences between the samples of the first visit (Figure 5-4). There was a small deflection in wind direction; therefore, there was a decrease in the emission concentrations during the second half of the sampling time (Samples 6, 7 and 8).

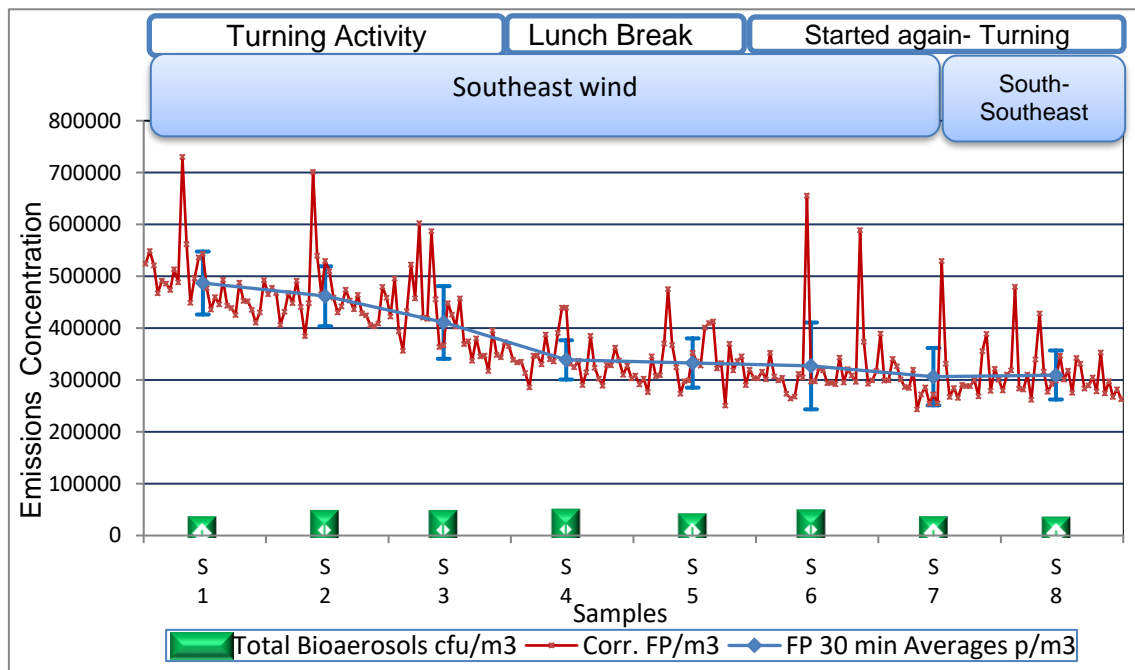


Figure 5-4: Fluorescent Particles and Total Bioaerosols concentrations from Ramsey composting site measured using the SIBS and the IOM/SKC during the second sampling visit in May-2016. The fluorescent particles data are presented as both 1 min average (Red line) and 30 min averages (blue line). Total bioaerosols (green bars) represent the combined value of the highest bacteria and the highest fungi regardless of incubation temperature.

In the third sampling visit (figure 5-5), the wind scenario was similar to what happened in the first visit where there was a significant change in wind

direction, which meant that the sampling location no longer directly downwind of the emission source. Furthermore, the scenario of emission concentration level was similar to what happened in the first sampling with differences in the general range of emission concentration.

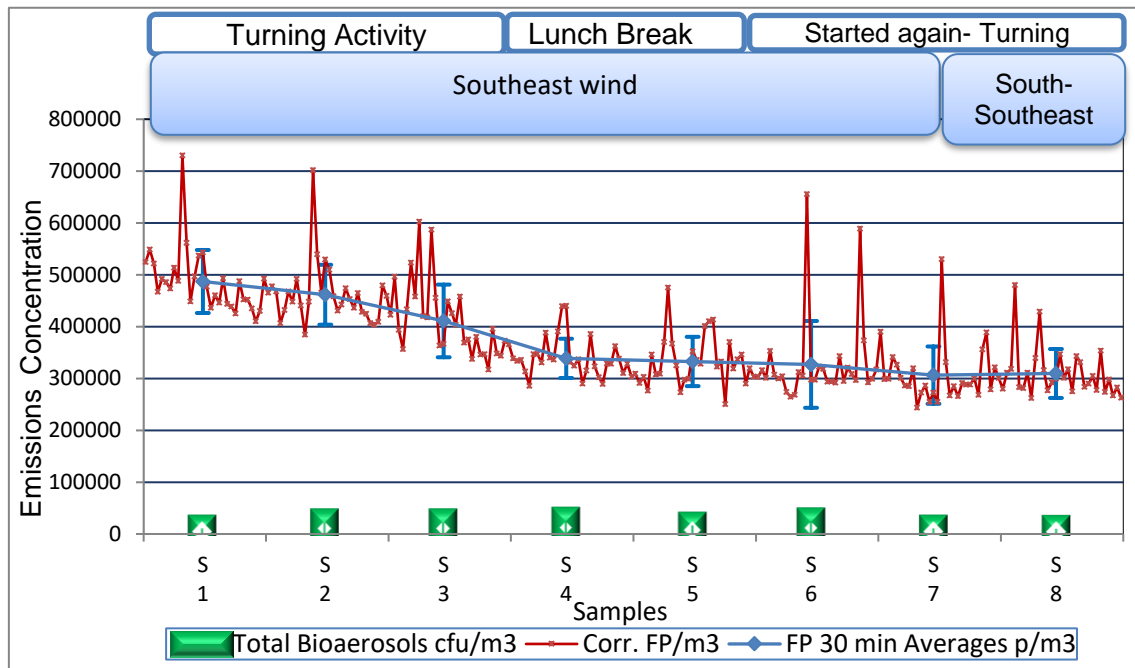


Figure 5-5: Fluorescent Particles and Total Bioaerosols concentrations from Ramsey composting site measured using the SIBS and the IOM/SKC during the third sampling visit in October -2016. The fluorescent particles data are presented as both 1 min average (Red line) and 30 min averages (blue line). Total bioaerosols (green bars) represent the combined value of the highest bacteria and the highest fungi regardless of incubation temperature.

The wind speed played an important role in the scenario of the third visit, where the wind speed was very slow (frequently below 1 m/sec). However, there were still significant levels of bioaerosols, although the wind direction changed during the sampling. This suggests that the emission dispersed in all directions because of the slow wind speed.

The SIBS data of the three sampling visits appears similar to the results of Pankhurst, (2010) who used the WIBS3 to detect fluorescent particles from composting activities. For example, Pankhurst (2010) measured the total

fluorescent particles from turning activity using the WIBS3 which reached 5.0×10^5 p/m³, where the mean concentration of turning activity detected using the SIBS was 4.9×10^5 p/m³.

Figures (5-3, 5-4 and 5-5) together show an important phenomenon, namely the highest concentrations always have a greater standard deviation, due to the nature of bioaerosol emission from the composting activity. For example, the highest 30 min average during the first sampling visit was 5.27×10^5 (first sampling period) with 1.22×10^5 standard deviation, where the lowest 30 min average (third sampling period) was 1.17×10^5 with 2.03×10^4 standard deviation. These results clearly highlight the intermittent nature of emissions from composting facilities.

Analysis of the real-time data for sample-2 in the first visit, shows a high variability in concentration with the time, even though there were only a few seconds between sample points. The variation of the bioaerosol concentration from composting agitation activities depended on factors such as wind conditions, and properties of the compost windrows (Taha et al., 2006).

For both the 1 min average and 30 min averages, the lowest concentration was detected during the first sampling visit, which was 116654 P/m³ and 155245 P/m³ for 1 min averages and 30 min averages respectively. Although these two lowest concentrations were detected during the same sampling visit, they were detected during two different sampling periods. The lowest 1 min average where during the fifth sampling period, where the lowest 30 min average was during the sixth sampling period. This shows the significance of the real-time data, by demonstrating that the lowest 30 min average does not correspond to the lowest of concentration monitored.

The 5th sampling period during the 1st visit (figure 5-2) shows that the average of this 30 minutes was 21588 P/m³, even though most of the samples for the same period were lower than this average. The average is influenced by a few very high concentrations. To be more specific; only in five samples (or readings) which is about 16.6% of all the samples of this 30 minutes were very high

concentration captured, and the rest of the samples (which were about 83.3%) were lower than the average. These high concentrations had a significant impact that made the average concentration higher than most samples.

The real-time concentration detected using the SIBS show the fluctuation of the emission concentrations with time. Furthermore, this fluctuation shows how the emission disperses from each kind of composting activities and how the emissions disperse when there is one composting activity or more. This advantage is not available in the traditional methods.

5.4 Discussion

The first and most important feature in the bioaerosol sampling using the SIBS are the 1 min average data, show the fluctuation of the emission concentration from the composting site during the operations (Figures 5-3, 5-4 and 5-5). These data show the relationship between the composting activities and the measured bioaerosol concentration, and how the bioaerosols disperse from the sources. For example, the highest 30 min average detected during the 1st sampling period was during the first sampling period (sample-2) which was (920187 FP/m³) as two activities were happening (turning and screening) and the sampler was downwind of the composting operations. However, this does not mean that this period has the highest peak concentration, because it is clear that the highest concentration was during the second sampling period (sample-2), exactly in the 10th minute of this period (1358025 FP/m³). However, the event record for this visit, shows that a lorry passed in front of the sampler which agitated the bioaerosol on the ground and therefore this high concentration did not result from the composting activities alone. In addition, as the 1 min average data consist of 60 seconds of data, there may be times when concentrations are higher than the highest 1 minute and 30 minute averages as well.

Table 5-2: The statistics of the three sampling visits to Ramsey. SIBS data (FP/m³) including 4 hours sampling average, standard deviation, median, maximum and minimum of 1 min average, and maximum and minimum of 30 min average.

Sampling	4 hours Average	Standard Deviation	Median	Average Time	Maximum	Minimum
First Visit	263487	183726	178411	1 min Average	1358025	116654
				30 min Average	526941	155245
Second Visit	371761	88657	341868	1 min Average	730219	243456
				30 min Average	487038	306386
Third Visit	652711	420510	560641	1 min Average	4593419	367545
				30 min Average	920187	496163

All the three sampling visits to Ramsey composting site have detected the bioaerosols concentration for four hours continuously. Four-hour average concentration has been calculated to compare all three visits. The highest average concentration was during the third sampling visit; because of the sampler location and wind speed. The sampler location in the third visit was very close to the composting activities (about 35 m from the screening activity and 45 m from the shredding activity), while the sampler was located about 100 m and 75 m from the emission source in the first and second sampling visit respectively (figure 5-2).

In addition to the sampler location, the wind speed during the third sampling visit was very low and sometimes less than 0.5 m/s which means that the

emission disperses very slowly and therefore the emissions accumulated around the source (Beychok, 2007).

On the contrary, the lowest four-hour average was in the first sampling visit. Although there were two composting activities (turning and screening), the distance between the sampler and the emission sources has likely resulted in this lower average. However, in all sampling visits, the concentration collected during active activities was always higher than the concentration from the passive sources (during activity breaks). This corresponds to what was found by Taha *et al.*, (2006).

Table (5-3) provides an analysis of fluorescent particles data collected from the Ramsey composting site during the three sampling visits. These three sampling visits differed in three main factors which are the season, composting activities, wind (speed and direction), and therefore the sampling locations. This table also shows the fluorescent particles when there was one composting activity and when there were two composting activities, as well as the fluorescent particles concentration from the passive source.

Furthermore, the impact of the wind direction on the measured concentration is highlighted by showing the fluorescent particles concentration detected when the sampler was downwind to the emission source and when the sampler was not directly downwind, due to a change in the wind direction.

Finally, this table includes the peak to mean ratios, which clarify the differences between the peak concentration and the mean concentration. However, some peak to mean ratios are by non-composting activities, such as vehicle movement in front of the sampler or vehicles transporting raw materials to the composting site.

Table 5-3: The statistics of the three sampling visits to Ramsey- SIBS data (P/m³) including 4 hour sampling average, an average of 30 min sampling, standard deviation, median and maximum and minimum of 30 min sampling.

Visit	Composting Activities	Wind Direction	Samples durations	Mean	Median	Peak/mean ratio
1st visit	Screening & Turning	Sampler in the plume	S1 + S2	533383	501287	1.65
	Break	Sampler in the plume	S3 + S4	177925	172614	1.30
	Screening & Turning	Sampler out of the plume	S6 + S7 + S8	163922	161866	1.71
2nd visit	Turning	Sampler in the plume	S1 + S2+S3	466540	457583	1.57
	Break	Sampler in the plume	S4 + S5	333925	329677	1.42
	Turning with a little change in wind	A slight deviation in the wind direction	S7 + S8	316480	298255	2.07
3rd visit	Screening and Shredding	Sampler in the plume	S1+ S2	868112	659498	5.29
	Break	Wind direction started to change	S5 + S6	570695	563515	1.37
	Screening and Turning with a change in wind	Sampler out of the plume	S7 + S8	644536	558468	3.33

Similarly, the IOM/SKC data have been analysed in table (5-4). The IOM/SKC sampler provides only the total bioaerosol concentration and not the average during the sampling time. There are two key influences on variability in the bioaerosol concentration as measure by the IOM/SKC. Firstly, there is between sample variation, which is expected as these samples were taken during different composting activities and in different wind direction.

