Environmental Research 212 (2022) 113597

Contents lists available at ScienceDirect

Environmental Research

journal homepage: www.elsevier.com/locate/envres

Microbial contamination and metabolite exposure assessment during waste and recyclable material collection

Fabiola R.D. Salambanga [a,b], Loïc Wingert [b], Isabelle Valois [a], Nancy Lacombe [a], François Gouin [a], Julien Trépanier [b], Maximilien Debia [a], Ewelina Soszczyńska [c], Magdalena Twarużek [c], Robert Kosicki [c], Marta Dias [d,e], Susana Viegas [d,e,f], Liliana Caetano [d,g], Carla Viegas [d,e,f], Geneviève Marchand [a,b,*]

[a] Department of Environmental and Occupational Health, School of Public Health, Université de Montréal, Canada
[b] Institut de Recherche Robert-Sauvé en Santé et Sécurité Du Travail, Canada
[c] Kazimierz Wielki University, Faculty of Biological Sciences, Department of Physiology and Toxicology, Chodkiewicza 30, 85–064, Bydgoszcz, Poland
[d] H&TRC- Health & Technology Research Center, ESTeSL- Escola Superior de Tecnologia da Saúde, Instituto Politécnico de Lisboa, Portugal
[e] Comprehensive Health Research Center (CHRC), Canada
[f] NOVA National School of Public Health, Public Health Research Centre, Universidade NOVA de Lisboa, Portugal
[g] Research Institute for Medicines (iMed.ULisboa), Faculty of Pharmacy, University of Lisbon, Lisbon, Portugal

ARTICLE INFO

Keywords:
Waste collectors
Truck drivers
Exposure
Bioaerosols
Microbial metabolites
ddPCR

ABSTRACT

Waste workers are exposed to bioaerosols when handling, lifting and dumping garbage. Bioaerosol exposure has been linked to health problems such as asthma, airway irritant symptoms, infectious, gastrointestinal and skin diseases, and cancer. Our objective was to characterize the exposure of urban collectors and drivers to inhalable bioaerosols and to measured the cytotoxic effect of air samples in order to evaluate their health risk.

Personal and ambient air sampling were conducted during the summer of 2019. Workers from 12 waste trucks collecting recyclables, organic waste or compost were evaluated. Bacteria and fungi were cultured, molecular biology methods were used to detect microbial indicators, cytotoxic assays were performed and endotoxins and mycotoxins were quantified.

Domestic waste collectors were exposed to concentrations of bacteria and endotoxins above the recommended limits, and *Aspergillus* section *Fumigati* was detected at critical concentrations in their breathing zones. Cytotoxic effects were observed in many samples, demonstrating the potential health risk for these workers.

This study establishes evidence that waste workers are exposed to microbial health risks during collection. It also demonstrates the relevance of cytotoxic assays in documenting the general toxic risk found in air samples. Our results also suggest that exposures differ depending on the type of waste, job title and discharge/unloading locations.

Credit Author Statement

Conceptualization: G.M., L.W., M.D.,[1] C.V., S.V., L.C.; Investigation: F.S., L.W., I.V., N.L., F.G., J.T., E.S., M.T., R.K., M.D.,[1] S.V., L.C., C.V., M. D.,[2] G.M.; Formal analysis: F.S., L.W., N.L., F.G., J.T., E.S., M.T., R.K., M. D.,[2] Resources M.D.,[1] G.M., C.V.; Writing - Original Draft: F.S., M.D.,[1] G. M.; Writing - Review & Editing: F.S., G.M., M.D.,[1] L.W., I.V., N.L., F.G., J. T., E.S., M.T., R.K., M.D.,[2] S.V., L.C., C.V.; Supervision: M.D.,[1] G.M., C. V.; Project administration: G.M., L.W., M.D.,[1] C.V., I.V.; Funding acquisition: G.M., M.D.,[1] L.W., C.V., S.V., L.C.

