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1           **Exposure to Bioaerosols at Open Dumpsites: A Case study of bioaerosols exposure**  
2           **from activities at Olusosun Open Dumpsite, Lagos Nigeria.**

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6                           **Abstract**

7           Activities associated with the open dumping of municipal solid waste has the potential to have  
8           greater impacts on the environment and public health compared to other forms of waste-to-land  
9           treatment of such wastes. However, there is a lack of quantitative data on the exposure to  
10          bioaerosols from open dumpsites, hence impeding the development of effective interventions  
11          that would reduce the risk of respiratory symptoms among scavengers and waste workers at  
12          such dumpsites. This study investigated exposure to bioaerosols at Olusosun open dumpsite,  
13          Lagos Nigeria using three methodologies; (1) Conducting a cross-sectional survey on the  
14          respiratory health of the population on the dumpsite, (2) Measuring bioaerosol concentrations  
15          in the ambient air by measuring four bioaerosols indicator groups (total bacteria, gram-negative  
16          bacteria, *Aspergillus fumigatus* and total fungi) using a Anderson six stage impactor sampler,  
17          (3) Measuring activity related exposures to bioaerosols using an SKC button personal sampler.

18          After a cross sectional health survey of 149 participants (waste workers, scavengers,  
19          middlemen, food vendors and business owners), smokers reported higher symptoms of chronic  
20          cough (21%) and chronic phlegm (15%) compared to non-smokers (chronic cough 15%,  
21          chronic phlegm 13%) . Years of work > 5 years showed no statistically significant association  
22          with chronic phlegm (OR 1.2, 95% CI 0.4-3.4; p>0.05) or asthma (OR 1.8, 95% CI 0.6-5.2;  
23          p>0.05). At the 95<sup>th</sup> percentile, the concentration of gram-negative bacteria was the highest  
24          (2188 CFU/m<sup>3</sup>), then total bacteria (2189 CFU/m<sup>3</sup>), total fungi (843 CFU/m<sup>3</sup>) and *Aspergillus*  
25          *fumigatus* (441 CFU/m<sup>3</sup>) after ambient air sampling. A comparison of the data showed that the

26 activity-based sampling undertaken using body worn personal sampler showed higher  
27 bioaerosols concentrations ( $10^4$  - $10^6$  CFU/m<sup>3</sup>), i.e. 2-3 logs higher than those recorded from  
28 static ambient air sampling. Bioaerosol exposure was highest during scavenging activities  
29 compared to waste sorting and site supervision . Particle size distributions showed that 41%,  
30 46%, 76% and 63% of total bacteria, gram-negative bacteria, *Aspergillus fumigatus* and total  
31 fungi respectively were of respirable sizes and would therefore be capable of penetrating deep  
32 into the respiratory system, posing a greater human health risk. This study has shown that  
33 exposure to bioaerosols can be associated with activities undertaken at open dumpsites and  
34 may contribute to the high prevalence of the chronic respiratory symptoms, among the workers  
35 in such environments.

## 36        **1. Introduction**

37        The rate of solid waste generation in developing countries has grown at a steady rate in recent  
38        years, especially due to urbanization and the accompanying urban population growth. Sub-  
39        Saharan Africa alone is estimated to generate 62 million tonnes per year of municipal solid  
40        waste (MSW) with a corresponding annual rate of change of urban population of 2.27 percent  
41        per year (UN-HABITAT 2009; Hoornweg and Bhada-Tata 2012b). Despite recording the  
42        highest rate of urbanization in the world, most developing countries unfortunately do not have  
43        the infrastructure to properly manage and treat their municipal solid waste. This has resulted in  
44        waste management authorities resorting to open dumping as a cheap method for managing their  
45        municipal solid waste (Srivastava *et al.*, 2015). Hence, it is unsurprising that 17 of the world's  
46        50 biggest dumpsites are located in the continent of Africa, of which 6 are in Nigeria (UNEP  
47        2015). This form of waste treatment involves the uncontrolled dumping of waste MSW on open  
48        land areas forming large waste hills with time. Microbial activities are central to the treatment  
49        of biodegradable MSW and agitation activities, such as tipping, spreading and waste sorting  
50        have been shown to aerosolise these microbial agents, including the pathogenic ones (Stagg *et*  
51        *al.*, 2010). Similarly, the analysis of the composition of MSW in sub-Saharan Africa have  
52        shown to have high biodegradable (40-60%) and high moisture content (Ogwueleka 2009;  
53        Hoornweg and Bhada-Tata 2012a). Furthermore, these characteristics in combination with the  
54        hot and humid weather conditions in the region favours the growth of microorganisms  
55        including pathogens that are eventually aerosolized during agitation activities at the dumpsite  
56        (Odewabi *et al.*, 2013; Epstein 2015). The individuals working on dumpsites such as  
57        scavengers, waste workers and vendors are often exposed to elevated levels of these pathogenic  
58        microorganisms majorly by inhalation as most have been noted to work in these conditions  
59        without personal protective equipment (Odewabi *et al.*, 2013). Workers exposure to organic  
60        dust laden with bioaerosols have been reported from composting and landfills facilities

61 typically associated by high emission of organic dust during agitation activities, a condition  
62 similar to open dumpsites (Ray *et al.*, 2005; Schlosser *et al.*, 2012). In addition to exposure by  
63 inhalation, other exposure routes such as ingestion and the skin contact presents additional  
64 exposure risk to the workers. These workers have been reported to be at risk of contracting  
65 gastrointestinal diseases, blood infections such as HIV and hepatitis, physical symptoms like  
66 impetigo and musculoskeletal symptoms (Thirarattanasunthon *et al.*, 2012; Odewabi *et al.*,  
67 2013).

