

**EVALUATION OF BIOAEROSOLS AT LLANDDULAS COMPOSTING FACILITY,
LLANDDULAS, CONWY – 6th SEPTEMBER 2016**

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1. BACKGROUND & SITE DESCRIPTION:

D&F Associates Ltd has been commissioned by FCC Environment (FCC) to evaluate bioaerosols at Llanddulas Composting Facility, situated at Llanddulas Landfill Site, Abergele Road, Conwy LL22 8PH. Located in a former limestone quarry, composting activities are carried out on a dedicated composting pad situated in the Southwest quadrant.

Quarry walls (~19m high) to the South and West provide partial shelter to the composting area. The pad has designated areas for receiving and sorting green waste, shredding, open windrows, product maturation (as open windrows), screening and product storage.

The nearest sensitive receptors to composting activities are Plas Farm (~125m West) and two residential dwellings situated at Plas yn Lysfaen (~175m and 190m West).

2. SAMPLING POINTS & SAMPLING PROTOCOL:

The Environment Agency Technical Guidance Note M17 (Monitoring of particulate matter in ambient air around waste facilities, version 2, 2013) offers guidance about aerosol emissions from composting sites.

The Association for Organic Recycling (AfOR) has published guidance on the evaluation of bioaerosols at open composting facilities ('A standardised protocol for the monitoring of bioaerosols at open composting facilities', 2009). The Protocol aims to aid the assessment of the risk and impact that an open windrow composting system may have on the level of naturally occurring airborne micro-organisms. The Protocol cannot be used to assess the occupational exposure of workers who may be engaged in composting activities and it cannot determine if a composting facility presents a risk to the health of people who may live or work near to a site. This is because satisfactory dose-response data is not available.

M17 and the 2009 AfOR Protocol have been used for this assessment as applicable. The Filter Method given in the 2009 AfOR protocol has been used to evaluate bioaerosol levels and deviations approved by the AfOR Protocol Steering Committee are presented in the Summary Table attached as Appendix 2 (Page 10).

2.1 Samples

Sample locations were chosen in accordance with the guidelines given in the 2009 AfOR Protocol. Sample points are dependent on weather conditions and are chosen with respect to the direction of the wind on the day of monitoring. Sample points may therefore be expected to change for each assessment.

The following sample points were used for the evaluation at Llanddulas. Access considerations and changeable winds influenced the positioning of both the downwind and upwind sampling locations and sampling at 75-150m downwind and 25-50m upwind was not possible. Sample locations and mean wind directions are illustrated on the site map attached as Appendix 1 (Page 9):

- Sample Point 1:** Sensitive Receptor (in field adjacent to Plas Farm, 75-150m West of the site boundary). Grid reference SH 90100 77166 (SP1).
- Sample Point 2:** Downwind (200-300m Northeast of the pad boundary on the track that surrounds the quarry surface). Grid reference SH 90443 77275 (SP2).
- Sample Point 3:** Upwind (100-150m South of the composting area on the track that surrounds the quarry surface). Grid reference SH 90305 77015 (SP3).

2.2 Sampling

Sampling was carried out using SKC AirChek XR5000 personal air sampling pumps and SKC Button Aerosol Samplers containing sterile 25mm 0.8µm polycarbonate filters. The units were calibrated to operate at 2 L/min using a float rotameter with a linear flowpath of 600-5000 ml/min (SKC 393-0650). Three samples were taken in parallel at each sampling location. The sampling heads were mounted on a tripod 1.6 metres high and spaced approximately 3.0-5.0 cm apart in a horizontal hanging position. The pumps were programmed to run for 30 minutes and the sampling start and stop times were recorded. Exposed filters were aseptically removed from the sampling head using sterile tweezers and they were placed into 10ml aliquots of sterile buffered saline water containing Tween 80 (0.9% NaCl and 0.01% Tween 80, buffered to pH 7.5). Two unexposed filters were aseptically transferred into 10ml aliquots of sterile buffered saline solution to form control samples. An electrically controlled coolbox was used to transport the samples to the laboratory at 4°C.