1. Introduction

Sorting, recycling and composting activities are constantly increasing in order to reduce the environmental impacts caused by depositing and treating waste. Many municipalities begin improving their domestic and recyclable waste organization by developing waste management programs. The waste management industry is included into the environmental protection category of “the green economy,” which is a concept based on sustainable development (Cheneval et al., 2015). Several studies have reported that workers in the waste

* Corresponding author. IRSST, 505 Boul de Maisonneuve Ouest, 14th floor, Montréal, Québec, H3A 3C2, Canada.
E-mail address: marchand.genevieve@irsst.qc.ca (G. Marchand).

https://doi.org/10.1016/j.envres.2022.113597
Received 14 December 2021; Received in revised form 24 May 2022; Accepted 30 May 2022
Available online 31 May 2022

management industry are exposed to bioaerosols during the handling, lifting, and dumping of waste (Poulsen et al., 1995; Lavoie et al., 2006; Ncube et al., 2017; Viegas et al., 2020a,b). Bioaerosols can be defined as solid biological particulates suspended in the air, with various aerodynamic diameters ranging from hundreds of nanometers to tens of micrometers (Sykes et al., 2011). They may consist of pathogenic or non-pathogenic live or dead bacteria and fungi, viruses, allergens, bacterial endotoxins, mycotoxins, peptidoglycans, and/or microbial volatile organic compounds (Douwes et al., 2003; Wéry, 2014).

Exposure to bioaerosols in the occupational environment of waste sorting and recycling is a major concern because it has been linked to a number of adverse health effects. Studies have reported an association with airway symptoms, asthma, respiratory and infectious diseases, nose irritation, gastrointestinal and skin problems (Douwes et al., 2003; Ivens et al., 1999; Wouters et al., 2002). Allergic bronchopulmonary aspergillosis (ABPA) has also been reported in two compost collectors, where *Aspergillus fumigatus* (*A. fumigatus*) was identified in the patients' sputum (Poole and Wong, 2013). *Aspergillus* species were proposed as target indicators in waste management industries primarily because *Aspergillus* section *Fumigati* is a well-described pathogen and a common component of bioaerosols from the waste industry, but also because of the large increase in resistance to azoles observed in *Aspergillus* isolates, which may represent a serious health problem (Viegas et al., 2015; Varga et al., 2015; Sabino et al., 2016; "Stop Neglecting Fungi", 2017; Viegas et al., 2020a,b; Roca-Barcelo et al., 2020, Viegas et al., 2021a). Toxic effects following exposure to airborne biological agents are also associated with endotoxin (Rylander, 2002) and mycotoxin exposure (Varga et al., 2015). The exposure to a mixture of bioaerosols may lead to potentially additive and synergic effects (Samake et al., 2017; Von Essen and Donham, 1999). The use of cell models to determine the biological effects of multiple stressors has been reported in a variety of settings, including the waste management industry (Huttunen et al., 2008; Segura et al., 2009; Madsen et al., 2012; Gniadek et al., 2017; Viegas et al., 2017a, Viegas et al., 2020b-c). Human hepatocellular carcinoma cells (HepG2) have been used to target the cytotoxicity of mycotoxin mixtures (McKean et al., 2006), which are produced by various toxigenic fungal species, and metabolized in the liver. Human lung epithelial cells (A549) are used in bioassays as a model for alveolar cells (Swain et al., 2010). The use of cell models provides valuable information to improve our understanding of the multiple toxicity pathways associated with adverse outcomes (Hernandez et al., 2019; Altenburger et al., 2018; Roy et al., 2020; Mack et al., 2019).