68 Hitherto, respiratory health risk from exposure to bioaerosols at open dumpsite has received  
69 limited attention from researchers compared to that associated with landfills and the  
70 composting processes (Douwes *et al.*, 2003; Heldal *et al.*, 2015; Van Kampen *et al.*, 2016).  
71 Although activities such as tipping and spreading of waste may be common to landfills and  
72 dumpsites, the latter has greater environmental and public health impact as there are little or no  
73 controls over the safe handling, treatment and exposure limits to bioaerosols and particulate  
74 matter (UNEP 2005; Hoornweg and Bhada-Tata 2012b). Moreover, there are currently very  
75 limited literature on the exposure of dumpsite workers to bioaerosol emissions and the impacts  
76 on their respiratory health in a single study. As such, this research aims to investigate the  
77 degree of exposure to bioaerosols associated with working at open dumpsites and the potential  
78 impact this might have on the respiratory health of the workers at Olusosun open dumpsite.

## 79 **2. Materials and Methods**

### 80 **2.1 Site description and waste characterization**

81 The open dumpsite used in this study was situated in Ojota, Lagos State Nigeria. Lagos State  
82 has a population of 21 million with an annual growth of rate of 3.2% (LBS 2013). Of the three  
83 principal dumpsites serving the urban population of Lagos i.e Ewu Elepe, Solous and Olusosun  
84 dumpsites (LAWMA 2016), Olusosun is the biggest at 42.7 hectares (105.5 Acres), and is  
85 located between 6°35'40.9"N and 3°22'38.4"E (Oyeku and Eludoyin 2010). Olusosun is  
86 estimated to receive approximately 2,100,000 tonnes of MSW, construction and E-waste per  
87 year and has received approximately 17,150,000 to 24,500,000 tonnes of waste already since  
88 it became active in 1992 (D-Waste 2014). Olusosun dumpsite was chosen because it receives  
89 about 40% of the total municipal solid waste deposits in Lagos and the dumpsite is surrounded  
90 by sensitive receptors located less than 200 m from the boundary of the dumpsite (Olorunfemi  
91 2011).

### 92 **2.2 Control site**

93 In order to be able to determine the impact of the open dumpsite on the concentration of  
94 bioaerosols it was necessary to identify a 'control' site at which the concentration of  
95 bioaerosols would not be impacted by activities at the open dumpsite. The two sampling points  
96 were located ~ 10.79 km away from Olusosun open dumpsite within 6°29'58.9"N 3°22'47.5"E  
97 and 6°29'45.3"N 3°22'45.0"E. Moreover, both locations and shared similar economic and  
98 demographic characteristics with Ojota, where Olusosun dumpsite is located. The median age  
99 of the sampled population at the control and Olusosun matched a national demographic  
100 characteristic: 70% of adult population in Nigeria are less than 30 years in age (Reed and Mberu  
101 2014) and are both part of the planned Lagos Central Business District.

## 102 **2.3 Respiratory Health Survey**

103 A cross-sectional health survey was conducted across the Olusosun dumpsite between the 12<sup>th</sup>  
104 -20<sup>th</sup> January 2017. Sample size was calculated using the metrics as recommended by WHO  
105 (1991). Previous data on the prevalence of respiratory symptoms in landfill workers in India of  
106 65%, was used to calculate the sample size (Ray *et al.*, 2004; 2005). With the confidence level  
107 at 95%, bound on error at 5%, and statistical power at 80%, the sample size was 145 ±4 to  
108 account for attrition.

109 Participants were classed into the five major occupational groups: Scavengers (those who  
110 ravage the waste pile with tongs for recyclable resources); Waste workers (staff of the  
111 Government agency in charge of the safe disposing of waste and maintenance at the dumpsite);  
112 Business Owners (those with kiosks rendering other side services to members of the local  
113 population); Middle Men (those who buy the recovered recyclable materials from the  
114 scavengers); Food vendors (people who sell food to the other members of the local population).  
115 Participants were selected at random and were required to have worked at the dumpsite for at  
116 least one year to be eligible. A written or verbal informed consent was sought from all  
117 participant before administering the questionnaires.

### 118 **2.3.1 Operational definitions**

119 These definitions were adapted from the operation guidelines of the European Community  
120 Respiratory Health Survey (ECRHS) II, and the American Thoracic Society Division of Lung  
121 Disease questionnaire (ATS-DLD-78A) (Ferris 1978; ECRHS 2002; Birring 2011; Shaikh *et*  
122 *al.*, 2012; Gibson *et al.*, 2016). **Chronic cough** was defined as a consistent cough as much as  
123 4 to 6 times a day, 4 or more days a week or a cough first thing morning or both for at least 3  
124 months. **Chronic Phlegm** was defined as expectoration of sputum for as much as twice a day  
125 for at least 4 days a week or expectoration of sputum first thing in the morning or both for at  
126 least 3 months. **Chronic wheezing** was defined as wheezing in a dusty environment that would

127 have triggered 2 or more episodes of shortness of breath. **Asthma** was classified as at least two  
128 reported asthma attacks or shortness of breath with wheezing in the past two months with  
129 normal breathing between episodes of shortness of breath or a diagnosed asthmatic by a  
130 physician.

## 131 **2.4 Air sampling and sample analysis**

132 Two different types of air sampling were undertaken; ambient static air sampling at points  
133 around the dumpsite and at the control site using a six stage Andersen sampler and activity-  
134 based personal sampling using a body worn SKC button sampler.