Table 5-4: The statistics of the three sampling visits to Ramsey for the IOM/SKC data (CFU/m³) including 4 hour sampling average, an average of 30 min sampling, standard deviation, median and maximum and minimum of 30 min sampling.

Sampling Visits	Kind of bioaerosol sample	4 hours Sample	Standard Deviation	Median	Maximum	Minimum
1st Visit	Bacteria 25°C	17951	22506	9444	52222	0
	Bacteria 37°C	51563	74848	15000	176944	278
	Fungi 25°C	5451	10366	278	27500	0
	Fungi 40°C	3854	9011	139	25833	0
	Total Bioaerosols	57049	84679	15139	204444	556
2nd Visit	Bacteria 25°C	5873	5873	10556	21944	10278
	Bacteria 37°C	5939	5939	13056	20556	3611
	Fungi 25°C	2083	1039	1667	4167	5278
	Fungi 40°C	3819	3796	2361	13056	833
	Total Bioaerosols	17951	6501	19583	25556	1667
3rd Visit	Bacteria 25°C	12118	20332	1111	46389	278
	Bacteria 37°C	28715	49685	3056	128333	0
	Fungi 25°C	3125	4186	1389	13056	556
	Fungi 40°C	1597	1730	972	5000	0
	Total Bioaerosols	31979	53541	4861	141389	1389

However, there is also a certain variability between the filters for the same sampling period. This disadvantage may reduce the accuracy of bioaerosol concentration measured using the IOM/SKC.

Total bioaerosols concentration shown in the table (5-4) and figures (5-2, 5-3 and 5-4) has been calculated by adding the highest bacterial concentration plus the highest fungal concentration for the same sample. The purpose of calculation to produce total bioaerosol was to compare between fluorescent particles measured using the SIBS and total emissions measured using the IOM/SKC rather than to compare fluorescent particles with two kinds of bacteria data and then with fungi data. While this is not a true total bioaerosol concentration, it is the closest approximation permissible with the data available.

The correlation coefficient (table 5-5) between SIBS samples and IOM/SKC samples is useful to analyse how SIBS data relates to the traditional data as present knowledge is based on the traditional methods.

Table 5-5: The correlation coefficient between FP/m³ (SIBS data) and CFU/m³ (IOM/SKC data) during the three Ramsey visits. The SIBS data includes the 30 min averages of every sampling visit. The IOM/SKC data include five readings; bacteria at two temperatures, fungi at two temperatures and total bioaerosols. Total bioaerosols is the sum of the highest bacteria and the highest fungi regardless of incubation temperature.

SIBS with:	1st Visit	2nd Visit	3rd Visit
Bacteria 25°C	0.960	-0.115	0.931
Bacteria 37°C	0.994	0.398	0.945
Fungi 25°C	0.968	0.053	0.860
Fungi 40°C	0.833	-0.211	0.793
Total Bioaerosols	0.997	0.081	0.942

The first sampling visit data shows the highest correlation between the SIBS and IOM/SKC data, whereas there was no relation during the second sampling visit for any of the data. However, if the first and last samples of bacteria data are excluded as an example, the correlation increases to 0.826. This means there was a positive correlation between most samples (6 samples) and the other two samples affected the final result of the correlation coefficient.

The data from the third sampling visit also shows a positive correlation between SIBS and IOM/SKC data. The highest correlation was always with bacteria data as most bacteria concentrations were higher than fungi. Overall, the correlation coefficients reveal that the IOM/SKC data does not always represent the real concentration of bioaerosols emitted.

Previously, several studies have measured bioaerosol concentrations during different composting agitation activities including turning, shredding and screening (Douglas, 2013; Lacey, J., 1997; Pankhurst et al., 2009, 2011; Sanchez-Monedero et al., 2003; Taha et al., 2006, 2007; Weber et al., 1993). Although these studies have used different sampling techniques and different sampling scenarios, the collected bioaerosols concentrations data differs from the data presented here using the SIBS. The traditional sampling methods that were used by the previous studies provided snapshots of bioaerosol concentration as the sampling time was very short if compared with the sampling time used in this study, which always was four hours continuously. However, the SIBS provided continuous monitoring for four hours with data recorded each minute. The SIBS data shows the fluctuations of the concentrations in relation to the agitation activities and the weather conditions.

5.4.1 Limitations

There were several limitations to the work presented here as summarised below:

- 1- It is difficult to sample close to the agitation activities due to the dangerous conditions. Sampling close to the emission source would provide important data about the bioaerosol concentration and dispersion from each type of agitation activities.
- 2- There was only one opportunity during all the sampling visits to the Ramsey composting site to sample the bioaerosol concentration from a single composting activity. This was during the second sampling visit, when only turning activity was occurring. Therefore, bioaerosol sampling for screening and shredding as individual sources is needed to analyse how bioaerosols are emitted from each type of activity in isolation.
- 3- The results presented here do not include a background bioaerosol concentration (as there was only one SIBS available), which would demonstrate the effect of the composting site on the bioaerosol concentration more accurately.
- 4- The time available to samples was limited by the filtration technique and the number of samples that could be processed for each visit due to issues such as limited space in incubators for agar plates and so on.
- 5- The high lower limit of detection of the IOM/SKC was a significant limitation as there was no growth on many agar plates.

5.5 Conclusion

The comparison between the SIBS and the IOM/SKC during the sampling visits can be summarised as follows:

- a- The SIBS data show how the emissions are released from the composting site over time, and that is not possible with the IOM/SKC technique. The fluctuation of emission concentration explains the relationship between composting operations or meteorological conditions and the emission concentration.

b- The wide differences between the range of fluorescent particle concentration using the SIBS and the bioaerosol concentration using the IOM/SKC indicate the higher sensitivity of the SIBS in comparison with the IOM/SKC.

For example, due to the change in the wind direction during the first and third sampling visit, both samplers were no longer within the emission plume , but the SIBS was still able to detect the low level of fluorescent particles where the IOM/SKC could not detect the low levels of bioaerosol.

5.5.1 Key findings

The main conclusions from the sampling campaign are:

- 1- The SIBS is more sensitive than the IOM/SKC technique. Therefore, it can sample at high concentrations (for example, during composting activities) and low concentration (background levels).
- 2- SIBS data show that the fluorescent particles concentration is much higher than bioaerosol concentration detected using traditional sampling technique.
- 3- The SIBS is a suitable technique to study and analyse the emission concentration levels whether during the operating time of the composting activities or any other time.
- 4- The SIBS detects the fluctuations of bioaerosol concentration from composting facilities, which is not possible by the IOM/SKC or other traditional techniques.

6 Emission rate calculations

6.1 Introduction

This chapter aims to improve the output of bioaerosol dispersion modelling by improving the inputs, specifically the emission rate and source representation. This work is linked to the second objective of this thesis which is to determine whether the SIBS data can be used to improve bioaerosol dispersion modelling from composting facilities.

An atmospheric dispersion model is a powerful means to evaluate downwind and air pollution concentration which gives information about pollutant emissions and the nature of the atmosphere. Air dispersion models are mostly used to identify the best solutions for particular environmental issues (Bluett et al., 2004). An air dispersion model uses mathematical equations of atmospheric flow to simulate air pollutant dispersion by depicting the atmosphere, dispersion, and physical and chemical processes within the plume to calculate concentration at various locations (Holmes and Morawska, 2006).

Dispersion can also be affected by turbulence in the atmospheric boundary layer, which is random by nature and therefore cannot be exactly predicted (Chang and Hanna, 2004). Bioaerosol dispersion could be affected by the particle size, the emission rate, buoyancy effects, atmospheric effects and local topography (Drew et al., 2006). Certain factors can cause doubts in the model results such as errors in the input data, model physics, and numerical representation (Chang and Hanna, 2004).

Meteorological and emissions data are the basic input parameters that are required in dispersion modelling (Sharma *et al.*, 2004). The pollutant emission rate is a significant model input, which represents the amount and rate of emitted material from the source of the release (Barratt, 2001). Emission rates used in previous studies differ by many orders of magnitude depending on the source type modelled (Douglas, 2013). Full details on the ADMS model and emission rate calculations were provided in Chapter 2. This chapter aims to

improve the accuracy of bioaerosol dispersion modelling. This comprised two main activities, firstly, using novel bioaerosol data to calculate the emission rates from composting agitation activities. This kind of data have not been used before in dispersion modelling of bioaerosol emissions. Usually, two methods are used to calculate the emission rate of bioaerosol emission from composting facilities which are direct calculation and back-calculation. In this research, back-calculation method was used because the data currently available have been collected downwind of the emission source, while direct calculation requires data collected directly at the emission source.

6.2 Methods

Calculating the emission rate using the back-calculation method involved two stages, namely modelling and calculations using the measured bioaerosol concentration from sampling visits to the Ramsey composting site. The modelling included many steps starting with define the sources through a number of source scenarios, followed by modelling tests, which revealed that some of these scenarios did not work as detailed later in this chapter. The second step of the modelling was defining the model inputs. Some inputs were measured during the sampling visits, but others were used from previous studies, especially Douglas (2013). The third step was running the modelling tests for all planned scenarios for all three visits, where each visit has its conditions such as weather, sampling locations, agitation activities, and so on. The calculation stage, involved adjusting the emission rate model parameter until the modelled outputs resemble the measured concentration data.

6.2.1 Modelling

Generally, three sets of inputs have been used to estimate the emission rate of the three sampling visits to the Ramsey composting site. Each set has been run with different source scenarios depending on the type of agitation activities, as well as the passive emission sources.

Source Scenarios

In this study, different scenarios were used to represent the emission sources of bioaerosol from Ramsey composting site depending on the previous literature and by direct observations at the composting site. The purpose of using different scenarios is to produce an ensemble of results and then compare the results with previous studies and to compare the different source scenarios with each other.

Point sources have been used extensively to represent bioaerosol emissions from different activities such as turning, screening and shredding (Millner, Bassett and Marsh, 1980; Danneberg *et al.*, 1997; Taha *et al.*, 2006, 2007; Drew *et al.*, 2007).

For turning activity, a new scenario has been used here in addition to the point source, which is the two points sources scenario. Douglas (2013) stated that the turning activity consists of three actions, namely removing material from existing windrow A with a front end loader (FEL), transportation of the material and depositing the material to form new windrow B. According to Douglas (2013) (Figure 6-1); the bioaerosol emissions were elevated from actions 1, and 3 as the bioaerosol emissions were negligible throughout action 2. Therefore, the two point sources scenario has been used to represent these two actions of turning activity. However, a third scenario for turning was also considered, which represents the movement of the FELs as a line source scenario.

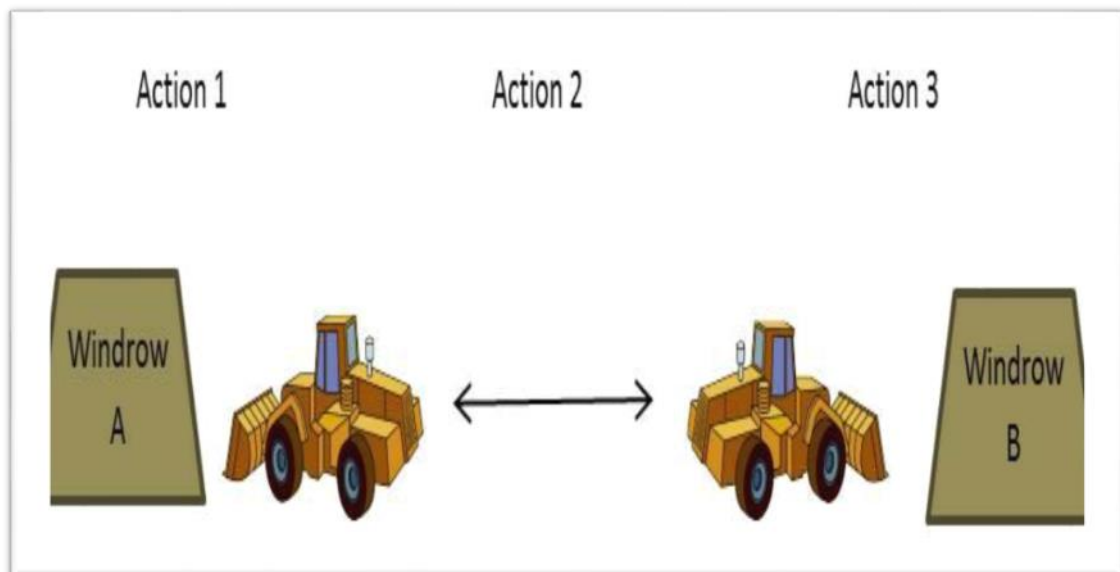


Figure 6-1: A profile illustration of the typical actions performed during turning activities with Front End Loader [FELs] (not to scale) (Douglas, 2013).