A variety of factors were identified as determinants of exposure to bioaerosols during waste and recyclable material collection. The collection location and type of waste may influence worker exposure (Lavoie et al., 2006; Park et al., 2011). Exposures to culturable bacteria and fungi based on air sampling have commonly been reported in occupational hygiene studies (Lavoie et al., 2006; Wouters et al., 2006; Park et al., 2011; Marchand et al., 2017a,b). Although culturing methods may underestimate less abundant taxa in a specific environment (Mbareche et al., 2019; Madsen et al., 2020), their analysis remains critical in assessing health risks, as viability affects inflammatory responses and cytotoxic effects (Madsen et al., 2020). Because culturable flora represents approximately 10% of total flora (Eduard, 2009; Russell et al., 1999), some studies have recommended that molecular biology methods be used to evaluate total flora (Madsen et al., 2016; Viegas et al., 2017b). The use of a next-generation PCR technology called droplet digital PCR (ddPCR) has been proposed for absolute quantification (Hindson et al., 2011; Taylor et al., 2015; Verhaegen et al., 2016). Droplet digital PCR provides absolute quantification of nucleic acids without depending on amplification efficiency and without the need for a standard curve, leading to more precision in quantification when compared to other PCR methods. In addition, ddPCR overcomes part of the problem of inhibition that could likely lead to underestimation when using qPCR (Verhaegen et al., 2016; Rački et al., 2014; Taylor et al., 2017). The ddPCR technology also has a better ability to detect rare events in a microbial load sample, enabling a more comprehensive risk assessment of targeted microbial indicators.

The purpose of this study was to characterize bioaerosol concentrations during waste collection by implementing a comprehensive characterization strategy combining sampling and analytical methods to obtain an extensive assessment of the microbial risk. Culture-based methods and molecular tools such as ddPCR were applied to compare drivers' and collectors' exposure to microbial contamination. Antifungal resistance was evaluated to determine the specific risk of exposure to resistant microorganisms, and cellular toxicity was determined to assess the toxicity of the aerosols as a whole.

2. Materials and methods

Measurements and sampling were performed in the city of Montréal in the summer of 2019, during the months of July and August, in order to evaluate the worst-case scenario, since higher humidity and warmer temperatures promote microbial proliferation. Twelve waste collection trucks were assessed during a full working day (Table 1). The research team followed the trucks throughout the day to document the workers' activities.

2.1. Workplace description and sampling approach

For each truck, the waste collection team was composed of one driver and at least one collector. Collectors manually or mechanically dumped the contents of garbage containers into the back of the trucks. Drivers occasionally helped the collectors. Trucks used a compaction system during collection. At least once a day, the truck was driven to the waste disposal site to dump the collected waste. Generally, waste can be separated into three categories: domestic waste (mixed fractions), recyclables (cardboard, glass, paper, plastic, etc.) and compost (grass, food leftovers, soil, etc.). An employee's working day was between three and 8 h.

2.2. Sampling approach

2.2.1. Ambient air samples

The bioaerosols in the ambient air of the cabin were sampled on a 44 mm diameter electret filter used as standard with the SASS 3100 air sampler (Research International Instrument Inc., Washington, DC). This filter has a collection efficiency of 50% at an aerosol particle diameter of 0.5 μm when the SASS 3100 is operated at a flow rate of 300 L/min. The SASS samplers were mounted on the passenger seat near the driver and centre of the cabin (Research International inc.) The number of samples per type of waste is detailed in Table 1. Depending on the duration of the working day, three or four sequential samples were taken. All samples were analyzed separately. Half of the samples were used to perform culture and endotoxin analyses, and for targeted molecular detection by ddPCR. The other half were used to analyze mycotoxin, cytotoxin, and azole resistance. The sample's collection duration was between one and 2 h. The filters were composed of polypropylene electret microfibres. For the first half of the samples, the filters were extracted in a 50 mL falcon® tube (Thermo Fisher Scientific, Burlington, ON, CA) containing 10 mL of PCR-grade Phosphate Buffered Saline (PBS) (Fisher Scientific, Burlington, ON, CA), with the addition of 0.05% Tween 20 (Sigma-Aldrich Canada Co., Oakville, ON, CA). Extraction was completed by

Table 1
Number of samples collected per each waste type

Waste type	Trucks (N)	Sample Numbers		
		Driver	Collector	Cabin
Compost	2	4	4	7
Recyclable	6	12	13	24
Domestic	4	9	7	13

vortexing at 2500 RPM for 10 min in a multi-vortex (Fischer Scientific, Ottawa, ON, CA). For mycotoxin analysis, half of the filters were extracted with 4.0 mL of the extraction solvent (ACN: H2O: AcOH 79:20:1) for 60 min using a rotary shake. As for the cytotoxin and azole resistance assay, the other half of the filters were extracted in a 50 mL falcon® tube containing 10 mL of NaCl 0.9% with the addition of Tween 80 (Frilabo, Maia, Portugal) to a final concentration of 0.05%. These extracts were kept at −80 °C until analysis.