### 135 **2.4.1 Ambient air sampling**

136 In Figure 1a, the sampling points and their distances away from the active part of the dumpsite  
137 are shown on a map. The sampling locations were chosen based on the prevalent wind direction  
138 as shown in the wind rose and were located as close as possible to the point at which activity  
139 was undertaken (Environment Agency 2017). The wind rose shows the average wind direction  
140 in the area over the period of 30 years (WMO 2017). The active area was considered as the  
141 point source as activities such as tipping of waste, spreading, compacting and scavenging took  
142 place here most frequently. Air samples were collected one day a week, for 13 weeks from the  
143 5<sup>th</sup> April to 28<sup>th</sup> August 2017. Due to the limited amount of equipment and manpower available  
144 to the researcher it was not possible to undertake concurrent sampling at all locations.

145 During sampling, air was pumped through the Anderson 6 stage sampler at the rate of 28.3 L  
146 min<sup>-1</sup> with a sampling time of 2.45 mins to avoid overloading of the petri dishes containing the  
147 selective media for the bioaerosols (Reinthal *et al.*, 1997). Samples were stored in sterile  
148 sealed bags and were taken to the laboratory for further analysis. It is important to note that at  
149 the time of sampling, site activities at Olusosun were at their peak. The microorganism, agar  
150 type, growth inhibitants, incubation temperature and duration for the different bioaerosols are



151 shown in the Table S1. After incubation, the number of colony forming units on each plate  
152 were counted, summed and subjected to positive-hole correction according to Macher (1989)  
153 and based on the volume of air sampled, were expressed as cfu/m<sup>3</sup>. This method, including  
154 bioaerosol quantification and enumeration, followed the protocol outlined in the Technical  
155 Guidance Note (M9) for monitoring of bioaerosols at regulated facilities by Environment  
156 Agency (2017) (England and Wales). Hence, the study was designed not to identify all  
157 mesophilic bacteria and fungi at the dumpsite but to align with emission standards from UK  
158 facilities, since Nigeria does not have any of such.

#### 159 **2.4.2 Activity-based air sampling**

160 Activity-based air sampling was carried out in order to measure bioaerosol exposure from  
161 specific activities engaged in by members of the population at the dumpsite. Three volunteers  
162 were recruited who were involved in scavenging, waste sorting and site supervision on the  
163 sampling days. They were required to spend most of their active time within 70 m from the  
164 active area of the dumpsite as exposure from these activities at this distance was assumed to  
165 be the highest compared to the other three sampling locations. The personal air sampler (Button  
166 Aerosol Sampler, SKC Inc., PA USA) was mounted vertically on the volunteers as close to the  
167 breathing zone as was practically possible. Sampling involved drawing sampled air at a flow  
168 rate of 4 L/min through a stainless cassette containing a 0.8 µm pore size gelatin filter while  
169 they went about their normal activities (Figure S1). The sampler captured air around the  
170 breathing zone for 30 mins for the three activities measured. After each sampling episode, the  
171 filters were removed from the sampler, dissolved in a sterile 30 ml vial of extraction fluid (0.01%  
172 Tween 80 in distilled water), and stored at 4 °C for transport back to the laboratory within 24  
173 h. Serial dilution were performed on all the raw samples with sterile buffered fluid (0.01%  
174 Tween 80 in distilled water) down to 10<sup>-4</sup> using standard aseptic techniques.. Subsequently, 10  
175 µl of the samples were plated out onto the appropriate selective media and incubated as shown

176 in Table S1. The bioaerosol density ( $c/m^3$ ) in the raw sample from the activity-based sampling  
177 was calculated using the following equation (Herigstad *et al.*, 2001):

$$178 \quad \text{Density (CFU } m^{-3}) = \frac{TC}{VP} \times DF \times BV$$

179 Where *TC*: total colony count, *VP*: Plated volume, *DF*: Dilution factor, *BV*: Volume of the raw sample.

## 180 **2.5 Statistical analysis**

181 a) **Qualitative health Survey**: Epi info™ 7 developed by the Centre for Disease Control and  
182 Prevention (CDC), USA., was used for preliminary data processing which were later  
183 transferred to Microsoft Excel for further processing. The relationship between predictor  
184 variables such as chronic cough, chronic phlegm, wheezing and asthma was analysed using a  
185 logistic regression statistic model. Covariates such as age, sex, smoking status, ‘years at study  
186 area’, ‘use of nose mask’ were adjusted for.

187 b) **Air sampling**: The test for normality was assessed by Kolmogorov-Smirnov test and  
188 nonparametric statistical tests were applied to the data set that violated the rule. The differences  
189 between the bioaerosol concentrations for the entrance, active area, dormant area and the  
190 boundary were assessed using the one-way ANOVA/Wetch ANOVA of normality. The  
191 homogeneity of variance was assessed by Levene’s test of homogeneity of variance. If violated  
192 by any of the variables, a Krustal-Wallis test was conducted to determine the difference in  
193 means across the sampling points at the dumpsite. Generally, the statistical analysis was carried  
194 out using IBM SPSS Statistics 22 for Windows (Version 22.0. Armonk, NY: IBM Corp., USA),  
195 Microsoft Excel and graphs generated using Origin (2015b, Origin Lab Corp., Northampton,  
196 MA, USA).