Observations of the composting activities during sampling, revealed that using a line source in this study was appropriate to represent two composting activities, namely turning and screening.

The length of the screening machine was about 48 metres (as shown in figure 6-2), and the width of the emission area was less than 1 metre. Emissions are almost all released along the length of the screening machine as well as from the movement of the shovel beside the machine; therefore emission can be represented as a line.



Figure 6-2: Illustrate how the Screening machine can be considered as a line source, where the machine length up to about 48 meters and the emission are emitted from all parts of the screening machine.

During the sampling when no activity was happening, there were still emissions released from the composting windrows and the samplers have detected bioaerosol concentrations. However, there was a marked difference between active and passive concentrations. Composting windrows extend to several square meters; therefore the most proper scenario to represent these windrows is an area source.

Table 6-1: All the source scenarios used to represent the source geometry in the modelling test to estimate the emission rates of bioaerosol emissions from Ramsey composting site during three sampling visits.

Composting Activity	Source scenario	Justification
Turning	2 points	Consider the activity of removing the compost material as a point source and depositing the material to form a new windrow as a second point source.
	Point	As used in previous studies, consider all turning actions as a point source in general.
	Line	The movement of the shovel vehicle between the old and the new windrows in addition to the activities of removing and depositing the compost material, all these activities can form a line source of emissions.
Screening	Point	Consider shredding machine as a point source of emissions.
	Line	Emissions are mostly released along the screening machine as well as from the shovel movement beside the machine; therefore emission can be released as a line (figure 6- 2).
Shredding	Point	The emissions disperse from the shredding machine in limited point shape, and the point source can be the nearest scenario to the fact.
Passive source	Area	The compost windrows were close to each other which formed an area of compost material, and the emission disperses from all these windrow during the passive dispersion of emissions.

Model inputs

The model inputs that were used in the modelling can be classified into two groups:

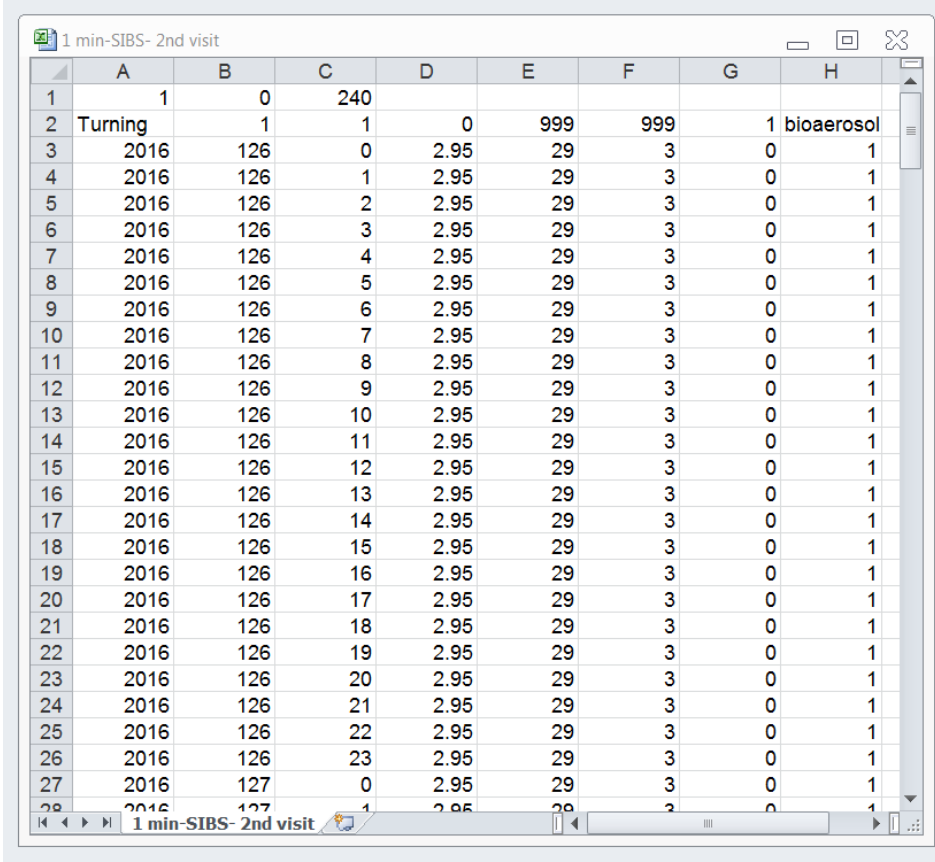
- a- Measured data which was collected and recorded during the sampling time, these inputs include:
 - Weather data: Kestrel Pocket Weather Tracker 4000 was used to measure weather conditions such as wind speed, temperature and humidity.
 - Locations of the composting activities and samplers.
 - types of the composting activities during each day of sampling.
 - The heights and dimensions of the emission sources.
- b- The second group of data that were used based on (Douglas, 2013) which are not easy to measure during the sampling and includes: pollutant exit velocity, pollutant temperature, pollutant heat capacity and pollutant molecular mass (Table 6-2).

Table 6-2: Model inputs that were used in the modelling tests depending on Douglas (2013) including pollutant exit velocity, pollutant temperature, pollutant heat capacity and pollutant molecular mass.

Input	Description or Value
Source Height	2.65
Pollutant exit velocity	2.95
Pollutant temp	29
Heat capacity	1519
molecular mass	28.996

6.2.1.1 Model options

Attempts were made to incorporate the SIBS data as real-time data (1-minute data) into the ADMS model. One of these attempts was using the time-varying source data option using the .var file. This file includes year, Julian day, hours, volume flow rate of the emission, source temperature, source diameter, the initial ratio of water of water and the emission rates. This input list appears for every line of modelled meteorological data (CERC, 2012). However, this option in ADMS can only model the emission dispersion hourly as smallest time unit. The SIBS data represents the emission concentration for each minute, so attempts were made to model this data by assuming the minutes as an hour as shown in figure 6-3.



	A	B	C	D	E	F	G	H
1	1	0	240					
2	Turning	1	1	0	999	999	1	bioaerosol
3	2016	126	0	2.95	29	3	0	1
4	2016	126	1	2.95	29	3	0	1
5	2016	126	2	2.95	29	3	0	1
6	2016	126	3	2.95	29	3	0	1
7	2016	126	4	2.95	29	3	0	1
8	2016	126	5	2.95	29	3	0	1
9	2016	126	6	2.95	29	3	0	1
10	2016	126	7	2.95	29	3	0	1
11	2016	126	8	2.95	29	3	0	1
12	2016	126	9	2.95	29	3	0	1
13	2016	126	10	2.95	29	3	0	1
14	2016	126	11	2.95	29	3	0	1
15	2016	126	12	2.95	29	3	0	1
16	2016	126	13	2.95	29	3	0	1
17	2016	126	14	2.95	29	3	0	1
18	2016	126	15	2.95	29	3	0	1
19	2016	126	16	2.95	29	3	0	1
20	2016	126	17	2.95	29	3	0	1
21	2016	126	18	2.95	29	3	0	1
22	2016	126	19	2.95	29	3	0	1
23	2016	126	20	2.95	29	3	0	1
24	2016	126	21	2.95	29	3	0	1
25	2016	126	22	2.95	29	3	0	1
26	2016	126	23	2.95	29	3	0	1
27	2016	127	0	2.95	29	3	0	1
28	2016	127	1	2.95	29	3	0	1

Figure 6-3: Emission source input in .var file format. The emission rate value was 1 in the modelling test.

Unfortunately, this method did not work, because when the ADMS simulates the emission dispersion, the time of dispersion has a significant impact on the nature of the dispersion. For example, the model output for 11 am is not the same as at 1 pm even though the input was exactly the same. Figure 6-5 shows this problem more clearly, where a regular repeating pattern of emissions is shown that does not relate to the meteorological or emission rate inputs.

It is also clear from the results, that the sampling period needs to align with the model time periods. While the SIBS provides a high temporal resolution, as ADMS cannot cope with such short timescales (one minute), the model is not able to utilise the high resolution data. It would be necessary to adapt the model or find an alternative model to fully utilise the SIBS data.

To illustrate this further, two modelling tests were completed using the same input including emission rate, weather condition, source geometry and all other input but with two different times. The results from these two modelling tests were different, where the concentration of both modelling tests at the same receptor point was different and thus the time is an important factor in ADMS.

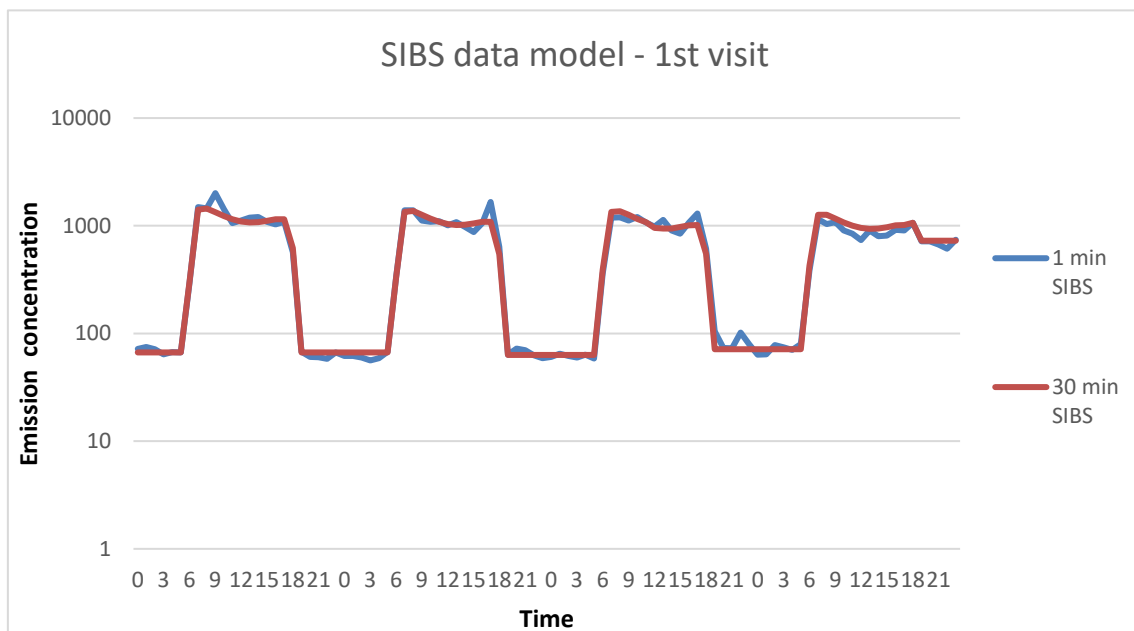


Figure 6-4: output emission concentration at the sampling location using .var file input data.

Furthermore, using the fluctuations option also did not work because the fluctuations that are considered by ADMS are the meteorological data with time, such as wind speed and direction. The focus of this research was on fluctuations in the emission concentration and not fluctuations in the meteorological data. Therefore, there was no option but to model one test for each condition and for each hour as well and using the output concentration as a base to calculate the emission rate for each minute. This involved completing a modelling test of a particular hour with measured meteorological conditions for this hour, and then use the output concentration to calculate the emission rate of the measured concentration using cross multiplication in Excel.

6.2.2 Calculations

From the three sampling visits, there were three different scenarios according to the composting activities and the season during each sampling visits. In the first scenario, the pollutant emission rates were estimated for the turning and screening agitation activities as the collected data represent the bioaerosol concentration from these two activities during the winter season (first sampling visit). In the second scenario, the pollutant emission rates were estimated for turning agitation activity as the collected data in this sampling visit represents the bioaerosol concentration from turning activity only during the spring season. Finally, in the third scenario, the calculated pollutant emission rates were for shredding and screening agitation activities performed during the autumn season. These bioaerosol data have been classified according to different criteria as shown in figure (6-5).

Generally, the data have been divided into the real-time data from the SIBS, and the average concentrations for the IOM/SKC and the SIBS as well. Then, the averages were classified as active and passive emission sources according to the sampling periods, and when agitation activities were occurring.

For all data, the modelling tests were performed with different source scenarios using emission rate = 1 and with weather conditions recorded during the sampling time to calculate the emission rates for these cases using the back-calculation method. This is summarized in figure 6-5.