2.2.2. Personal samples

Personal measurements were taken on both drivers and collectors in the breathing zone (Fig. S1). The inhalable fraction (D_{50} = 100 μm) of airborne biological agents was collected using a CIP10-M (Tecora, Villebon-sur-Yvette, France). The flow rate was adjusted to 10 L/min, in accordance with the INRS sampling method (INRS, 2017). CIP10-M cupules were decontaminated with isopropanol for 1 min, then in DNA AWAY for 5 min (Thermo Fisher Scientific, Burlington, ON, CA), and finally autoclaved for 30 min. Two millilitres of sterile PCR grade PBS were added to the cupule just before turning the sampler on. To prevent dryness, 1 mL of PBS was added every hour. On each sampling day, two sequential samples were taken for each worker. Sampling time was adjusted to correspond to half of the working day (1.5–4 h). After the collection, the sample was harvested in a 2 mL Eppendorf tube (Thermo Fisher Scientific, Burlington, ON, CA). At the end of the sampling day, samples were brought to the laboratory, kept at 5 °C and analyzed separately the next day. All samples from the CIP-10 M were completed with PBS to a final volume of 2 mL. Aliquots of 500 μl and 1000 μl were taken for endotoxin and PCR analysis, respectively. The rest of the sample was used to make a 1/6 dilution for the culturable analysis.

Due to analytical limitations, not all analyses were performed on all samples. Table 2 provides a summary of the analyses performed on each sample type. Fewer analyses were performed on personal samples (collected with the CIP10-M) because of the limited sample quantity available.

2.3. Analyses performed

2.3.1. Culture analysis

All samples (N = 93; collectors (24), drivers (25) ambient air (44)) were analyzed for culturable microorganisms on two different culture media: tryptic soy agar (TSA) for the total culturable bacteria and malt extract agar (MEA) for the culturable fungi. In addition, 19 of the ambient air samples were analyzed using violet-red bile agar (VRBA), dichloran-glycerol agar 18% (DG18) and Sabouraud dextrose agar (SDA) to evaluate microbial diversity and SDA with itraconazole (ITR), posaconazole (POS) or voriconazole (VOR), in order to determine the susceptibility pattern of the collected fungi, following EUCAST breakpoints. Detailed protocols for culturable analyses have been published in previous reports (Viegas et al., 2020c) and are available in the Supplementary Material file.

Table 2
Analysis performed per type of sample

Analysis	Microbial target	Culture media	Sample type	
			Personal	Ambient air
Culture	Bacteria	TSA	X	X
	Gram-negative	VRBA		X
	Fungi	MEA	X	X
	Fungi	DG18		X
	Fungi	SAB		X
	Fungi-Azole	ITRA		X
	Fungi-Azole	VORI		X
	Fungi-Azole	POSA		X
Endotoxin			X	X
Mycotoxin				X
Cytotoxicity				X
dd-PCR	Fungal Biomass		X	X
	Asp/Pen		X	X
	A.section Fumigati		X	X
	A.section Flavi		X	X
	E.coli		X	X
	Enterococcus		X	X
	Legionella		X	X

TSA: trypticase soya agar; VRBA: violet red bile agar; MEA: malt extract agar; DG18: dichloran-glycerol agar; SAB: Sabouraud dextrose agar; ITRA: 4 μg/mL itraconazole supplemented SDA media; POSA: 0.5 μg/mL posaconazole supplemented SDA media; VORI: 1 μg/mL voriconazole supplemented SDA media

2.3.2. Endotoxin analysis

Endotoxin samples (N = 93) were either analyzed the same day or kept at 4 °C for 24 h. Analyses were performed following IRSST method 332 (Marchand, 2009).