## 197 **3. Results and Discussion**

### 198 **3.1 Ambient air sampling**

199 The result of the one-way ANOVA conducted to determine if the difference in concentration  
200 of total bacteria and gram- negative bacteria across the four sampling locations were significant  
201 showed no statistically significant difference in their mean concentration, irrespective of the  
202 sampling location (total bacteria:  $F = 1.144$ ,  $p > 0.05$ , gram-negative bacteria:  $F= 2.463$ ,  $p >$   
203  $0.05$ ). Furthermore, running a Kruskal-Wallis one-way ANOVA on total fungi and *Aspergillus*  
204 *fumigatus*, total fungi indicated no statistically significant differences in mean concentration  
205 across the all sampling locations ( $\chi^2 (3) = 5.799$ ,  $p = 0.122$ ). *Aspergillus fumigatus* however,  
206 showed a significant difference between the means of the concentration at the active area and  
207 every other sampled location ( $\chi^2 (3) = 14.725$ ,  $p = 0.002$ ), suggesting a steep drop in the  
208 concentration of *Aspergillus fumigatus* as it travels beyond 50 m from the active area.

209 The dumpsites entrance recorded a median concentration of  $1.312 \times 10^3$  cfu/m<sup>3</sup> ( $231-1.849 \times 10^3$   
210 cfu/m<sup>3</sup>) and  $1.144 \times 10^3$  cfu/m<sup>3</sup> ( $786-2.043 \times 10^3$  cfu/m<sup>3</sup>) for total bacteria and gram-negative  
211 bacteria respectively, a 1-2 log higher concentration compared to *Aspergillus fumigatus* and  
212 total fungi (Table 1). The reason for this disparity is unclear, however it is possible that the use  
213 open-back waste trucks (1 in 4 waste trucks on dumpsite is open-back) and the extended  
214 waiting time at the entrance before tipping (mean: 2 hours), would likely contribute to  
215 bioaerosols concentration at that sampling location, as bioaerosols can also be emitted during  
216 the transport of waste to tipping point (Schlosser *et al.*, 2016). The weather conditions were  
217 favourable for most days, except for visit day 9, which started with a light shower but ended  
218 just before sampling begun at the entrance location. For the dormant area, the form of activities  
219 observed to take place here were mostly sorting and loading of recovered recycled materials  
220 into trucks; moreover, scavenging was also observed to sparsely take place as well. Bioaerosols  
221 sampling result shows the concentration of bacteria were  $> 10^3$  cfu/m<sup>3</sup> except for total bacteria

222 on visit day 6 (884 cfu/m<sup>3</sup>) and gram-negative on visit day 9 (939 cfu/m<sup>3</sup>). Total fungi on the  
223 other hand had concentration < 10<sup>3</sup> cfu/m<sup>3</sup> for the entire sampling visit (See Figure 2 M-P).  
224 The boundary was the closest sampling location to the nearest sensitive receptor. Bioaerosols  
225 concentration up to 1.63×10<sup>3</sup> cfu/m<sup>3</sup> and 2.26×10<sup>3</sup> cfu/m<sup>3</sup> for total bacteria and gram-negative  
226 bacteria respectively were measured at this location even though *aspergillus fumigatus* and  
227 total fungi were < 10<sup>2</sup> cfu/m<sup>3</sup> and 10<sup>3</sup> cfu/m<sup>3</sup> respectively.

228 Described in Table S2 are the meteorological conditions monitored during the sampling  
229 operation. The mean air temperature at the site varied from a low of 27°C up to a high of  
230 35.7 °C over the entire period. The relative humidity ranged between 64.3 – 88% with average  
231 wind speed of 1.2 (minimum) and 4.2 (maximum) during the same period. . Assessing the  
232 relationship between meteorological factors and bioaerosol concentrations across the dumpsite,  
233 the coefficient of Person's product moment correlation indicated that there was a statistically  
234 significant positive correlation between the concentration of gram-negative bacteria taken at  
235 the dormant area and atmospheric temperature (r = 0.649 n= 13, p < 0.05). Wind speed on the  
236 other hand showed a positive and negative correlation with the concentration of *Aspergillus*  
237 *fumigatus* at the boundary (r = 0.571, n =13, p < 0.05) and dormant area (r = -0.57, n= 13, p<  
238 0.05) respectively. Relative humidity showed no statistically significant correlation with the  
239 concentration of bioaerosols with respect to the sampling locations. However, upon conducting  
240 a multiple regression to further explain the overall change in the concentration of bioaerosols  
241 across the dumpsite in response to the independent variables (meteorological factors), the result  
242 indicated that, of the three independent variables tested, only relative humidity added  
243 statistically significantly to the prediction (p< 0.01) and that total bacteria concentration [F(3,  
244 48) = 4.140, p< 0.05, adj. R<sup>2</sup> = 0.156] was the most affected by the independent variable.

245 In general, the results of the bioaerosol sampling shows a progressive decline in concentration  
246 with distance, from the source point, downwind (see Figure 2). However, the one-way ANOVA

247 indicated no statistically significant difference in mean concentrations for the sampling points  
248 across the dumpsite for total bacteria ( $p>0.05$ ) and gram-negative bacteria ( $p>0.05$ ). This is  
249 surprising given that some of the sampling points were located a considerable distance from  
250 the active area (see Figure 1a). The result suggests a ‘fairly-uniform’ concentration for total  
251 bacteria and gram-negative bacteria across the dumpsite, which may be due to either  
252 meteorological factors or further bioaerosol emission from other sampling locations or both  
253 (Ogden *et al.*, 1969; Giner *et al.*, 1999). Moreover, although the prevailing wind direction was  
254 identified prior to sampling, during sampling it may be that the wind direction was variable  
255 and there may have been swirling locally. This could mean that some sampling locations may  
256 have been influenced by emission from other places and that would not have happened if the  
257 prevailing wind direction was maintained throughout the sampling period. Gram-negative  
258 bacteria particularly have been observed to be in high prevalence at open dumpsites located in  
259 these regions, hence their higher concentrations in this study is not a surprise (Odeyemi 2012).  
260 The steep decline in the concentration of *Aspergillus fumigatus* observed between the sampling  
261 location at the active area (50 m from active point; Median: 283 CFU/m<sup>3</sup>), the dormant area  
262 (531 m; median: 43 CFU/m<sup>3</sup>) and the boundary (788 m; 51 CFU/m<sup>3</sup>) showed an 80-81%  
263 reduction in concentration downwind. This behaviour of *Aspergillus fumigatus* was similar to  
264 what was observed by Schlosser *et al.*, (2016) where the decline was up to 77% between the  
265 tipping point at the landfill and the property boundary.