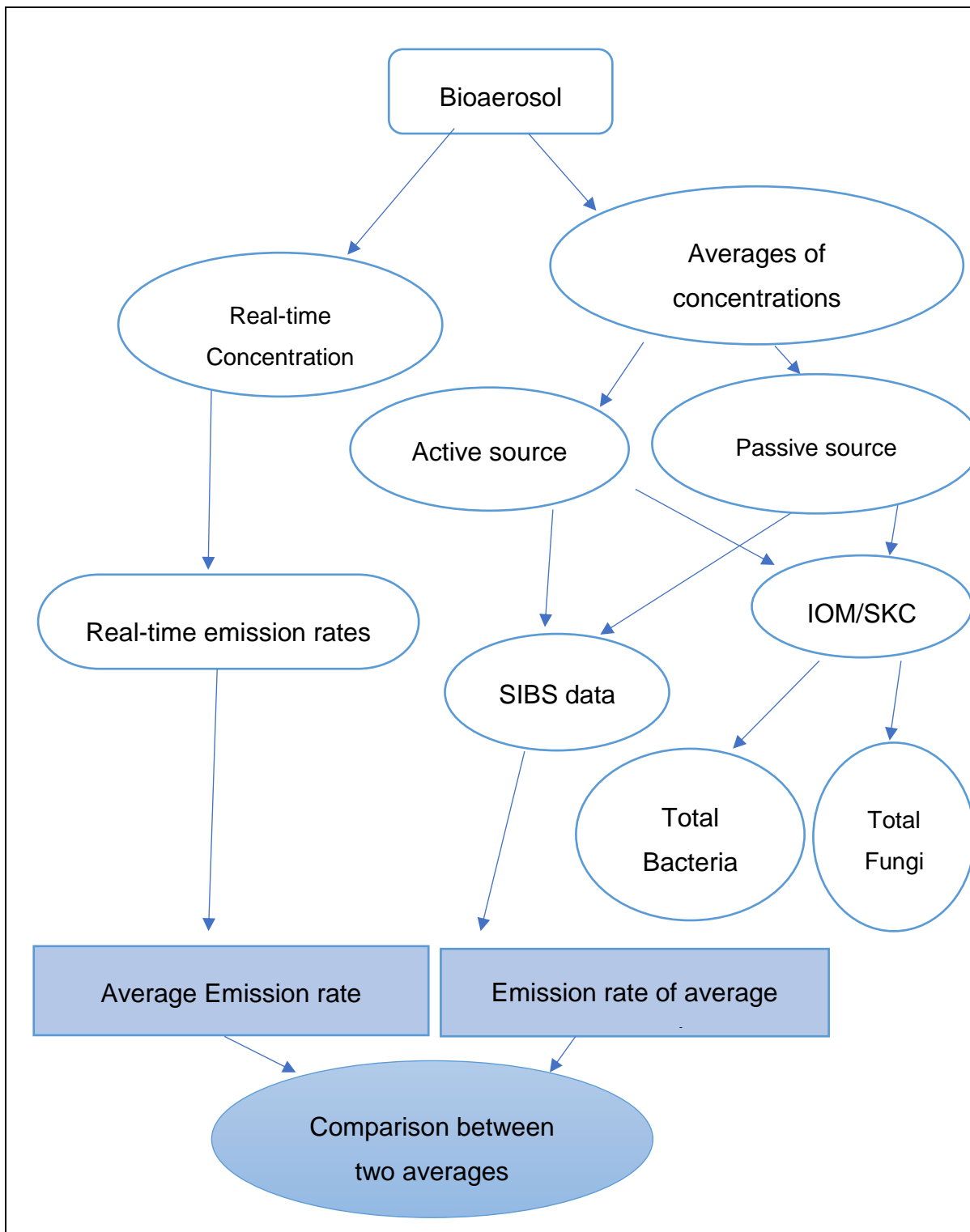


Figure 6-5: An overview of the structure of the bioaerosol data used in the emission rates calculation.

6.3 Results

The emission rates presented here for the fluorescent particles are the first time that emission rates of these particles have been calculated (as detailed in chapter 4).

6.3.1 Emission rates from active sources

All estimated emission rates of pollutants from active sources are presented in table (6-4), alongside the composting agitation activities, seasons during the sampling visits and the source scenarios used in the models.

Table (6-3) shows that the calculated emission rates for the fluorescent particles cover several orders of magnitude. Fluorescent particle emission rates calculated for point source varied from 4.92×10^9 P/s to 6.39×10^9 P/s, and for line source were 8.05×10^7 P/m/s and 4.21×10^8 P/m/s. All fluorescent particle emission rates were always much higher than the emission rates of total bacteria and fungi because the measured concentrations of fluorescent particles were always higher than the measured concentration of total bacteria and fungi.

Bioaerosol emission rates calculated for the point sources varied from 1.19×10^8 CFU/s to 1.55×10^9 CFU/s for total bacteria, and from 1.94×10^7 CFU/s to 2.48×10^8 CFU/s for the total fungi. For the line source scenario, the calculated emission rates of bioaerosol were 2.48×10^6 CFU/m/s and 9.27×10^7 CFU/m/s for the total bacteria, and 4.04×10^5 CFU/m/s and 1.48×10^7 CFU/m/s for the total fungi.

Table 6-3: summary of the parameters and consequential estimated emission rates for active sources calculated based on the bioaerosol concentrations collected in this study.

Composting activities	Season	Sources geometry	Bioaerosol kind	Bioaerosol Concentration	Estimated emission rate
Turning and Screening	Winter	Line and Line	Fluorescent particle	518005 (P/m ³)	4.21x10 ⁸ (P/m/s)
			Total Bacteria	114028 (CFU/m ³)	9.27x10 ⁷ (CFU/m/s)
			Total Fungi	18264 (CFU/m ³)	1.48x10 ⁷ (CFU/m/s)
Turning and Screening	Winter	Point and point	Fluorescent particle	518005 (P/m ³)	9.13x10 ⁹ (P/s)
			Total Bacteria	114028 (CFU/m ³)	2.01x10 ⁹ (CFU/s)
			Total Fungi	18264 (CFU/m ³)	3.22x10 ⁸ (CFU/s)
Turning and Screening	Winter	2 Points and point	Fluorescent particle	518005 (P/m ³)	4.39x10 ⁹ (P/s)
			Total Bacteria	114028 (CFU/m ³)	9.66x10 ⁸ (CFU/s)
			Total Fungi	18264 (CFU/m ³)	1.55x10 ⁸ (CFU/s)
Turning	Spring	Line	Fluorescent particle	432952 (P/m ³)	8.05x10 ⁷ (P/m/s)
			Total Bacteria	13333 (CFU/m ³)	2.48x10 ⁶ (CFU/m/s)
			Total Fungi	2176 (CFU/m ³)	4.04x10 ⁵ (CFU/m/s)
Turning	Spring	Point	Fluorescent particle	432952 (P/m ³)	6.59x10 ⁹ (P/s)
			Total Bacteria	13333 (CFU/m ³)	1.88x10 ⁸ (CFU/s)
			Total Fungi	2176 (CFU/m ³)	3.07x10 ⁷ (CFU/s)
Turning	Spring	2 Points	Fluorescent particle	432952 (P/m ³)	3.86x10 ⁹ (P/s)
			Total Bacteria	13333 (CFU/m ³)	1.19x10 ⁸ (CFU/s)
			Total Fungi	2176 (CFU/m ³)	1.94x10 ⁷ (CFU/s)
Shredding and Screening	Autumn	Point and point	Fluorescent particle	888222 (P/m ³)	3.87x10 ⁹ (P/s)
			Total Bacteria	75972 (CFU/m ³)	3.95x10 ⁸ (CFU/s)
			Total Fungi	6319 (CFU/m ³)	3.29x10 ⁷ (CFU/s)

6.3.2 Emission rates from passive sources

The area source scenario was the only one scenario used to estimate the emission rates from the passive source term because the static compost windrows form an area relatively wider than point or line as used for the active sources (Taha et al., 2007). The calculated emission rates of the fluorescent particles from static windrows using the SIBS were 7.43×10^6 P/m²/s, 2.21×10^8 P/m²/s and 1.31×10^{14} P/m²/s for the first, second and third sampling visits respectively (table 6-4).

These emission rates were naturally higher than the passive emission rates for the IOM/SKC data. The calculated emission rates for the total bacteria were 6.64×10^5 CFU/m²/s, 7.76×10^7 CFU/m²/s and 1.91×10^{11} CFU/m²/s for each sampling visit respectively. Emission rates of bacteria were always higher than emission rates of fungi as the measured concentrations of bacterial bioaerosols were higher. The calculated emission rates for fungi emissions were 1.17×10^4 CFU/m²/s, 3.50×10^7 CFU/m²/s and 1.59×10^{11} CFU/m²/s for each sampling visit respectively.

Table 6-4: A summary of the parameters and consequential estimated emission rates for passive sources calculated based on the bioaerosol concentrations collected in this study.

Season	Source geometry	Bioaerosol kind	Bioaerosol Concentration	Estimated emission rate
Winter	Area	FP	185271 P/m ³	7.43x10 ⁶ P/m ² /s
		TB	15833 CFU/m ³	6.64x10 ⁵ CFU/m ² /s
		TF	278 CFU/m ³	1.17x10 ⁴ CFU/m ² /s
Spring	Area	FP	323721 P/m ³	2.21x10 ⁸ P/m ² /s
		TB	11389 CFU/m ³	7.76x10 ⁷ CFU/m ² /s
		TF	5139 CFU/m ³	3.50x10 ⁷ CFU/m ² /s
Autumn	Area	FP	571866 P/m ³	1.31x10 ¹⁴ P/m ² /s
		TB	833 CFU/m ³	1.91x10 ¹¹ CFU/m ² /s
		TF	694 CFU/m ³	1.59x10 ¹¹ CFU/m ² /s

6.3.3 Real-time emission rates

As the SIBS can detect the fluorescent particles concentration in real-time (minute by minute) it is possible to calculate the emission rate for every minute concentration using the back-calculation method. Figure (6-6) shows both the concentration of the fluorescent particles detected from turning activity in Ramsey composting site during the second sampling visit and the calculated

emission rates for turning activity as well. The emission rates in figure (6-6) have been calculated using a point source scenario; the other scenarios show the same pattern with the real-time concentration.

The purpose of the figure (6-6) is not to compare the concentration of fluorescent particle and the emission rates of the fluorescent particles, but to show and compare the trends. The emission rate of fluorescent particles was absolutely affected by the concentration, for example, when the concentration increases, the emission rate also increase and vice versa.

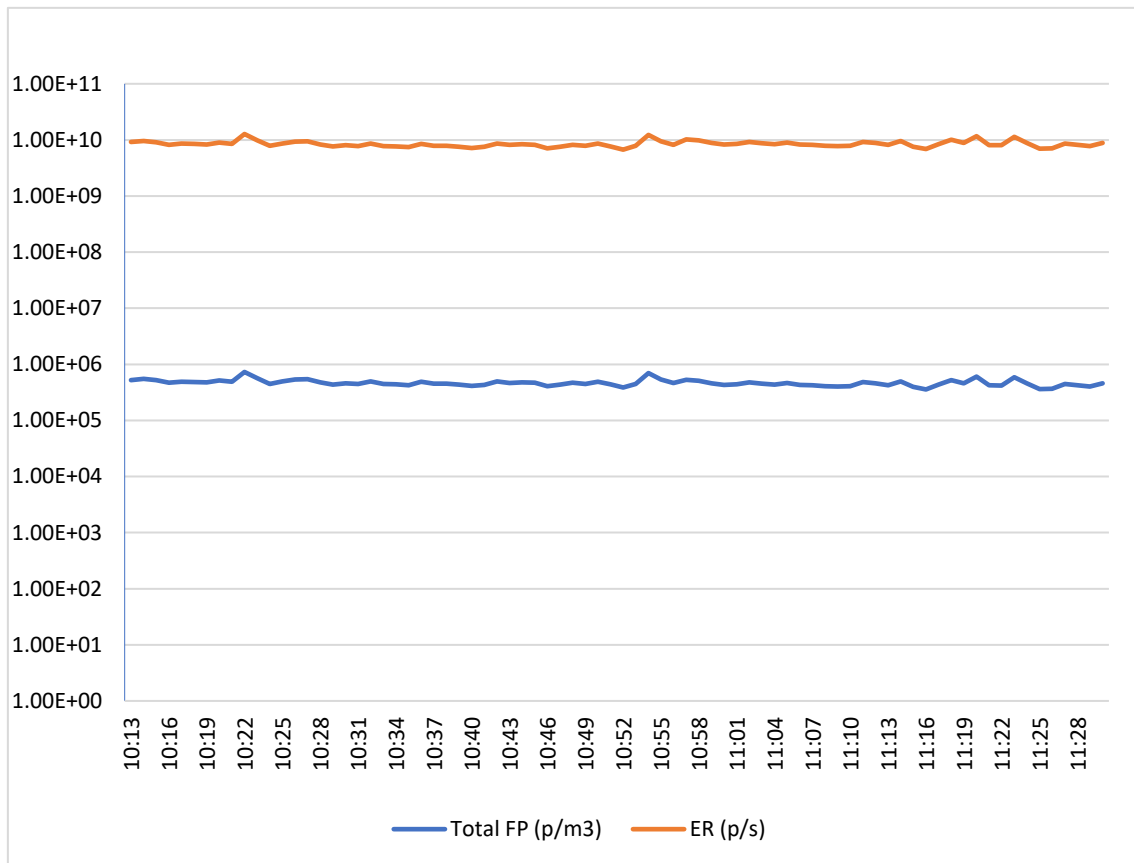


Figure 6-6: Real-time emission rates and the measured real-time emission concentration for the turning activity modelled as a point source during the second sampling visit to Ramsey composting site

6.4 Discussion

The calculated emission rates of fluorescent particles were always higher than the calculated emission rates of all traditional bioaerosol data, whether from this study using the IOM/SKC sampler or from other previous studies. Figure (6-7) shows the emission rates from turning activity using a point source scenario, which calculated for various types of bioaerosol. The emission rates of fluorescent particles calculated in this study were higher than the emission rates of total bacteria and fungi which were sampled using the IOM/SKC sampler alongside the SIBS. Another two studies have been displayed for comparison; firstly Douglas (2013), as the emission rates were calculated using the direct calculation methods and Shi and Hodson, (2010) used the back-calculation methods.