2.3 Laboratory Enumeration

The samples were shaken and allowed to stand for 10-15 minutes in order to equilibrate to room temperature. A 10-fold series of dilutions were prepared from the filter suspensions. 0.5ml aliquots were plated onto prepared agar plates containing half strength Nutrient Agar (NA for the enumeration of mesophilic bacteria) and Malt Extract Agar (MEA for the enumeration of *Aspergillus fumigatus*).

Plates to evaluate mesophilic bacteria were inverted and incubated at 37°C. Plates to evaluate *Aspergillus fumigatus* were inverted and incubated at 40°C. All plates were counted after 48 hours. NA and MEA plates showing fewer than 100 colonies were returned to their respective incubators. Growth for *Aspergillus fumigatus* was rechecked after 3-days and growth for mesophilic bacteria was rechecked after 5-days.

Microbiological growth media was prepared in accordance with BS EN ISO 11133:2014: Microbiology of food, animal feed and water, preparation, production, storage and performance testing of culture media. Following sterilisation and cooling to 47°C, MEA was treated with Penicillin G Sodium Salt (20 units/ml) and Streptomycin sulphate (40 units/ml), and NA was supplemented with 0.01% w/v cycloheximide (dissolved in <2.0ml acetone). Prepared plates were pre-incubated and quality checked before use.

2.4 Calculation of Results

Exposed filters placed into 10ml aliquots of sterile buffered saline water containing Tween 80 produce microbial suspensions in which the micro-organisms have been diluted by a factor of 10 (i.e. 1:10 or 10^{-1}). For each subsequent dilution step, the micro-organisms are further diluted by a factor of 10 (i.e. 1:100 or 10^{-2} , 1:1000 or 10^{-3} , 1:10000 or 10^{-4} , etc). Serial dilution allows the number of viable cells in a microbial suspension to be quantified as colony forming units per millilitre (cfu/ml).

The number of micro-organisms in 60 litres of air (pump rate of 2 L/min x 30min sampling period) was obtained by multiplying the average number of micro-organisms in 1.0ml of the original suspension by 10. The number of micro-organisms in one cubic metre of air (m^3) was obtained by multiplying the number of micro-organisms determined in 60 litres of air by 16.667.

The minimum level of detection for mesophilic bacteria and *Aspergillus fumigatus* is 167 cfu/ m^3 . Concentrations that were found to be below the minimum level of detection are given as <167 cfu/ m^3 (i.e. less than 167 colony forming units per cubic metre of air).

2.5 Other Considerations

Equipment, consumables and microbiological growth media were assessed in the laboratory for quality assurance before use or release. Results and records of QA and QC checks are maintained as part of Good Laboratory Practice procedures. Industrial Methylated Spirits (IMS) was used to sterilise equipment in the field and 'Control Samples' were collected as part of the assessment to confirm aseptic handling. Procedures are followed to prevent external contamination.

3. METEOROLOGICAL/ WEATHER CONDITIONS:

Wind speed and direction, air temperature and relative humidity were logged at 1-minute intervals using a Kestrel 4500 portable weather station.

4. SITE ACTIVITIES:

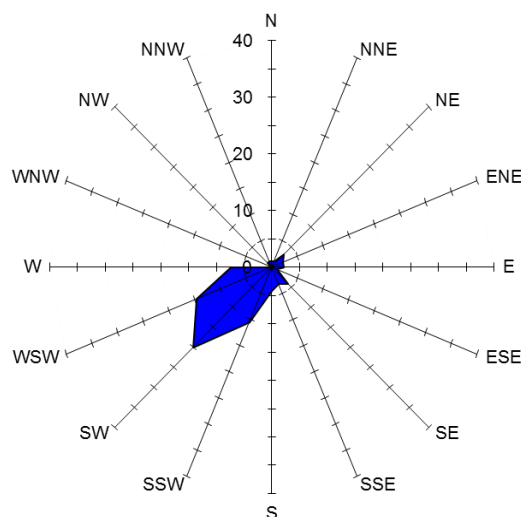
Material reception, material movement and screening occurred during sampling at SP1 (Sensitive Receptor) and SP2 (Downwind). Material reception and material movement occurred during sampling at SP3 (Upwind).