266 The mean bioaerosol concentration at the control site was generally 1-log lower than the  
267 concentrations observed at Olusosun dumpsite ( Table 3), supporting the fact that the Olusosun  
268 dumpsite was a major source of bioaerosols within the study area. In general, the bacteria  
269 concentration at the Olusosun dumpsite was typically 1-log higher than the fungi concentration.  
270 This is not surprising as Xu and Yao (2013) also reported that there are usually higher viable  
271 bacteria concentrations when compared to viable fungi concentrations when using an Anderson

272 impactor for air sampling. When the concentration of bioaerosols at all the sampling points  
273 were added together, the 95<sup>th</sup> percentile of concentration of total bacteria (2189 CFU/m<sup>3</sup>), gram  
274 negative bacteria (2352 CFU/m<sup>3</sup>), *Aspergillus fumigatus* (300 CFU/m<sup>3</sup>) and total fungi (824  
275 CFU/m<sup>3</sup>) were 2-3 log lower than reported in literature where similar activities such as tipping,  
276 spreading and compaction takes place (Kalwasińska *et al.*, 2014; Sangkham *et al.*, 2014). The  
277 main explanation for such differences probably relates to the sampling locations on-site and  
278 the sampling method employed. In this study using the Anderson six stage impactor, the closest  
279 sampling location to the active area was 50 m downwind; compared to other studies where  
280 sampling was either carried out at the working area or up 20 m away (Breza-Boruta 2012;  
281 Schlosser *et al.*, 2016).

282 Assessing bioaerosols emissions at Olusosun dumpsite against the expected limits set in the  
283 UK by the Environment Agency (2017), the concentration limits of total bacteria and gram-  
284 negative bacteria were exceeded on most visits to the sampling site during ambient air sampling  
285 (Figure 2, Table 2). Although the *Aspergillus fumigatus* and total fungi concentrations never  
286 exceeded the expected limits, the set limits were based on an 8-hour time weighted average  
287 estimation of exposure (HSE 2014). In this study however, the median daily exposure duration  
288 for participants at Olusosun dumpsite was 11 hours, indicating longer exposures, hence greater  
289 chances of inhaling more bioaerosols that could potentially cause related respiratory health  
290 problems to the population.

### 291 **3.2 Activity-based air sampling**

292 The result of the activity-based air sampling shows that the scavengers and workers alike were  
293 exposed to concentrations 2 -3log higher than the mean bioaerosol concentration measured  
294 during the static ambient air sampling (Table 3). The activity-based sampling using the  
295 personal body worn sampler allowed a picture to be obtained of the exposure of workers to  
296 bioaerosols as a result of engaging in multiple activities typical of those at a dumpsite. Activity

297 such as scavenging typically involves material collection and sorting with material collection  
298 being most prevalent during tipping and spreading of the waste while sorting tends to take  
299 place further away from the active area (Afon 2012).

300 Table 2 shows the concentration of total bacteria, gram-negative bacteria and *Aspergillus*  
301 *fumigatus* during site supervision, waste sorting and scavenging activities compared to the  
302 control and ambient air sampling. Although Schlosser *et al.*, (2016) alluded to the fact that the  
303 use of different equipment for bioaerosol sampling will yield different result, but the use of a  
304 combination of different sampling methods in this study produces a better validation of the  
305 results (Sandelowski 2000). Unlike several other studies where bioaerosol exposure  
306 measurements at landfills and dumpsites were restricted to static measurements from impaction  
307 and/or sedimentation, this study combined personal sampling and static sampling,  
308 consequently providing a robust dataset for the study. Furthermore, using bioaerosol data from  
309 static sampling only is problematic since the participants are usually not stationed at the same  
310 spots throughout the day's activities (Małecka-Adamowicz *et al.*, 2007; Schlosser *et al.*, 2016;  
311 Frączek *et al.*, 2017).

312 In Figure S2, exposure to bioaerosols from the various activities are compared to ambient air  
313 sampling and the result indicated higher exposure to total bacteria and gram-negative bacteria  
314 from scavenging up to  $10^6$  CFU/m<sup>3</sup>. Exposure to *Aspergillus fumigatus* on other hand was  
315 found to be high when undertaking site-supervision activities ( $3 \times 10^5$  CFU/m<sup>3</sup>) with lower  
316 exposure from sorting ( $9 \times 10^4$  CFU/m<sup>3</sup>) compared to scavenging (Table 3). This unusual spike  
317 in *Aspergillus fumigatus* during site monitoring may not be unrelated to the property of the  
318 waste (waste rich in fungi) tipped at the dumpsite at the time of sampling. Bioaerosols exposure  
319 from scavenging and sorting activities accounted for the highest exposure levels at the dumpsite,  
320 levels comparable to or higher than other forms waste-to-land treatment of municipal solid

321 waste such as landfilling and open windrow composting reported in some literature (Lis *et al.*,  
322 2004; Odeyemi 2012; Wéry 2014b; Breza-Boruta 2016).