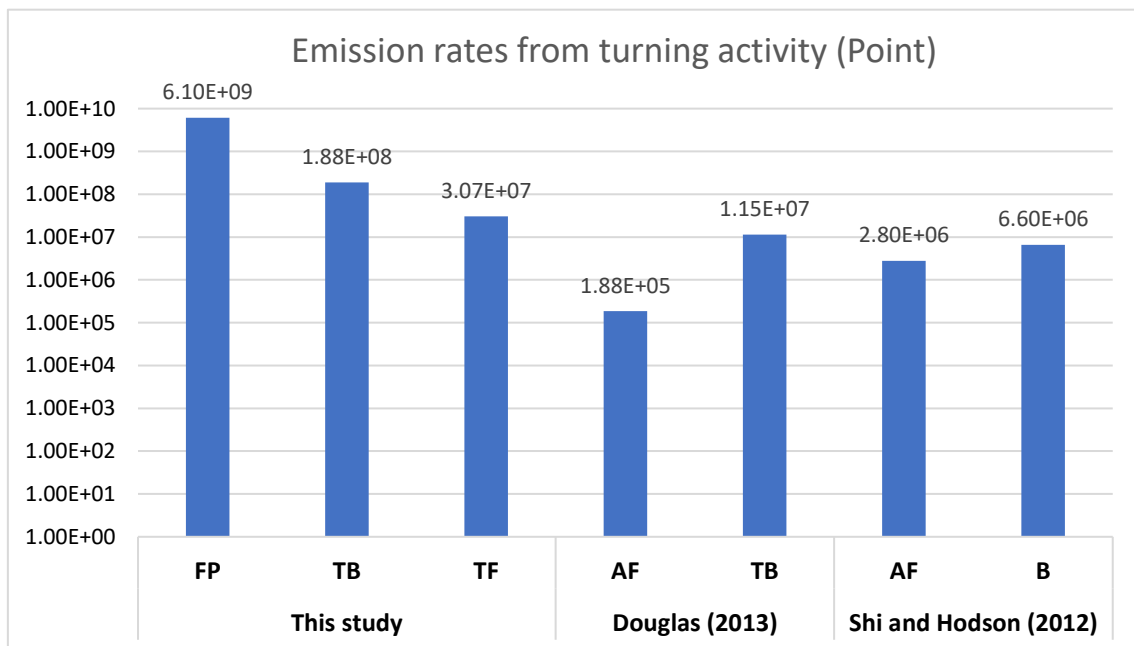


Figure 6-7: Emission rates for turning activity as a point source calculated in this study and from Douglas (2013) and Shi and Hodson (2012). The abbreviations correspond as FP to fluorescent particles, TB to total bacteria, TF to total fungi, AF to *Aspergillus fumigatus* and B to bacteria.

The bioaerosol emission rates calculated in this study depended on the fluorescent particle concentration detecting using the SIBS, and bioaerosol

concentration (bacteria and fungi) using the filtration sampling technique as a traditional data. Several studies have tried to calculate the emission rates based on traditional bioaerosol data detected at different distances from different agitation activities (Douglas, 2013; Drew et al., 2007; Millner et al., 1980; Taha et al., 2006, 2007). The average emission rates estimated in this study using the traditional methods are within the same orders of magnitude as previously published studies, but the emission rates of the fluorescent particles were higher. However, the more recent emission rates calculated by Douglas (2013) may be considered as more accurate because the bioaerosol concentration that was used to calculate the emission rates was measured at the source using novel sampling approach. Furthermore, Douglas (2013) monitored for *Aspergillus fumigatus*, whereas this study monitored total fungi, so the emission rates are higher as would be expected.

The most significant advantage of the emission rates calculated in this study are the fluctuations that can be calculated using the SIBS data. These fluctuations can demonstrate how bioaerosols disperse from the source when these emission rates are used in the model, as will be shown in the next chapter. This data was not available before when the emission rates were calculated from previous studies using traditional sampling techniques.

When comparing all emission rates calculated in this study, figure 6-8 shows that the highest emission rate was calculated from the first sampling visit. Whereas the lowest emission rates were calculated from the third sampling visit, even though the concentration of this sampling visit was the highest concentration. This was because the wind speed during this sampling time, which was always less than 1 m/s. This suggests that the particles are staying around the site and disperse very slowly.

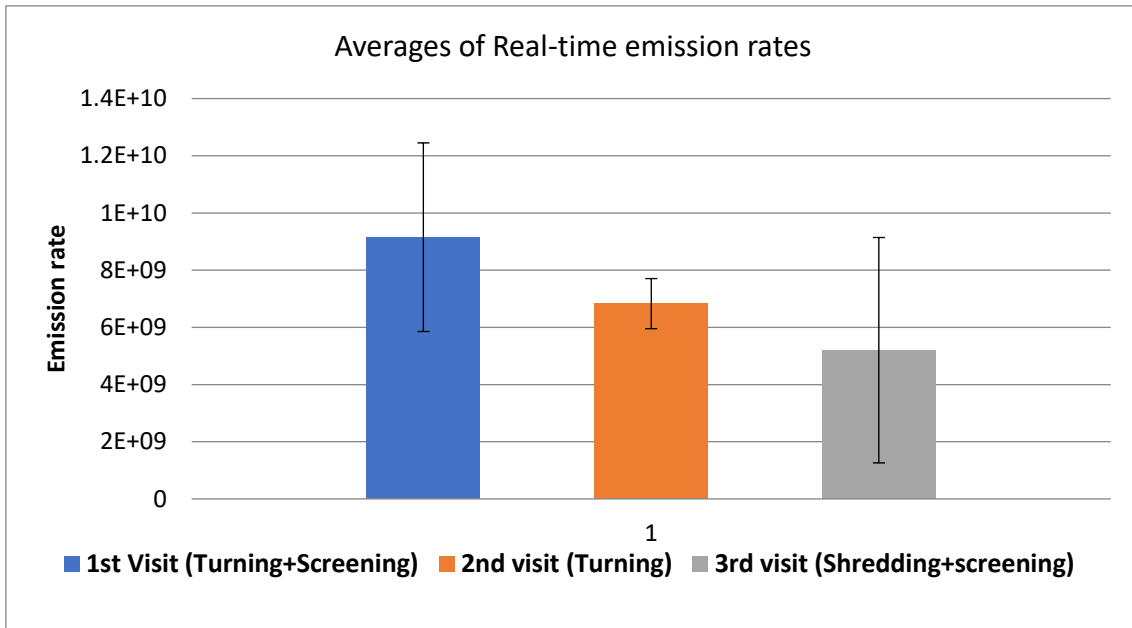


Figure 6-8: Emission rates based on the fluorescent particles collected from three sampling visits to the Ramsey Composting site, emission sources were modelled as point sources.

However, the emission rates from the second sampling visit can be considered as the most accurate emission rate, because it represents one composting activity (turning). The emission rates from the first and third sampling visits represent two composting activities which were turning and screening during first sampling visit and shredding and screening during the third sampling visit. Therefore, the emission rate represents two composting activities equally, which is unlikely to be accurate. It is almost impossible that two emission sources emit bioaerosol emissions at exactly the same rate.

In addition the real-time emission rates presented in (figure 6-6), demonstrate the fluctuation in the emission concentrations and emission rates for the turning activity. However, where there are multiple activities occurring, it is not possible to clearly identify their individual contributions, so the fluctuations cannot be accurately assigned to a source. Therefore, it is not appropriate to calculate the emission rates using the back-calculation method except if the concentration was collected from one agitation activity.

However, the emission rates calculated using the average concentration were equal to the average of real-time emission rates using the same data for the same hour. Statistically, the average of real-time emission rates is more useful than the emission rate of the average concentration as in the first the standard deviation, distribution and other statistical measurements can be calculated but cannot for the second.

Figure (6-9) shows the distribution of the average of the real-time emission rates from turning activity during the second sampling visit to the Ramsey composting site. The highest emission rates was between 7.00×10^9 P/s to 8.99×10^9 P/s, where the average was 6.83×10^9 P/s. Averages do not represent the full picture of emissions, particularly where the average is less than most of the samples of bioaerosol in this period in particular.

This shows the importance of real-time detection (fluctuations) of bioaerosol concentration to study bioaerosol dispersion and so to improve risk assessments regardless of whether this depends on direct measurement or modelling.

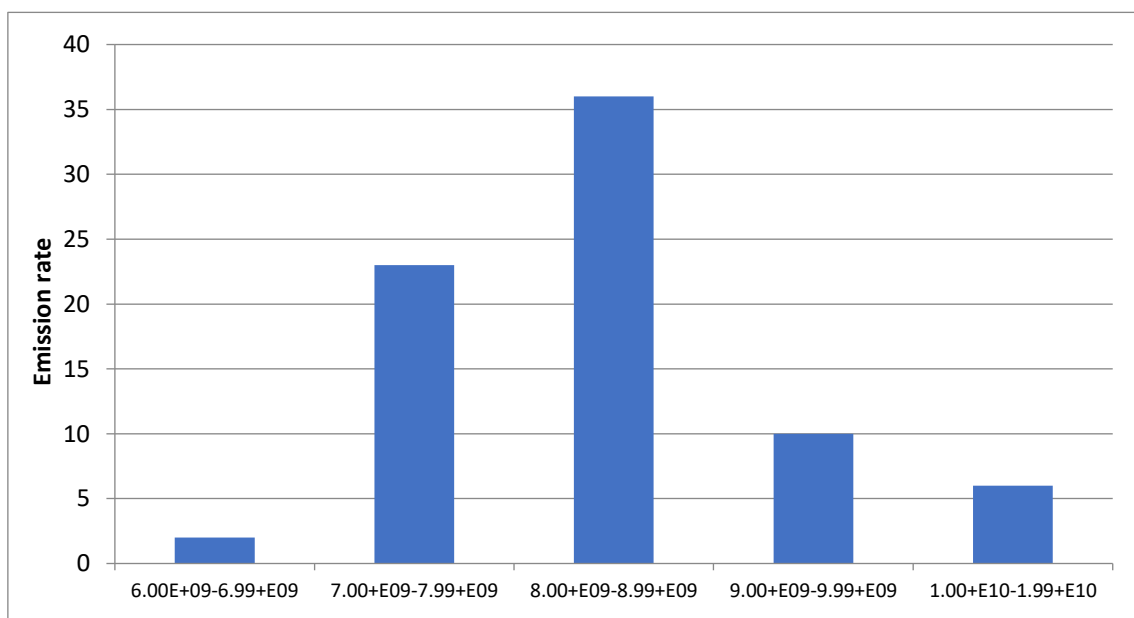


Figure 6-9: Distribution of the real-time emission rates average calculated from turning activity during the second sampling visit in May 2016.

6.4.1 Limitations

There were some limitations faced during the modelling studies. ADMS models dispersion hourly as the smallest time unit. The main objective of this project was to use the bioaerosol real-time data detected using the SIBS to improve the dispersion modelling of bioaerosols from composting sites. This limitation will be encountered by anyone who wants to use real-time data to model the bioaerosol dispersion using the current version of ADMS. However, the emission rates of fluorescent particles have been estimated using the method mentioned above.

Another limitation was the lack of data representing the background bioaerosol concentration. The data collected in this project and used to calculate the emission rates, represents the total concentration of background and composting as well. So, upwind sampling would have provided more accurate results.