5. RESULTS OF EVALUATION:

5.1 Meteorological Information

Meteorological data recorded at each sample point is summarised in the Meteorological Conditions Table (Page 6). The wind direction at the surface of the quarry was variable with wind swirling recorded. The mean wind direction for the assessment was calculated from 224° (Southwest). Winds were recorded from North, North Northeast, Northeast, East Northeast, East, East Southeast, Southeast, South Southeast, South, South Southwest, Southwest, West Southwest, West, Northwest and North Northwest (Figure 1, Page 4).

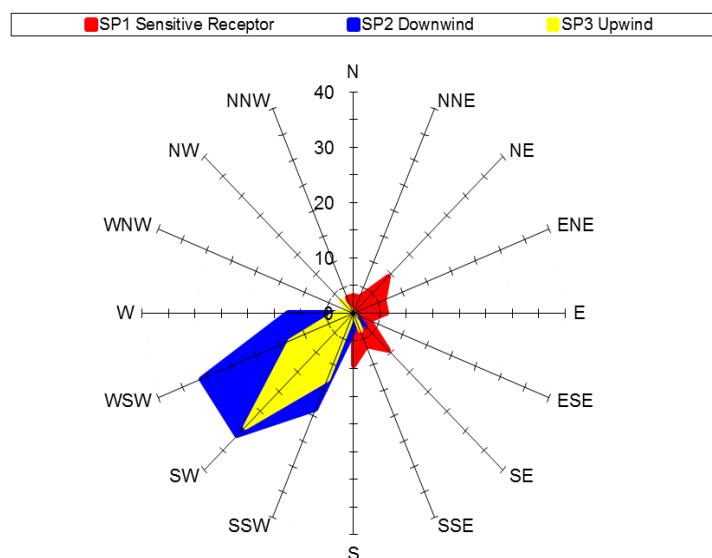
Figure 1: Wind Rose Illustrating the Mean Wind Direction from the Southwest.



Note: Units in Percentage Contribution. Wind speed ranged from 0.0-5.4m/s.

Figure 2 illustrates the wind patterns experienced at each sample location during the sampling periods. The mean wind bearings given in the Meteorological Conditions Table have been calculated using the AfOR method described on Page 30 of the Protocol. However, the mean wind bearing at each sample point may not reflect the bearing calculated for the whole of the assessment. It should also be noted that the mean bearing calculated for the assessment may be different than the predominant wind direction. This can be due to the influence of landscape, topography and nearby structures on air movement at the sample location and this should be considered if wind direction patterns at different sampling locations are compared. The calculated mean wind directions are illustrated on the site map attached as Appendix 1 (Page 9).

Figure 2: Wind Rose Illustrating Wind Patterns at Sample Locations SP1, SP2 and SP3.



The mean wind direction at SP1 (Sensitive Receptor) was calculated from 136° (Southeast). Changeable winds were observed with influences recorded from North, North Northeast, Northeast, East Northeast, East, East Southeast, Southeast, South Southeast, South, West and North Northwest. Wind speeds were measured within the range 0.0-2.2 m/s with an average wind speed of 0.6 m/s calculated.

The mean wind direction at SP2 (Downwind) was calculated from 227° (Southwest). Winds were recorded from Southeast, South, South Southwest, Southwest, West Southwest and West. Wind speeds were measured within the range 1.0-5.4 m/s with an average wind speed of 3.0 m/s calculated.

The mean wind direction at SP3 (Upwind) was calculated from 224° (Southwest). Changeable winds were recorded from South Southeast, South Southwest, Southwest, West Southwest, West and Northwest. Wind speeds were measured within the range 0.0-1.2 m/s with an average wind speed of 0.4 m/s calculated.