### 323 **3.3 Particle size distribution**

324 To further evaluate the potential degree of exposure to bioaerosols at Olusosun dumpsite, the  
325 size distribution of bioaerosols were analysed and related to typical tidal volumes inhaled by  
326 humans. During sampling using the six stage Andersen sampler, bioaerosols are deposited out  
327 onto six separate agar plates located in the six stages of the sampler. The six stages of the  
328 sampler are designed to mimic the human respiratory system with collection of progressively  
329 smaller particles from stages 1 (top, upper respiratory tract) to stage 6 (bottom, lower  
330 respiratory tract and alveoli) (Jensen *et al.*, 1992; Tisch Environmental Inc. 2018). Particles of  
331 inhalable fraction with aerodynamic diameter  $>4.7 \mu\text{m}$  are deposited in stages 1 and 2,  
332 representing deposition in the nasal area. Stages 3 and 4 (thoracic fraction with aerodynamic  
333 diameter  $2.1 - 4.7 \mu\text{m}$ ) represents bronchial deposition, while deposition in the alveoli is  
334 represented in stages 5 and 6 (respirable fraction with aerodynamic diameter  $< 2.1 \mu\text{m}$ )  
335 (Andersen 1958; HSE 2003, 2014).

336 Figure 3(a)-(d) shows the percentage size distribution of bioaerosols as deposited on the  
337 different stages of the Anderson 6 stage sampler measured at the four sampling locations across  
338 the dumpsite. Taking into account all the bioaerosol collected from the 13 visit days,  $\sim 41\%$  of  
339 the total bacteria (ambient air:  $1.9-3 \times 10^2 \text{ CFU/m}^3$ ; Scavenging:  $1.2 \times 10^6 \text{ CFU/m}^3$ ; Site  
340 supervision :  $6.0 \times 10^5 \text{ CFU/m}^3$ ; Sorting:  $4.8 \times 10^5 \text{ CFU/m}^3$ ) were sized at  $< 2.1 \mu\text{m}$  aerodynamic  
341 diameter. Gram-negative bacteria on the other hand were slightly higher with  $\sim 46\%$  of the  
342 gram-negative bacteria at Olusosun dumpsite being  $< 2.1 \mu\text{m}$  aerodynamic diameter. The data  
343 for total fungi which also includes *Aspergillus fumigatus* indicates that  $\sim 76\%$  of all the fungi  
344 collected at Olusosun dumpsite were in the  $< 2.1 \mu\text{m}$  size range.



345 The aerodynamic diameter of bioaerosols is key to their dispersion, potential risk from  
346 inhalation and disposition in either the tracheal, bronchial or the alveolar regions of the lungs  
347 Brągoszewska *et al.*, (2017) and Ferguson *et al.*, (2017) had observed that bioaerosol exist  
348 either as single cells or agglomerates depending on the season and the meteorological  
349 conditions at the time of sampling. Their existence in an aggregated form, they argued, are  
350 likely responsible for their settlement in the upper stages (aerodynamic diameter > 3.3  $\mu\text{m}$ ) of  
351 the Anderson sampler either as aggregates of cells, cells associated with water droplets or  
352 particulate matter. Moreover, high relative humidity can further increase both the size and  
353 weight of the particle from absorption of ambient moisture, a phenomenon that favours higher  
354 deposition at upper stages of the Anderson sampler (Total Bacteria: ~59%; Gram-negative  
355 bacteria: ~54%) or dry deposition, giving a low reading for bioaerosol in the ambient air as  
356 observed in this study (Liu *et al.*, 2015; Smets *et al.*, 2016). This is perhaps not surprising as  
357 the multiple regression carried out in this study showed that, of the three metrological factors  
358 tested, only relative humidity showed a statistically significant association with the  
359 concentration of bioaerosols ( $p < 0.01$ ), especially total bacteria ( $F(3, 48) = 4.140$ ,  $p < 0.05$ , adj.  
360  $R^2 = 0.156$ ). The results shown in Figure 3 suggests a higher percentage of fungi (*Aspergillus*  
361 *fumigatus* ~76%; and total fungi ~63%) were within respirable fraction compared to bacteria  
362 (Total bacteria ~41%; gram-negative bacteria ~43%). Moreover, as observed by Deacon *et al.*,  
363 (2009), the recovery of *Aspergillus fumigatus* is usually highest within instruments pore size  
364 range of 3.3 -1.1  $\mu\text{m}$  as they are typically 2-3.5  $\mu\text{m}$  in diameter. Particles of inhalable fraction  
365 deposited in the nasal area and upper respiratory tract can usually be removed by the action of  
366 the nasal and tracheobronchial escalators, which is a combined mucociliary function of  
367 trapping deposited bioaerosol particles in mucus and removal by the action of cilia (Mason and  
368 Nelson 2005). However, this does not happen when particles of respirable fraction get down in  
369 to the pulmonary region where there is much more potential for health problems if deposited

370 down there, especially those who are immunocompromised (Yoshida and Whitsett 2004;  
371 Thomas 2013).

372 The tidal volume, which is the expected volume of air displaced during normal breathing, is  
373 approximately 7 mL/kg or 500 ml for a healthy young adult (Quanjer *et al.*, 1993; Ricard 2003).  
374 With a respiratory rate of 12 breaths per minute (Cretikos *et al.*, 2008; Erden *et al.*, 2015), the  
375 scavengers, waste sorters and waste workers working 11-hours (median) at Olusosun dumpsite,  
376 will inhale approximately 3.96 m<sup>3</sup> of air containing approximately 4.8×10<sup>5</sup> CFU/m<sup>3</sup> total  
377 bacteria, 1.4 ×10<sup>6</sup> CFU/m<sup>3</sup> of gram-negative bacteria and 5.2×10<sup>4</sup> CFU/m<sup>3</sup> of *Aspergillus*  
378 *fumigatus* during scavenging; 9.6×10<sup>4</sup> CFU/m<sup>3</sup> of total bacteria, 8×10<sup>5</sup> CFU/m<sup>3</sup> of gram-  
379 negative bacteria and 6.93×10<sup>4</sup> CFU/m<sup>3</sup> of *Aspergillus fumigatus* during waste sorting; 2.4  
380 ×10<sup>5</sup> CFU/m<sup>3</sup> of total bacteria, 9.7×10<sup>4</sup> CFU/m<sup>3</sup> gram-negative bacteria and 2.3 ×10<sup>4</sup> CFU/m<sup>3</sup>  
381 of *Aspergillus fumigatus* when carrying out site supervision , Table S3.