Finally, there was only one dataset that represented a single composting activity, and the data that represents more one activity is not suitable to calculate the emission rates using the back-calculation method.

6.5 Conclusion

This chapter presents the first attempts to calculate emission rates from a real-time bioaerosol sampler, and highlights a number of challenges faced in this process. This chapter has also analysed the usefulness of existing and new source scenarios and suggests improvements to the representation of the source geometry. These improvements will enhance the use of the bioaerosol dispersion modelling in bioaerosols risk assessments.

As a conclusion from this section, several source scenarios can be used to represent the emission source in the model, however, determining which is the most appropriate is challenging. To represent the turning activity as an emission source; point, area and line scenarios can be used. Additionally, two points can

also be used, but this scenario still needs further research such as of the concentrations at both points as they will not emit the same concentrations.

For screening activity, the line scenario may be suitable as illustrated above, but this scenario has not been validated yet due to a lack of sampled data representing the screening activity only. Currently, the point source scenario is the best option to represent the shredding source. The emission that is emitted from the shredding machine is emitted from a known slot or gap. However, further research is required to improve and validate the scenarios for shredding, particularly data sampled at the source.

6.5.1 Key findings

- The calculated emission rates represent all biological particles as detected by the SIBS, whereas previous studies have only used culturable bioaerosol for modelling.
- The SIBS emission rates (emission rates of the fluorescent particles) were higher than the emission rates based on the traditional data from this and other studies.
- Real-time emission rates were calculated for the first time, and that will contribute greatly to improve the using of the bioaerosols dispersion modelling in the risk assessment.
- The SIBS data collected downwind of the emission sources can only be used if there is one composting activity occurring.

7 SIBS data modelling

7.1 Introduction

Many modelling studies have attempted to predict bioaerosol dispersion from composting sites but the prediction are still unreliable. The reasons for this are the model input, and the model itself. Most modelling studies are based on data showing only a snapshot of bioaerosol concentration. Furthermore, these data represent viable bioaerosols, and non-viable components will significantly increase the total concentration (Taha et al. 2007). Therefore, the emission rates calculated using these limited data are limited.

This research aims to improve bioaerosol dispersion modelling by using novel data collected at Ramsey composting site. The second objective of this thesis is to determine whether the SIBS data can be used to improve bioaerosol dispersion modelling from composting facilities. However, as the SIBS was used for the first time in this project to sample bioaerosol concentrations from a composting facility, it was also the first time that the SIBS data was used with dispersion modelling.

This objective was divided into two section, namely emission rates calculation (chapter 6) and modelling with the calculated emission rates (chapter 7). In the previous chapter, emission rates were calculated based on the bioaerosol data using the SIBS and the filtration sampling technique (IOM/SKC).

Finally, the new source scenarios were used to represent the emission sources, with the aim of improving the output of bioaerosol dispersion modelling.

Therefore, the purpose of this chapter was to model the bioaerosol concentrations from Ramsey composting site using the calculated emission rates depending on the bioaerosol data. Both the average emission rates and the real-time emission rates were used to model the bioaerosols concentrations downwind to the composting site.

Several studies have used models to predict downwind bioaerosol concentrations dispersed from the composting facilities. Millner et al. (1980) modelled the dispersion of *Aspergillus fumigatus* released from composting sewage sludge. This study concluded that under unstable atmospheric conditions, the bioaerosols could disperse 0.5 – 0.6 km downwind from the source before reaching background concentrations. Danneberg et al., (1997) also modelled the concentrations of *Aspergillus fumigatus* downwind of a composting plant. Danneberg et al., (1997) calculated an emission rate and was similar to the results from Millner et al. (1980). In addition, the conclusion of both studies was similar, which was that bioaerosol could travel 500 m before reaching the background concentrations of 500 CFU/m³.

Taha et al. (2007) modelled the bioaerosol from static windrows as a passive emission source, and agitation activities as an active emission source. Taha et al. (2007) used SCREEN3 and ADMS to estimate the emission rate depending on the measurements taken at source during static conditions and agitation activities, from the compost of different ages. The results added further weight to the conclusion of Taha *et al.*, (2006) which was the bioaerosol concentration from agitation activities is always higher than from static windrow. Furthermore, bioaerosol emissions from turning activity during the early stages may be higher than during the later stages of the composting process.

In this study, fluorescent particles concentrations from turning activity were modelled in addition to the traditional data which included bacteria and fungi concentrations to estimate the bioaerosol concentrations downwind of the composting site.

7.2 Method

Modelling methods and inputs, including the source type, source geometry, weather conditions and so on were described in chapter 6. However, for chapter 6, the sampling point was the only receptor point used, but for this modelling, there were additional receptor points. The purpose of this was to produce a depletion curve for each type of emissions including fluorescent particles

detected using the SIBS, and bacteria and fungi detected using the filtration technique. All data used in this modelling were from the second sampling visit to the Ramsey composting site, as this represented a single agitation activity (turning). Passive sources have also been modelled.

For active source data, a point source scenario was used to represent the turning activity. The passive source has been represented as an area source as used in the previous chapter and previous modelling studies such as Taha et al., (2007).

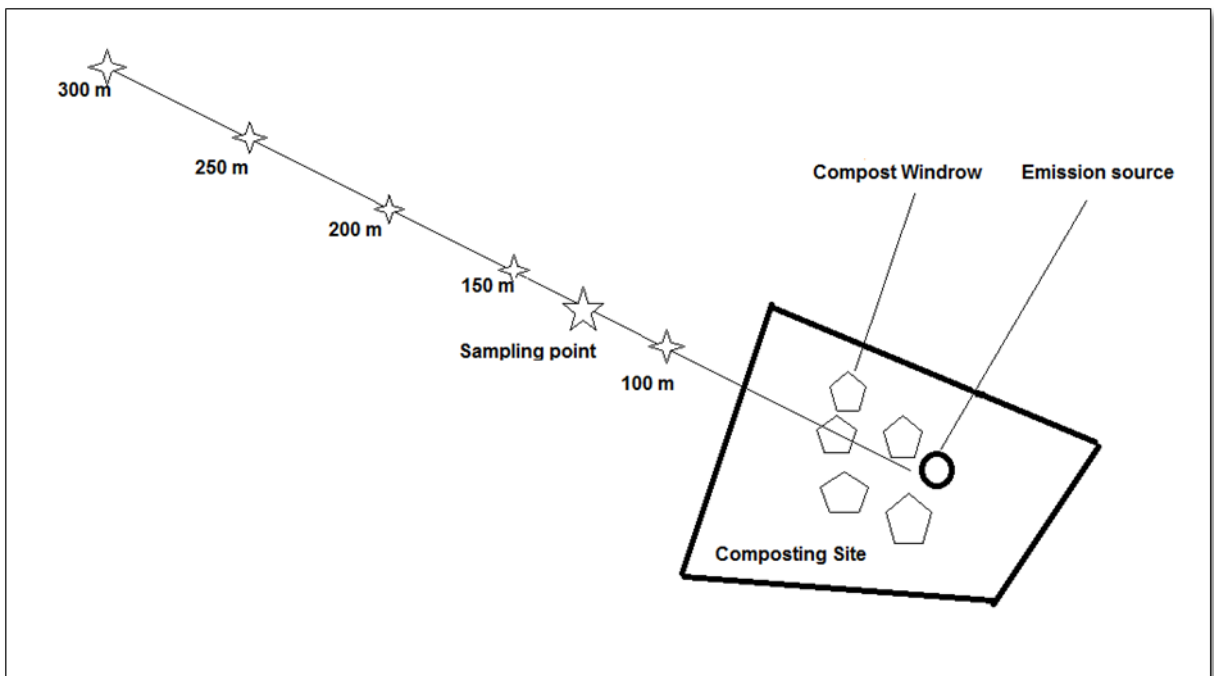


Figure 7-1: Composting site layout including the emission sources, sampling point and the distances of modelled concentration. For active source, a point scenario was used to model the emissions from turning activity, and for passive, an area scenario was used to model the emissions from compost windrows during the break where there was no agitation activity.

a-

Figure (7-1) shows the general layout of the composting site to illustrate the locations of the turning activity and compost windrow. As well as the sampling point, five downwind distances have been chosen to model the bioaerosol concentrations from the compost site, which were 100, 150, 200, 250, and 300 metres from the emission source.

7.3 Results

Figures (7-2 and 7-3) shows the real-time concentrations of fluorescent particles modelled at different distances downwind of the emission source. Figure (7-2) represents the emission concentrations during the turning activity. The results for the passive source, which was the compost windrow during the break (no activity) are presented in figure (7-3).

The concentrations of emissions were inversely proportioned with the distances. As the plume moves away from the source, the pollutants in the plume become more diluted as the plume expands laterally and vertically due to the effects of turbulence (Beychok, 1994).

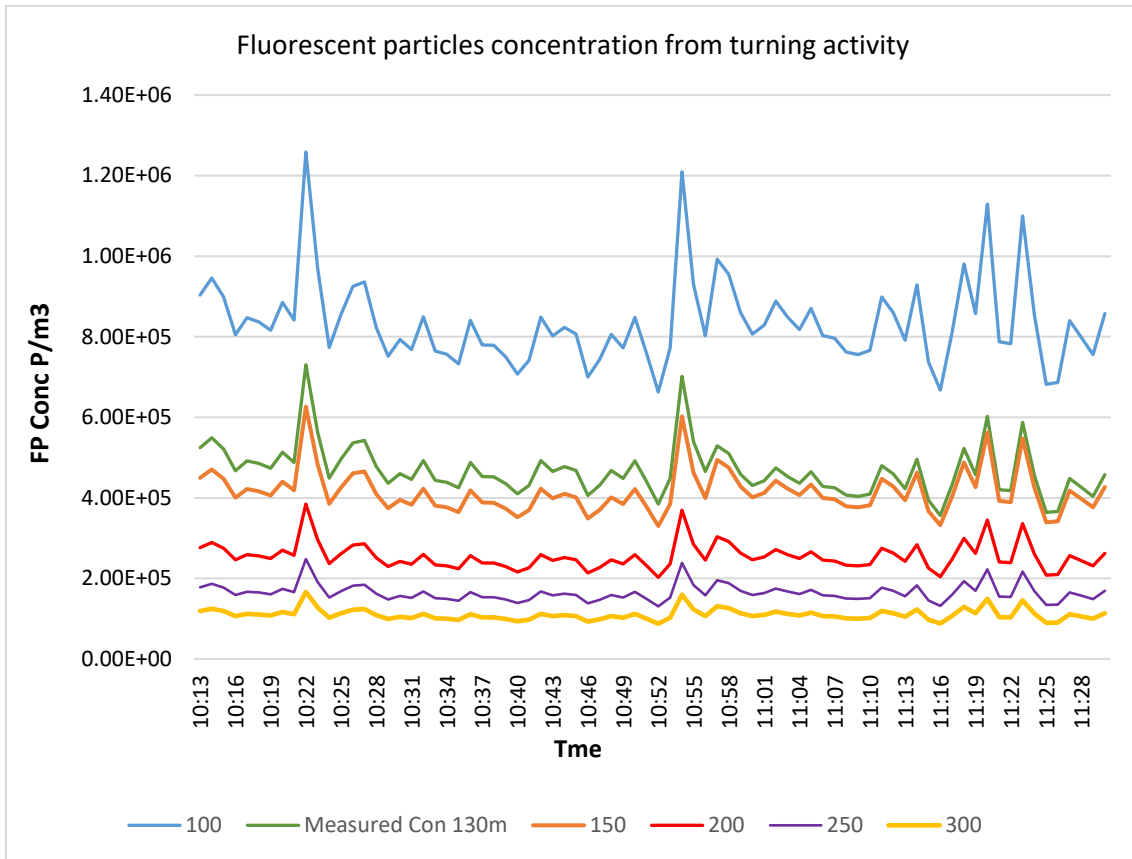


Figure 7-2: The real-time modelled concentration of fluorescent particles from turning activity at different distances downwind of the source. The 130 m distance is also the sampling location of the second sampling visit.

The results show how the bioaerosol emissions disperse from the composting site. The fluctuations in the emission concentrations show how the emission concentration is continuously changing. This continuous change is due to the weather conditions and the level of activity.

Generally, the pattern of the emission concentrations at all distances shows the same trend. In the modelled results, the correspondence appears to be identical regardless of the concentration levels. However, this is not correct, as the emissions need time to move from the source to a particular distance. As discussed in chapter 6, ADMS can only model the emission dispersion hourly, whereas the SIBS detects the emission minute by minute. The method described in chapter 6 was used here and is not able to tag the time lag for the

emissions to move downwind to the next receptor into account. There is a difference in the timing of the modelled concentration, and this difference depends on the distance and the wind speed. For these results, the delay is less than 1 minute between each modelled distance as the wind speed was about 3 metres/second.