5.2 Results for Airborne Micro-organisms:

5.2.1 Current Limits (based on Open Windrow Composting Systems)

The Agency considers acceptable bioaerosol levels as;

- i. Those before the start of the composting process, or,
- ii. Bioaerosol levels that are no greater than 1,000 cfu/m³ for total bacteria, 500 cfu/m³ for the thermophilic fungus *Aspergillus fumigatus* and 300 cfu/m³ for Gram negative bacteria.

5.2.2 Microbiological Results

The microbiological results obtained for this assessment are given in the Estimated Concentrations of Airborne Micro-organisms Tables (Pages 7-8). Unless otherwise stated, the following results discuss colony forming units per cubic metre of air (cfu/m³) as calculated average means.

Sensitive Receptor (SP1)

Sampling was undertaken in a field adjacent to Plas Farm, 75-150m West of the site boundary. Due to changeable winds SP1 was both upwind and downwind of the composting and landfill areas during the sampling period. Mesophilic bacteria and *Aspergillus fumigatus* were each estimated at less than 167 cfu/m³.

Downwind (SP2)

Sampling was undertaken 200-300m Northeast of the composting pad on the track that surrounds the quarry surface. SP2 was downwind of composting operations during the sampling period. Mesophilic bacteria were estimated at 333 cfu/m³ and *Aspergillus fumigatus* at less than 389 cfu/m³.

Upwind (SP3)

Sampling was undertaken 100-150m South of the composting pad on the track that surrounds the quarry surface. SP3 was upwind of the composting area during the sampling period. Mesophilic bacteria were estimated at less than 333 cfu/m³ and *Aspergillus fumigatus* at less than 167 cfu/m³.

6. CONCLUSIONS & DISCUSSION:

1. Mean results for mesophilic bacteria and *Aspergillus fumigatus* were below EA guidance levels at Plas Farm, 200-300m downwind and 100-150m upwind of composting activities (*i.e.* at SP1, SP2 and SP3).
2. Plas Farm was both upwind and downwind of composting operations due to changeable winds during the sampling period.
3. The results suggest that the risk of exposure at Plas Farm and at Plas yn Lysfaen to composting bioaerosols was likely to be very low.
4. The mean wind direction for the evaluation was calculated from the Southwest.

7. SUMMARY:

The evaluation undertaken at Llanddulas Composting Facility on 6th September 2016 completed an appraisal of composting bioaerosols during material reception and screening activities.

Mean results for mesophilic bacteria and *Aspergillus fumigatus* were calculated below EA guidance levels at Plas Farm, 200-300m downwind of composting operations and 100-150m upwind of the composting area. Access considerations and changeable winds influenced the positioning of both the downwind and upwind sampling locations and sampling at 75-150m downwind and 25-50m upwind was not possible.

The mean wind direction for the assessment was calculated from the Southwest and due to changes in the wind direction Plas Farm was both upwind and downwind of composting operations. However, the results suggest that the risk of exposure at Plas Farm and at Plas yn Lysfaen to composting bioaerosols was likely to be very low.

METEOROLOGICAL CONDITIONS

Site:		Llanddulas Composting Facility			Site Operator:		FCC Environment	
Sampling Date:		6 th September 2016			Commissioning Laboratory:		D&F Associates Ltd	
Estimated Mass of Materials:		5,000-10,000 Tonnes			Type of Material Processed:		Green Waste	
Location	Sample Reference Number	Bearing of samplers from boundary of operational area (° from true north) Grid Reference	Mean direction the wind blows from during the sampling period (° from true north)	Difference in bearing between location of samplers from boundary/ source and mean direction of wind (°)	Mean wind speed during sampling (m/s)	Arithmetic mean of air temperature (°C)	Estimated relative humidity (%)	Prevailing weather conditions
Sensitive Receptor	SP1	40-120 (not visible) SH 90100 77166	136 (SE)	16-96	0.6 (Range 0.0-2.2)	21.2	82.1	Cloud cover = 8/10 Dry and bright.
Downwind	SP2	200-260 SH 90443 77275	227 (SW)	0	3.0 (Range 1.0-5.4)	20.0	84.2	Cloud cover = 9/10 Dry and dull.
Upwind	SP3	310-20 SH 90305 77015	224 (SW)	86-156	0.4 (Range 0.0-1.2)	21.8	80.4	Cloud cover = 9/10 Dry and dull.