2.3.3. Mycotoxin analysis

The same 19 ambient air samples analyzed with VRBA, DG18 and SDA were screened for mycotoxin presence. Mycotoxins were analyzed by HPL-MS ((HPLC) Nexera (Shimadzu, Tokyo, Japan) with a mass spectrometry detector API 4000 (Sciex, Foster City, CA, USA), following the same laboratory procedures described in previous papers (Viegas et al., 2021b). The mycotoxin concentration was calculated using external calibration. The Limit of Detection (LOD) obtained for each mycotoxin with the analytical method used are presented in Table 3S in the Supplementary Material file.

2.3.4. Molecular detection

Samples were concentrated by centrifugation before DNA extraction. Extracted DNA samples were kept at −20 °C until analysis. *Escherichia coli (E.*coli), total fungi *(FungiQuant), Aspergillus* section *Fumigati, Enterococcus* spp., *Legionella* spp., *Aspergillus* section *Flavi* and *Aspergillus/Penicillium (*Asp/Pen) detections were performed by ddPCR. Table 1S in the Supplementary Material file provides the sequences of the primers and probes used for detection (Integrated DNA Technologies IDT, Coraville, IA, USA). Wells with fewer than 12,000 accepted droplets were not included in the analysis (Viegas et al., 2020b). The Detection Limit (DL) was set at 3 copies/20 μl. For each gene that was amplified, a non-template control and a positive control were analyzed. The complete protocol for molecular detection by ddPCR is presented in the Supplementary Materials file.

2.3.5. Cytotoxicity evaluation

The same 19 ambient air samples were also screened for cytotoxicity, using the MTT assay to evaluate the metabolic activity of human lung epithelial (A549) and human hepatocarcinoma (HepG2) cells. The level of cytotoxicity was determined through spectrophotometric analysis of ten test dilutions of samples in an ELISA microplate reader (ELISA LEDETECT 96, biomed Dr. Wieser GmbH; MikroWin 2013 SC software). The lowest concentration of the samples causing a drop in absorption to <50% of cell metabolic activity (IC_{50}) was considered the threshold toxicity level. The complete protocol for cytotoxicity evaluation is presented in the Supplementary Materials file.

2.4. Statistical analysis

NCSS™ software was used to determine the distribution of the data. The Martinez-Iglewicz test was used to evaluate the normality of data distribution. All data were log-transformed to normalize them before performing the statistical analysis. The halved detection limit was used for static analyses when sample results were below the detection limit. The General Linear Model Analysis of Variance (GLM-ANOVA) and Tukey-Kramer multiple comparison tests were performed on the log-transformed data. A significant level of p = 0.05 was used for all statistical tests. The expostats tool was used to estimate geometric mean and geometric standard deviation (available from www.expostats.ca).

Table 3
Geometric mean (GM) and geometric standard deviation (GSD) of inhalable culturable bacteria and fungi from personal (drivers and collectors) and ambient air inside the cabins during waste collection

Type of waste	Site	Bacteria CFU/m^3 (n) GM (GSD)	Fungi CFU/m^3 (n) GM (GSD)
Recyclable	Cabins	(24) 1,620 (6.3)	(24) 320 (2.6)
	Drivers	(11) 1,480 (11.0)	(12) 340 (2.9)
	Collectors	(12) 2,070 (4.3)	(13) 1,130 (4.7)
Compost	Cabins	(7) 3,380 (5.3)	(7) 945 (2.1)
	Drivers	(4) 420 (4.0)	(3) 370 (2.4)
	Collectors	(4) 5,200 (9.0)	(4) 3,400 (3.0)
Domestic	Cabins	(13) 4,280 (3.3)	(13) 1,090 (3.1)
	Drivers	(9) 2,380 (3.7)	(9) 1,420 (4.8)
	Collectors	(7) 20,800 (12.1)	(7) 4,940 (5.9)

3. Results

3.1. Bacteria and fungi

The personal (collector and driver) and ambient air (cabin) concentrations of culturable bacteria and fungi are presented in Table 3. No trends were observed over the course of a day in relation to workers' exposure to culturable microorganisms.