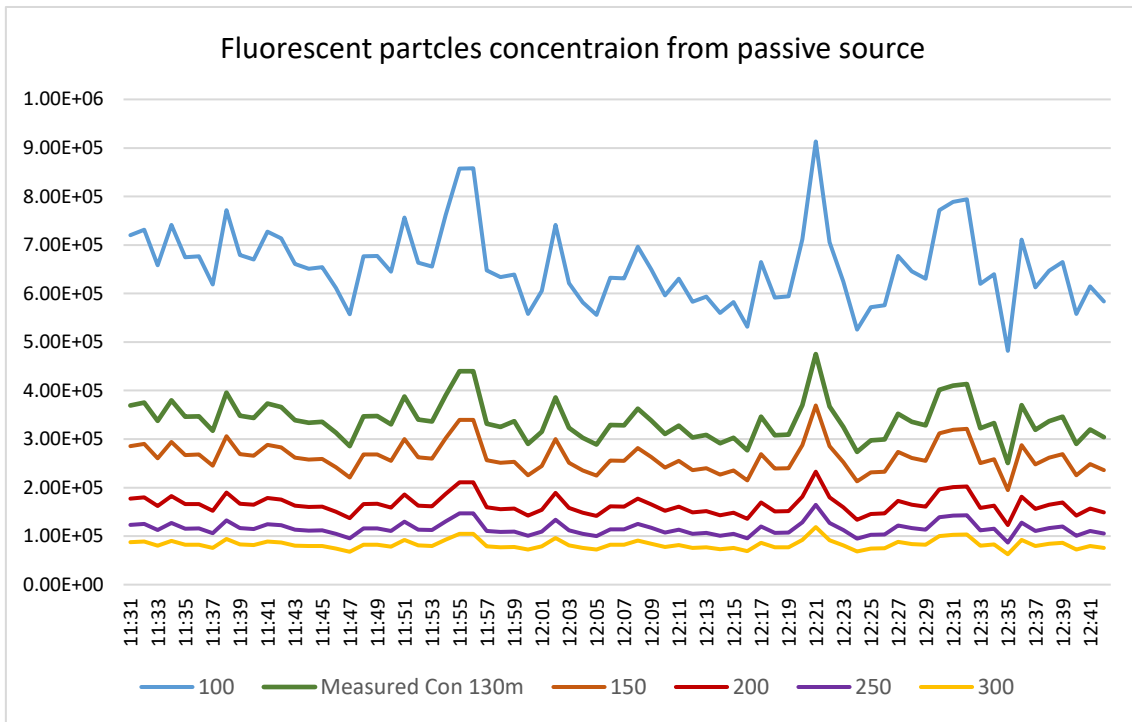


Figure 7-3: The real-time modelled concentration of fluorescent particles from compost windrows (passive source) at different distances downwind of the source.

Figure (7-3) shows the modelled concentration of fluorescent particles when there was no activity. As there was a difference between the concentrations monitored during the agitation activities and the break (no activity), so there was also a difference in the modelled concentrations at all distances between active and passive sources.

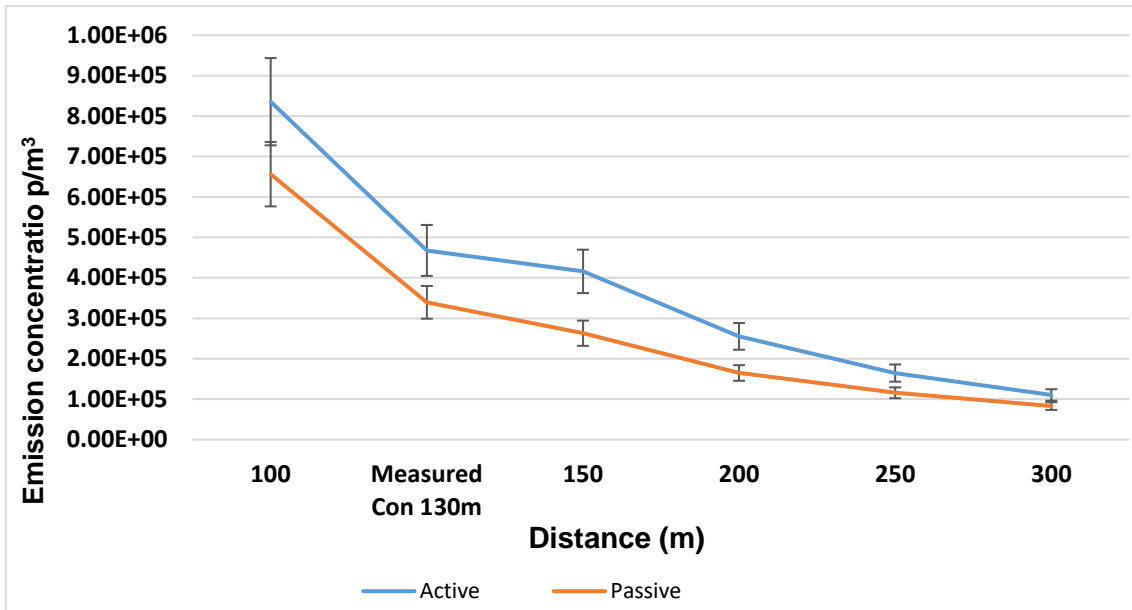


Figure 7-4: Depletion curves of fluorescent particle concentrations at downwind distances from the emission sources during active and passive emission sources.

Figure 7-4 shows the average concentrations of the fluorescent particles at different downwind points during the active and passive dispersions. The differences between the concentrations show the effect of the agitation activity on the concentration levels. Some interesting patterns emerge, for example, the fluorescent particles concentration during no activity at 200 metres was 1.65×10^5 FP/m³, which is the exact concentration during the active dispersion at 250 metres from the emission sources. This suggests that the emissions can disperse further during the agitation activities.

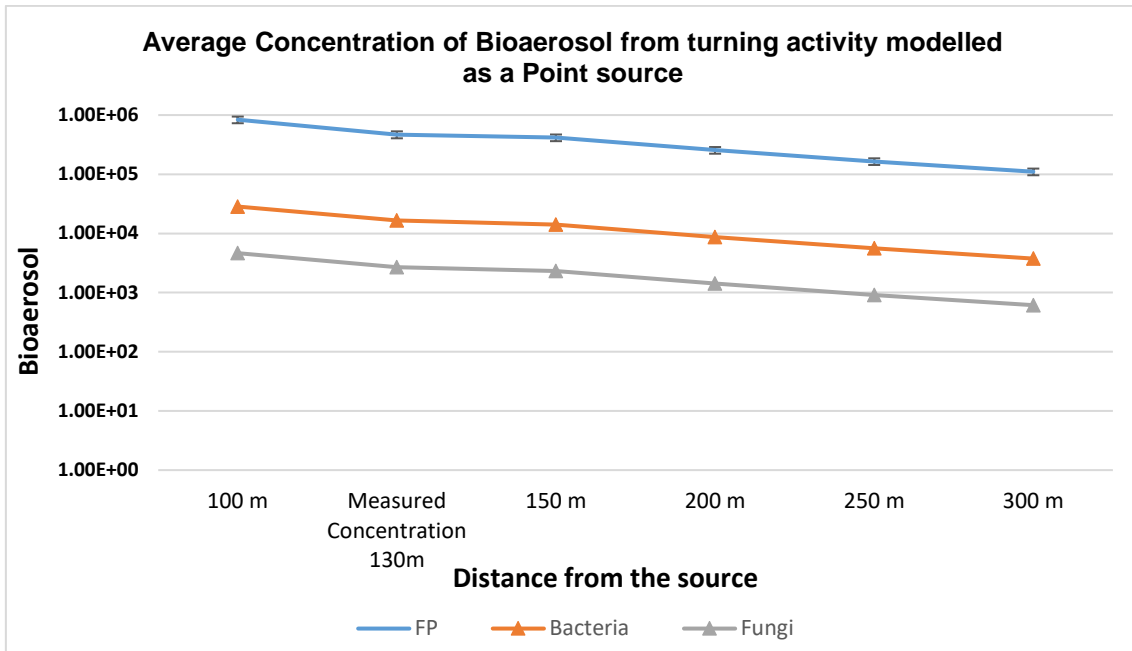


Figure 7-5: Modelled concentration of bioaerosols downwind of the emission source depending on the average emission rates of fluorescent particles and emission rates of bioaerosols detected using filtration sampling technique.

Figure (7-5) shows the modelled concentrations of different bioaerosol types including fluorescent particles, bacteria and fungi. The modelled concentrations of fluorescent particles were based on the average emission rate of the fluorescent particles calculated using the SIBS. At 250 metres from the source, all types of bioaerosols were higher than the trigger levels that are recommended by M9 (2018), which are 1000 and 500 CFU/m³ for total bacteria and *Aspergillus fumigatus*, although the data presented here is for total fungi and not just *A. fumigatus*. While not directly comparable, the fluorescent particles concentration was much higher than the trigger levels. The SIBS data represents a wide range of bioaerosol components including viable and non-viable bioaerosols, and the fluctuation in the SIBS data indicates that the bioaerosol concentration is constantly changing. As the average data are only snapshots, this suggests that the average data does not show the real picture of dispersion, where the real-time data from the SIBS is expected to be closer to reality.

7.4 Discussion

The significant difference between the results in figure (7-2) and (7-3) are the fluctuations. More details can be analysed with the real-time data and the fluctuations (such as peak to mean ratios, and outliers), which was not possible with the traditional data. These details can contribute to risk assessments of bioaerosols through determining the peak, lowest and epidemic emission concentration for any particular distance around the site.

As detailed in chapter 4, the fluorescent particles concentration collected from Ramsey composting site were always higher than the traditional bioaerosols concentrations that were collected at the same time. Therefore, the modelled concentrations of the fluorescent particles must be higher than the traditional bioaerosol concentrations.

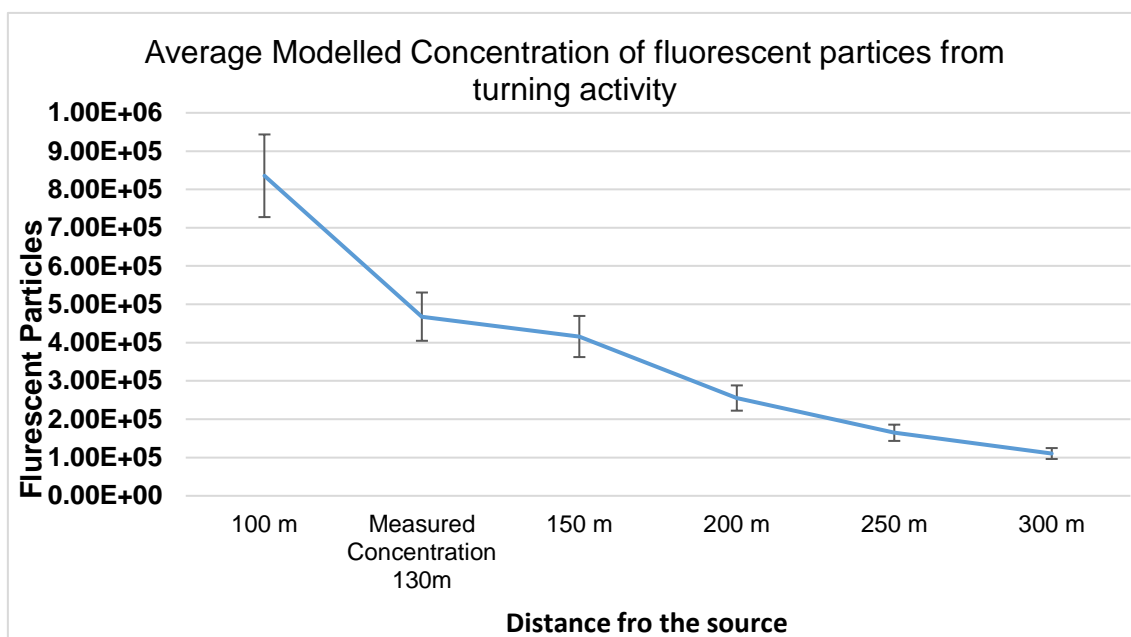


Figure 7-6: modelled average concentrations of fluorescent particles at different downwind distances. The standards deviation decrease with the increasing the distance from the source.

Another significant issue found from this modelling was the relationship between the distance and the fluctuations in the emission concentration. This relation may not be very noticeable in figures 7-2 and 7-3, but it is very clear in figure 7-5. Figure 7-6 shows the average concentrations at different distances downwind of the emission source, and it is clear that the standard deviation decreases with distance. Therefore the variations in the real-time concentrations decrease with distance, suggesting that the emission concentration decreases and homogenises during dispersion. Therefore, whenever the SIBS was closer to the source, it recorded more details about the emission concentrations.