N/A = Not Available

ESTIMATED CONCENTRATIONS OF AIRBORNE MICRO-ORGANISMS										
Site:			Llanddulas Composting Facility			Site Operator:			FCC Environment	
Sampling Date:			6 th September 2016			Commissioning Laboratory:			D&F Associates Ltd	
Estimated Mass of Materials:			5,000-10,000 Tonnes			Type of Material Processed:			Green Waste	
Location	Sample Reference Number	Distance From Operation (Metres)	Sample Volume (Litres)	Microbial Type	Site Activity	Materials Processed	Calculated Concentration of Airborne Micro-organisms (cfu/60 L)	Calculated Concentration of Airborne Micro-organisms (cfu/m ³)	Calculated Mean of Samples (cfu/m ³)	Comments
Sensitive Receptor SP1 (11:01:55-11:31:56 hrs)	1A 1B 1C	75-150 (Boundary)	60	MB	Material Reception, Material Movement & Screening	Green Waste	10 <10 <10	167 <167 <167	<167	Mean results for mesophilic bacteria and <i>Aspergillus fumigatus</i> were below EA guidance levels. Sampling was undertaken in a field adjacent to Plas Farm. SP1 was both upwind and downwind of the composting and landfill areas due to changeable winds. The mean wind direction was calculated from 136° (SE).
	1D 1E 1F			AF			<10 <10 <10	<167 <167 <167	<167	
Downwind SP2 (12:09:58-12:39:59 hrs)	2A 2B 2C	200-300 (Boundary)	60	MB	Material Reception, Material Movement & Screening	Green Waste	30 10 20	500 167 333	333	Mean results for mesophilic bacteria and <i>Aspergillus fumigatus</i> were below EA guidance levels. Sampling was undertaken Northeast of the pad on the track that surrounds the quarry surface. SP2 was downwind of composting operations. The mean wind direction was calculated from 227° (SW).
	2D 2E 2F			AF			<10 <10 50	<167 <167 833	<389	

Controls = <10 cfu per filter

MB - Mesophilic Bacteria

AF - *Aspergillus fumigatus*

ND - Not Determined

TNTC - Too Numerous To Count

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ESTIMATED CONCENTRATIONS OF AIRBORNE MICRO-ORGANISMS										
Site:			Llanddulas Composting Facility			Site Operator:			FCC Environment	
Sampling Date:			6 th September 2016			Commissioning Laboratory:			D&F Associates Ltd	
Estimated Mass of Materials:			5,000-10,000 Tonnes			Type of Material Processed:			Green Waste	
Location	Sample Reference Number	Distance From Operation (Metres)	Sample Volume (Litres)	Microbial Type	Site Activity	Materials Processed	Calculated Concentration of Airborne Micro-organisms (cfu/60 L)	Calculated Concentration of Airborne Micro-organisms (cfu/m ³)	Calculated Mean of Samples (cfu/m ³)	Comments
Upwind SP3 (12:52:03-13:22:04 hrs)	3A 3B 3C	100-150 (Boundary)	60	MB	Material Reception & Material Movement	Green Waste	20 30 <10	333 500 <167	<333	Mean results for mesophilic bacteria and <i>Aspergillus fumigatus</i> were below EA guidance levels. Sampling was undertaken South of the pad on the track that surrounds the quarry surface. SP3 was upwind of the composting area. The mean wind direction was calculated from 224° (SW). Changeable winds were recorded.
	3D 3E 3F			AF			<10 10 <10	<167 167 <167	<167	