382 Several inhalation studies have investigated the effect of bioaerosols particle size on bioaerosol  
383 lungs deposition and the lethal dose required to initiate an infection. Although there are several  
384 animal models to this effect, no human studies exist investigating this relationship (Thomas  
385 2013; Dabisch *et al.*, 2017). The use of mouse models to demonstrate infection from  
386 *Aspergillus fumigatus* have been widely reported in literature, and that perhaps is because mice  
387 displays pathological consequences similar to humans (Kupfahl *et al.*, 2006; Dagenais and  
388 Keller 2009). Sheppard *et al.*, (2004) administered a dose of 2.4 ×10<sup>3</sup> CFU/mouse of  
389 *Aspergillus fumigatus* conidia to immunocompromised mice, resulting in a lethal pulmonary  
390 infection in most of the mice, surviving between 5 to 12 days. The exposure to *Aspergillus*  
391 *fumigatus* in this research (from scavenging, waste sorting and site supervision ) however, was  
392 up to 2.74 x 10<sup>5</sup> CFU/day , a 2-log higher than reported by Sheppard *et al.*, (2004). However,  
393 considering the complex nature of the structure and defence mechanism of the human  
394 respiratory system, it is unlikely that a similar dose will be lethal to humans, however, a prolong

395 exposure and neutropenia to will increase the risk of developing invasive aspergillosis by the  
396 exposed workers especially, the immunocompromised (Dagenais and Keller 2009; Wéry  
397 2014a). Gram negative bacteria have also been reported to be a potent pro-inflammatory  
398 stimulus in the human lungs due to flagellin release from their flagella (Liaudet *et al.*, 2003).

### 399 **3.4 Respiratory symptom from workers**

400 Summarised in Table 4 is the socio-demographic description of the participants in the cross  
401 sectional health survey. Of the 149 participants, 130(87%) and 19(13%) were males and  
402 females respectively. The result also shows that participants spent 11 hours daily (median) at  
403 the dumpsite and recorded a median “year at work” as 5. Moreover, the median age of the  
404 participants was 30 years, and the smoker and non-smoker population was 61(41%) and  
405 84(56%) respectively.

406 As shown in Figure 4 (A), smokers reported higher symptoms of chronic cough (21%) and  
407 chronic phlegm (15%) compared to non-smokers (chronic cough 15%, chronic phlegm 13%).  
408 This was however different with chronic wheezing and asthma, as non-smokers recorded  
409 higher prevalence (chronic wheezing 4%, asthma 1.3) when compared to smokers (chronic  
410 wheezing 3.5%, asthma 0.7%). Figure 4 (B) further shows reported symptoms among the non-  
411 smoker population in relation to how long the members have been at the study location. The  
412 occurrence of chronic cough (10%) and chronic phlegm (10%) were the highest for members  
413 of the population that have been at the study location in the first 5 years. Chronic phlegm was  
414 also noticed to be consistently reported to varying degree by non-smokers, irrespective of the  
415 years spent working at the dumpsite. Generally, the reported chronic respiratory symptoms  
416 were observed to have reduced as the years of spent at the dumpsite increased.

417 The result of the logistic regression showed a weak association between the independent  
418 variable ‘Use of Nose Mask’ and occurrence of chronic cough ( $p > 0.05$ ). Another independent

419 variable was ‘Years of work’ which showed no statistically significant association with chronic  
420 cough, chronic phlegm and asthma, for participants with years of work > 5 years (Table 5).

421 The prevalence of respiratory symptoms among the sampled population is not surprising since  
422 previous researches have also reported similar respiratory symptoms among waste workers and  
423 dumpsite scavengers. Athanasiou *et al.*, (2010) and Garrido *et al.*, (2015) for instance reported  
424 high prevalence of asthma and COPD symptoms among solid waste workers in municipality  
425 of Keratsini, Greece and city of Hamburg, Germany, respectively. Black *et al.*, (2019) also  
426 reported that 70% of the 1278 informal waste workers surveyed in their study at the dumpsite  
427 in Nuwakot district Nepal, reported one form of respiratory symptoms or other. Moreover,  
428 increased alveolar macrophages, neutrophils, eosinophils and lymphocytes in the sputum  
429 samples of examined female rag pickers (female scavengers) working on a waste dumpsite in  
430 Kolkata, India have been reported by Ray *et al.*, (2009). Although the study did not report the  
431 possible exposure concentration, the observed biomarkers from the body of the female rag  
432 pickers were evidence indicating a heightened immune system, which would suggest that they  
433 have been exposed to bacteria, endotoxins and viruses through inhalation.