Previously, dispersion modelling studies have showed the dispersion of bioaerosols in two dimensions only, namely the concentrations and the distances from the source (similarly to figures 7-5 and 7-6). Where the new emission rates calculated in this study using the SIBS real-time data, added a new dimension to the dispersion modelling, which is the time (figure 7-2 and 7-3). Many studies have modelled the dispersion of bioaerosol using data from traditional methods (Taha, 2005; Taha *et al.*, 2006, 2007; Douglas *et al.*, 2017; ADAS and SWICEB, 2005; Drew *et al.*, 2006; Tamer Vestlund, 2009; Douglas, 2013). However, they used different scenarios and different emission rates to model the dispersion of bioaerosols, whether from static compost windrows, agitated compost windrows or agitation activities. For example, Taha *et al.* (2006) modelled the dispersion of *A. fumigatus* from static compost windrows of varying geometry and load, and agitated compost windrows depending on bioaerosol data detected at 10 m distances from various compost processing activities. Douglas *et al.* (2017) also modelled the dispersion of *A. fumigatus* from composting activities. However, Douglas *et al.* (2017) calibrated and validated an existing numerical dispersion model applied to the emission of *A. fumigatus* from composting facilities, and found a good correlation and coincidence between predicted concentrations and measured data after validation.

7.5 Conclusion

This chapter has presented the results from using the SIBS data in the model for the first time to model both the average fluorescent particles and fluctuations in emissions. As with the earlier chapters focussing on sampling (chapters 4 and 5) at the composting site, improvements were made to the model output. This improvement was done using the emission rates of the fluorescent particles. These data provided new insights into the emission and dispersion from the composting site. The ADMS software needs further develop to be able to model the real-time data from the SIBS. As a first step, the SIBS data can improve the bioaerosols dispersion model input, but the ADMS itself cannot fully utilise the SIBS data.

7.5.1 Key findings

1. As the measured bioaerosol emissions from composting site using the SIBS provides more details about bioaerosol emission and dispersion, so modelling with these data also provides new insights and details about emission and dispersion. These details can contribute to improving bioaerosol risk assessment.
2. As the relationship between the emission concentration and the distance was reverse, the relation between the fluctuations of the concentration was also reverse. This means, when the distance increases, the emission concentration and the fluctuations reduce.
3. The ADMS software needs a significant update to fully model the SIBS data.

8 Final Outcomes

8.1 Reminder of the project aim

This project aimed to improve the sampling and dispersion modelling of bioaerosols from composting facilities. The purpose of this improvement is to increase the reliability and accuracy of the risk assessment of bioaerosols from composting facilities.

The improvements included using a novel sampling technique to detect a new type of bioaerosol data from open windrow composting facilities and determining the most appropriate methods to use these data in bioaerosol dispersion modelling.

The aim of this project was:

To examine to what extent a new sensor (the SIBS) can be useful to apply dispersion models more effectively to model bioaerosol dispersal in the open windrow composting environment.

The results presented in this research have enhanced knowledge on how bioaerosols are released and dispersed. Dispersion modelling plays an important role in the prediction of exposure of close receptors to bioaerosols. It is anticipated that these results will further support both risk assessments and development of Environment Agency policy, as well as aid in the planning of new composting facilities in the future. This aim was addressed by defining two objectives, developed to confront the gaps in knowledge associated with this subject field.

First Objective: Analyse bioaerosol concentrations measured using the SIBS alongside traditional sampling technique.

The project undertook a series of sampling experiments to measure the concentration of bioaerosol emissions at composting facilities and other environments using the SIBS and traditional sampling technique. The resulting data has been subjected to statistical analysis to compare the two methods.

The data from the SIBS, as was anticipated, have some advantages over the data from the traditional method, because of the ability of the SIBS to continuously sample and measure the concentration of living and non-living biological particles.

The objectives of this comparison were:

1- Quantifying and characterising the fluorescent particles from the composting facilities using the SIBS.

2- Analyse how SIBS data relates to data collected using traditional methods as the present knowledge is based on traditional methods. This new analysis helped to place the SIBS data within the context of previous findings.

Second Objective: Determine whether SIBS data can be used to improve bioaerosol dispersion modelling from composting facilities.

Currently, dispersion modelling of bioaerosols from composting facilities depends on the traditional measured data. These data represent a snapshot of bioaerosol concentrations where the composting sites emit the bioaerosol continuously and variably. Furthermore, the traditional data measures only the viable bioaerosol, where there are non-viable bioaerosols may also have harmful effects. The input data needs to be more accurate to improve model output accuracy. Therefore, improving the bioaerosol data may improve the model input and thus improve the model output. The new bioaerosols data measured by the SIBS was anticipated to be useful for modelling of bioaerosol emissions from composting sites.

This objective, therefore, used the database that was collected during the sampling campaigns to determine the most appropriate methods to incorporate the SIBS data into the ADMS dispersion model.

8.2 Key findings

Generally, the key findings of this thesis can be classified according to the main objectives, namely sampling and modelling findings:

8.2.1 Sampling key findings

- 1- The SIBS is more sensitive than the IOM/SKC technique. Therefore, it can sample at both high bioaerosol concentrations and at low concentrations as well, which was very clear when the composting activities were operating and even during no activity. In contrast, the filtration technique has a high lower limit of detection and other traditional techniques (such as impaction) may be prone to over-loading in high concentrations.
- 2- SIBS data show that the fluorescent particles concentration is much higher than bioaerosol concentration detected using traditional sampling technique, confirming that the actual emitted concentrations of bioaerosols are higher than the traditional techniques are capable of monitoring.
- 3- The SIBS is a suitable technique to study and analyse the emission concentrations, whether during the operating time of the composting activities or any other time.
- 4- The SIBS detects the fluctuations of bioaerosol concentration from composting facilities, which is not possible with the IOM or other traditional techniques.
- 5- The SIBS data showed the relationship between the bioaerosol dispersion and the weather conditions during the sampling time.

8.2.2 Modelling key findings

- 1- With the use of the real-time data, the range of the model output increased because the range of input has increased. Therefore, the

- probability of the “correct” answer being within the range of modelled outputs is higher. This provides greater confidence in the model outputs.
- 2- The calculated emission rates represent all biological particles as detected by the SIBS, whereas previous studies have only used culturable bioaerosol for modelling.
 - 3- The SIBS emission rates (emission rates of the fluorescent particles) were higher than the emission rates based on the traditional data from this and other studies.
 - 4- Real-time emission rates were calculated for the first time and will improve the use of bioaerosols dispersion modelling in risk assessments by being able to model the dispersion of bioaerosol emission for a long term.
 - 5- The SIBS data collected downwind of the emission sources can only be used to calculate the emission rates if there is one composting activity occurring, because if there are two activities or more, the data will be combined and cannot be associated to a specific emission source.
 - 6- As the relationship between the emission concentration and the distance was reverse, the relation between the fluctuations of the concentration was also reverse. This means, when the distance increases, the emission concentration and the fluctuations reduce.
 - 7- The ADMS software needs a significant update or adaptation to fully capitalize on information provided by the SIBS data.
 - 8- In order to accurately model bioaerosol emissions from composting sites, real-time bioaerosol concentration should be known and bioaerosol particles in ADMS need to be correctly defined.

8.3 Contribution

The research in this thesis presents a significant contribution to knowledge regarding improving the understanding of the characterisation and dispersal of bioaerosols emitted from composting facilities. As discussed before, the understanding of factors that affect bioaerosol behaviour at source, pathway

and receptor is essential to the risk assessment of bioaerosol from composting facilities.

However, there are gaps in knowledge which may affect the analysis of bioaerosols emitted from composting facilities. This is the first study that has used the new biosensor (the SIBS) to detect the real-time concentration of the bioaerosol emissions from compost agitation activities. This sampling technique was used to generate novel data to characterise bioaerosols emitted from composting sites. This novel data showed the relationship between the bioaerosol concentration and the agitation activities, and also showed the effects of weather conditions on the dispersion and concentration of bioaerosol. Furthermore, these data were compared with traditional bioaerosol data detected using the filtration sampling technique at the same time.

As detailed before, the traditional sampling technique, such as filtration technique, can provide only snapshot data of bioaerosol concentration. This data does not show the nature of the bioaerosol dispersion which the SIBS data has clearly shown that the concentration of bioaerosol from agitation activities is always fluctuating and is not static.

In the modelling, new scenarios for source geometries were used to model the dispersion of bioaerosol from composting site. The novel data were also used to improve the modelling input by calculating the emission rates of bioaerosol from the composting site. This has the potential to provide a more continual indication of emissions spatially and temporally. Although the ADMS could not model the emission dispersion using the minute averages, the method that was used in chapter 7 has shown the potential of the SIBS data and how it could be useful within risk assessments of bioaerosol from composting facilities.

However, improving the ADMS software for this purpose will make a quantum leap in dispersion modelling of bioaerosol from composting facilities. So that, this is the first library of such data generated for compost related bioaerosols. In addition to the novel data, new scenarios of source geometries were used to model the dispersion of bioaerosol from composting site. Finally, a set of

recommendations have been provided for more improvement to the bioaerosol dispersion modelling based on the SIBS.

In conclusion, the results achieved by this research have presented new insights into the characterisation and dispersal of bioaerosols emitted from composting facilities. As such, these new insights would be anticipated to make an important contribution to the needs of new and existing composting facilities in quantifying bioaerosol site exposures to meet increased regulatory requirements, as well as the needs of the regulatory body, the Environment Agency, in evaluating the exposure risks of bioaerosols from composting facilities.

Therefore, the aim of this project, “To examine to what extent a new sensor (the SIBS) can be useful to apply dispersion models more effectively to model bioaerosol dispersal in the open windrow composting environment” has been achieved.

8.4 Recommendations

The limitations highlighted above can be addressed by following the suggestions listed below:

- 1- Although the SIBS can detect bioaerosol emissions based on the fluorescence of the biological particles, there is still ambiguity about the exact nature of all the particles that are detected by the SIBS. Further experimental studies should be undertaken to clarify what particles the SIBS detects and under which circumstances. This experiment should include a series of tests with different kinds of source emission in the controlled environment; and will show the ability of the SIBS to detect both biological and non-biological particles.
- 2- Using more than one SIBS during sampling such as using several SIBSs simultaneously at locations including upwind, at the source, at sensitive receptors and downwind during different times such as daytime, night etc. This will detect the real dispersion of bioaerosol and will also

contribute to validating the prediction of bioaerosol dispersion using the model.

- 3- Source representation: there are some source scenarios that can be used to represent the emission source in the model. To represent the turning activity as an emission source; point, area and line scenarios can be used. Additionally, two points scenario can also be used, but this scenario still needs further improvement, such as detecting the variability between both points because the emissions from these points are unlikely to have the same pattern or concentrations. For screening activity, the line scenario may be suitable as illustrated above, but this scenario has not been validated yet due to a lack data representing the screening activity only. Currently, the point source scenario is the best option to represent the shredding source. The emission that is emitted from the shredding machine is emitted from known gap. However, this needs further to improve and validate the scenarios for shredding activity and which requires more data detected at source.
- 4- Emission rate: in this study, three kinds of emission rates have been calculated depending on the measured data at the composting site which were fluorescent particles, bacteria and fungi. For all bioaerosol types, the emission rates represented one composting activity, which was turning. However, the emission rates of fluorescent particles for turning activity, that can be used in the model, are 8.08×10^7 (P/m/s) and 6.54×10^9 (P/s) for line and point source scenarios respectively. The emission rate of bacteria from turning activity is 2.48×10^6 (CFU/m/s) and 1.88×10^8 (CFU /m/s) for line and point scenarios respectively. Finally, the emission rate of fungi for turning activity are 4.04×10^5 (CFU /m/s) and 3.07×10^7 (CFU /s) for line and point scenarios respectively.
- 5- Sampling using two SIBS (one at source and one downwind) would provide data to calculate true source emission rates and to compare the measured concentration with the modelled concentration. This

comparison will validate the method of emission rate calculation and validate the model as well.

- 6- Model the dispersion of bioaerosol from each activity individually, and from all sources as well. This will show the effects of the composting site on the surrounding environment generally and on the sensitive receptors specifically.

8.5 Further research

As a result of all results, conclusions, key findings and limitations, further research needs to focus on the following points to further improvement the sampling and modelling of bioaerosols from composting facilities:

- 1- The relationship between the weather conditions, seasons and the concentration of bioaerosol using the SIBS.
- 2- The depletion curve of bioaerosol concentrations from the compost windrow starting with the highest concentration during the agitation action and ending with the lowest concentration when leaving the windrow.
- 3- Calculate the emission rate of fluorescent particles from composting activities using fluorescent particles concentration detected at the source. This will be useful to compare the direct calculation and back-calculation methods.

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