The bacterial geometric mean (GM) ranged from 420 CFU/m^3 (compost/drivers) to 20,820 CFU/m^3 (domestic/collectors). The fungal GM ranged from 320 CFU/m^3 (recyclable/drivers) to 4940 CFU/m^3 (domestic/collectors). Regarding the bacteria and fungi, domestic waste collectors were significantly more exposed than recycling workers, with p values of 0.017 and 0.000183 respectively, but not more than workers collecting compost. The maximum per sample concentrations measured were 2.5×10^5 CFU/m^3 for bacteria and 4.3×10^4 CFU/m^3 for fungi. These concentrations are all higher than the proposed exposure limit in industrial environments characterized by the persistent presence of microorganisms (Table 4) (Marchand, 2021).

Coliform bacteria cultured on VRBA represented about 6% of the bacterial flora cultured on TSA. When the average concentrations were sorted by the type of waste (Table 5), the highest percentage was observed in the ambient air of the trucks during recycling material collection, with coliform bacteria representing 10% of the general bacteria recovered on TSA.

The average concentrations of xerophilic fungi grown on DG18 (1430 ± 1280 CFU/m^3) are higher than the concentrations observed on the general broad spectrum MEA (630 ± 560 CFU/m^3) as well as on SDA (430 ± 380 CFU/m^3). The genera *Penicillum* and *Aspergillus* are the most abundant on all media. In terms of diversity, the SDA enabled the characterization of eight different genera compared to six on the other two media (Fig. 1). Regarding the *Aspergillus* genera, species from the *Circumdati* section were predominant on both xerophilic (DG18) (86%) and clinical (SDA) (40%) media. On the other hand, on MEA, the most abundant *Aspergillus* were from the *Nigri* section, representing 68% of all the *Aspergillus* species cultured. Only the *Flavi* and the *Nigri* sections were retrieved on all three media. Species belonging to 8 of the 21 *Aspergillus* group sections could be identified using these 3 media. The highest average concentration was for the *Circumdati* section cultured on DG18 with more than 1000 CFU/m^3 (Table 6).

Table 5
Total and coliform culturable bacteria in the ambient air of the driver's cabin

Waste type	Total	Coliform	Coliform/Total
	CFU/m^3	CFU/m^3	%
	(n) Average (SD)	(n) Average (SD)	Proportion
Recyclable	(11) 5,900 (4,200)	(12) 590 (1,900)	10%
Compost	(3) 9,900 (6,200)	(3) 120 (110)	1%
Domestic	(5) 4,700 (2,600)	(5) 70 (80)	1%

3.2. Fungal azole susceptibility profile

Exposure to triazole-resistant fungi is presented in Fig. 2. In the ambient air from the cabins, the highest concentrations of fungal strains resistant to triazoles were obtained on media supplemented with voriconazole and posaconazole antifungal agents. The average concentration obtained on these two media represents nearly 40% of the concentration recovered on the non-supplemented SDA medium. The supplemented media did not show significantly different concentrations between them, but itraconazole-resistant fungal strains were the least common in the ambient air from cabins.

Penicillium was the most abundant genus observed on all azole-supplemented media, followed by *Cladosporium* sp. on itraconazole supplemented media, *Rhizopus* on voriconazole media and *Chrysonilia sitophila* on posaconazole media. Three different *Aspergillus* sections (*Candidi, Nigri* and *Nidulantes*) were observed on the posaconazole and voriconazole supplemented media, but not on the itraconazole supplemented media.

3.3. Molecular detection and quantification

Table 7 summarizes the concentration in DNA copies/m^3 of air for total fungi, *Asp/Pen* group, *Aspergillus* sections *Flavi* and *Fumigati*, *Legionella*, *E. coli* and *Enterococcus*, in the cabins as well as in the breathing zones of the drivers and collectors. The fungal biomass concentration during recycling collection was significantly lower ($p = 0.000007$) than that measured during the collection of compost and domestic waste. The peak concentration reaches 2,000,000 DNA copies/m^3 of air and was observed in the collectors' breathing zones during compost and domestic waste collection. The *Asp/Pen* group concentrations observed in the collectors' breathing zones were significantly higher ($p = 0.015601$) than those observed for the drivers. The *Asp/Pen* ($p = 0.000002$) and Aspergillus section *Fumigati* ($p = 0.000067$) concentrations during domestic waste collection were significantly higher than other types of waste. Section *Flavi* was detected in 25% of samples and at much lower concentrations (13–3410 DNA copies/m^3) than section *Fumigati* (16–145,200 DNA copies/m^3). No significant difference