Controls = <10 cfu per filter

MB - Mesophilic Bacteria

AF - *Aspergillus fumigatus*

ND - Not Determined

TNTC - Too Numerous To Count

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Sample Points Evaluated at Llanddulas Composting Facility on 6th September 2016



Mean Wind Direction at Sample Point



Indicates Compost Pad Boundary



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Summary of Approved Deviations/Amendments for the Filter Method Described in the 2009 AFOR Protocol

Deviation/Amendment	Reason for the Deviation/ Amendment
5.5. Quality assurance.	All equipment, consumables and microbiological growth media was checked for sterility in the laboratory before use or release. Results and records of QC checks are maintained. Equipment may be swabbed in the field using IMS and additional field controls are collected to assess aseptic handling. Procedures are followed to prevent external contamination. Impact on the results: None
5.6. Summary of equipment.	SKC Button Aerosol sampling heads were used to house the filters instead of IOM sampling heads. The sampler is a patented reusable filter holder with a porous curved surface sampling inlet. SKC claim that the sampler improves the collection of bioaerosols for viable or non-viable analysis whilst minimising wind sensitivity, and provides equal distribution of particle loading and low intersample variation. The sampler follows the ACGIH/ISO sampling criteria. Impact on the results: The porous surface minimises the collection of particles >100µm which may be projected into the IOM sampler. Bioaerosols attached to particulate matter >100µm will not be assessed. Particles >100µm are thought to be less inhalable and bioaerosol levels may therefore be lower than those obtained using IOM samplers. The sample is less likely to be affected by wind direction variations at the sample location during the sampling period.
6.2. Filters stored in separate resealable bags.	Individual filter membranes were wrapped and sterilised in tinfoil pouches. QC and sterility checks were carried out on the sterilised batch of filters before use (5.5) Impact on the results: None
6.2. Transportation of loaded filters to the laboratory.	Exposed filters were aseptically removed from the filter holder using sterile tweezers on site. The filters were placed into 10ml aliquots of sterile buffered saline water containing Tween 80 to prevent osmosis. Two unexposed filters were also aseptically placed in 10ml aliquots of sterile buffered saline water on site to i) demonstrate aseptic handling, and ii) to create blank controls. Impact on the results: Micro-organism quality and recovery is safeguarded during the storage and transportation of exposed filters as long as the filter is not exposed to other contamination sources. Good aseptic handling is critical.
6.2. Initial suspension of the impacted filter into 5ml of sterilised physiological saline solution.	The sterilised physiological saline solution is chemically the same as the buffered saline water used by D&F but the D&F solution is buffered to pH 7.5. The impacted filters were placed into 10mls of solution instead of 5mls. The resulting suspension was plated out directly and this allowed the minimum limit of detection to be reduced from 417 cfu/m ³ (AFOR) to 167 cfu/m ³ (in 60L of air). Impact on the results: Improved detection level and reduced calculation errors due to the multiplication factor that is applied.
6.2. Filters shaken at 35°C to 40°C for 15-minutes.	The filter suspensions were allowed to stand at room temperature for 10-15 minutes to allow them to equilibrate. This was to reduce the effects of thermal shock on the microbes following transportation at 4°C. The samples were shaken but they were not heat-treated at 35-40°C prior to plating out. The total number of viable bacteria in the mesophilic temperature zone (20-45°C) including the recovery of stressed micro-organisms were therefore assessed following standard incubation at 37°C. Impact on the results: Heating the sample above 37°C will adversely affect the recovery of micro-organisms that are grown at 37°C.
6.2. 0.1ml of each respective step is plated on to culture medium with a pipette and spread out by circular movements.	0.5 ml of each dilution (including the original suspension) was plated onto culture media and the bacterial suspension was gently spread across the surface of the medium using a sterile L-shaped spreader. This is a traditional microbiological technique used for the cultivation of micro-organisms. The inoculated plates were left for a few minutes to dry prior to incubation. Impact on the results: Plating 0.5ml instead of 0.1ml reduces the minimum level of detection.
11.1. Data calculation for filters.	The results were calculated as described in section 2.5 of this report.
Additional:	