434 Figure 6A shows that chronic cough and chronic phlegm recording a higher prevalence among  
435 smokers was not surprising, since chronic respiratory symptoms are most of the time associated  
436 with smoking (Abramson *et al.*, 2002; Frank *et al.*, 2006; Garrido *et al.*, 2015). The differences  
437 in percentage points in the prevalence of the reported respiratory symptom between smokers  
438 and non-smokers was marginal (chronic cough – 6%, Chronic phlegm- 2%, chronic wheezing-  
439 0.5% and asthma-0.6%) and suggests that the reported respiratory symptoms were largely of  
440 occupational origin than life style. This further supports growing evidence that chronic cough  
441 and chronic phlegm are risk factors for chronic obstructive pulmonary disease (COPD) and  
442 that reported cases of COPD among non-smokers are on the rise in developing countries (Salvi  
443 and Barnes, 2009; de Marco *et al.*, 2007).

444 It was expected that the non-use of a nose mask would greatly increase the odds of developing  
445 these outcomes; however, the model was not sensitive enough to measure this. This perhaps is  
446 because 87% of the respondents admitted not to have used a nose mask while working at the  
447 dumpsite, and the few that did (13%), used them only occasionally when they had access to  
448 PPE. This result should not be viewed as undermining the need to provide appropriate safety  
449 interventions, as such interventions have been recommended by researchers to reducing the  
450 risk of acquiring work related respiratory health problems (Lavoie *et al.*, 2006; Kuijter *et al.*,  
451 2010). It was also observed that all the respiratory health outcomes were not associated with  
452 ‘Years of work’ > 5 years ( $p > 0.05$ ) (Table 5). In their study of the respiratory health of brick  
453 kiln workers, Shaikh *et al.*, (2012) observed a similar result, however only for workers with  
454 ‘years of work’ > 10 years. The median ‘years of work’ at Olusosun dumpsite was 5 years and  
455 this represented 46% of the participants (Table 4). By implication, the majority of the  
456 participants who had reported to suffer chronic respiratory symptoms, would have contracted  
457 either chronic phlegm or asthma within the first 5 years of working at Olusosun open dumpsite.  
458 In a different report of a 5-year follow-up study of compost workers, Bünger *et al.*, (2007)  
459 observed a doubling of the number of workers with chronic bronchitis (RR 1.41; 95% CI 1.3-  
460 1.5) over the study period. Van Kampen *et al.*, (2016) also observed an increase relative risk of  
461 cough (RR 1.28; 95% CI 1.2-1.5) and phlegm (RR 1.32; 95% CI 1.2-1.5) and among compost  
462 workers working for at least 5 years at the composting facility irrespective how long their years  
463 of work was. They however, reported COPD as having no significant association with years of  
464 work. This study has demonstrated that working at dumpsites such as Olusosun may increase  
465 the odds of contracting chronic respiratory symptoms. This perhaps is primarily due to the  
466 limited use of PPE by the population, limited control over activities on the dumpsite and longer  
467 exposure hours, rather than longer years of work in such environments. In undertaking this  
468 study, there were some limitations which were largely due to limited resources and lack of

469 health records. Firstly, there was no morbidity record for the workers at the dumpsite making  
470 it difficult to compare the number of those who would have left the dumpsite due to ill health  
471 from exposure, with those currently working at the dumpsite within the same period. This may  
472 result in the underestimation of the extent of the respiratory health problems suffered by the  
473 workers in Olusosun dumpsite. Secondly, the findings for this study were largely based on a  
474 subjective inquiry without a complementary objective clinical measure. Spirometry and  
475 haematological profiling could not be carried out to assess the lungs functions and detect  
476 inhaled PM in the blood. The third limitation was that only the areas downwind of the  
477 dumpsite (selection based on the predominant wind direction across the dumpsite) was selected  
478 for sample group near the dumpsite. This may present a bias in the data, as there was regular  
479 swirling in the local wind during the survey activity, hence a possible exclusion of an important  
480 exposed group. Other cofounding factors such as vehicular emissions and industrial emissions  
481 at the study locations were not considered and consequently adjusted for in this study. Lastly,  
482 the probable underestimation of the actual bioaerosol exposure concentration due to  
483 uncultivable and non-viable fraction of airborne microorganisms; a weakness inherent in the  
484 use of the impaction sampling technique (HSE 2003; Persoons *et al.*, 2010).

#### 485 **4. Conclusion**

486 This study has demonstrated that open waste dumpsites are a major source of bioaerosols that  
487 can contribute adversely to the respiratory health of the waste workers, scavengers and others  
488 that are working at the dumpsite. Bioaerosols concentrations at Olusosun open dumpsite were  
489 2-3 log higher when compared to the control site. The results from the activity-based air  
490 sampling suggests that exposure to bioaerosol at Olusosun open dumpsite may also depend on  
491 the activities engaged in at the dumpsite. Scavenging recorded the highest level of bioaerosols  
492 exposure when compare to waste sorting, site supervision and the sampled ambient air. Gram-  
493 negative bacteria represented the highest bioaerosol concentration in Olusosun dumpsite. The

494 particle size distribution for *Aspergillus fumigatus* and total fungi showed a greater portion  
495 approximately 76% and 63% respectively were sizes < 3.3 µm. These particle sizes are  
496 respirable and can be deposited at the lower respiratory tracts. Observed also was the  
497 prevalence of chronic respiratory symptoms such as chronic cough and chronic phlegm, with  
498 earlier showing equally prevalence in both smokers and non-smokers and the later, more  
499 prevalent among the non-smoker population. Further clinical investigation into the respiratory  
500 health of the workers at the Olusosun dumpsite is necessary to determine the lungs function in  
501 responds to exposure to the bioaerosols.

## 502 **5. Supplementary data**

503 Supplementary data associated with this article is available at <https://doi.org/10.5518/449>

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## 508 **Ethics approval**

509 Ethical approval was obtained from the Engineering Faculty Research Ethics Committee,  
510 University of Leeds, UK.

511

512

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