Table 4
Proposed occupational exposure limit for industrial environments characterized by the persistent presence of microorganisms (Marchand, 2021)

Bioaerosols	Proposed limits
Culturable bacteria	10,000 CFU/m^3 of air (Malmros et al., 1992)
Culturable gram-negative	1,000 CFU/m^3 of air (Malmros et al., 1992)
Endotoxins	90 EU/m^3 of air ***or*** Concentration 30 times higher than the basic concentration (in presence of respiratory symptoms, the concentration should not be more than 10 times higher) (DECOS 2011)
Fungi	10,000 spores/m^3 of air ***or*** 1,000 CFU/m^3 of air (Eduard, 2007; SUVA, 2021)

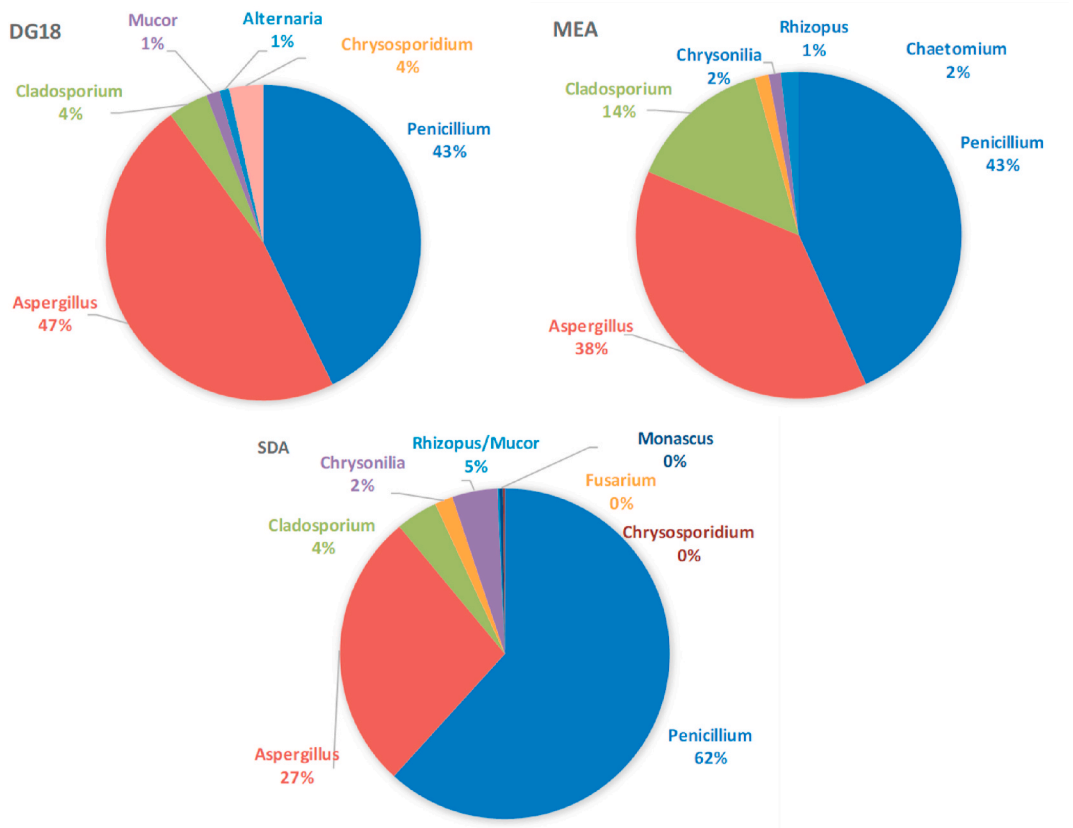

Fig. 1. Distribution of culturable fungi burden obtained from the ambient air on three culture media

Table 6
Aspergillus section distribution on three cultured media

Aspergillus	MEA		DG18		SDA	
	Average	Prevalence	Average	Prevalence	Average	Prevalence
Sections	CFU/m^3	%	CFU/m^3	%	CFU/m^3	%
Aspergilli			29	2		
Candidi					24	9
Circumdati			1048	86	110	40
Flavi	39	13	94	8	86	31
Fumigati	32	11	15	1		
Nigri	206	68	25	2	78	29
Restricti	27	9				
Nidulantes			11	1		

could be observed between the types of waste collected, but the collectors were significantly more exposed than the drivers. The greatest concentration of *Legionella* were again observed during domestic waste collection: 67% of samples had detectable levels. The collectors and drivers ($p = 0.000000$) were exposed to significantly higher concentrations than those measured in the cabin. As for *Enterococcus*, the concentrations observed in the breathing zones of the collectors were higher ($p = 0.008598$) than in the cabin and no statistical difference could be observed between the different types of waste. Only 13% of samples were positive for *E. coli*, with the highest concentration (3400 copies/m^3) measured for one domestic waste collector.

3.4. Metabolites (endotoxins and mycotoxins)

There were no detectable mycotoxins in any cabin air samples and most of the GM for endotoxins were under 100 EU/m^3 (Table 8). The highest GM was obtained for personal samples from the domestic waste collectors (96 EU/m^3). Overall, collectors were exposed to significantly higher endotoxin concentrations than drivers, no matter the type of waste collected. The highest concentration of endotoxins measured for a single sample was 450 EU/m3.

3.5. Cytotoxicity effects

Cytotoxic effects were observed in 37% of air samples in A549 cells, and in 100% of air samples in HepG2 cells, as presented in Table 9.

4. Discussion

The purpose of this study was to assess the exposure of drivers and collectors to airborne microbial agents and metabolites and to document the potential cytotoxic effects of bioaerosols inside the cabin during the collection of recyclable materials, compost and domestic waste. A comprehensive sampling strategy was carried out to perform this analysis and characterize the health risk for workers in this industry.

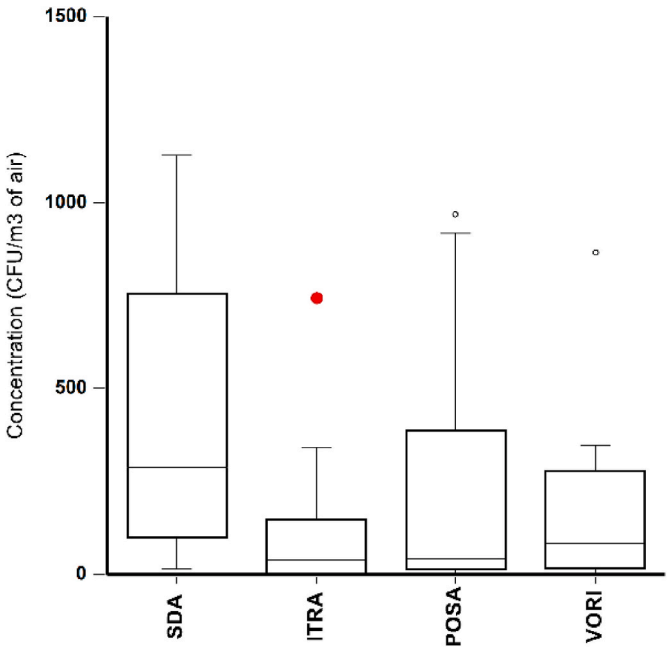

Fig. 2. Fungal burden in azole screening media, from samples collected in the ambient air of the waste truck's cabin (N=19) (red dots are outliers). SDA: Sabouraud Dextrose Agar; ITRA: 4 μg/mL itraconazole supplemented SDA media; POSA: 0.5 μg/mL posaconazole supplemented SDA media; VORI: 1 μg/mL voriconazole supplemented SDA media

4.1. Exposure to culturable microbiota

The evaluation of workers' exposure to bioaerosol from culturable microbiota is the traditional and most widespread approach used in environmental and occupational studies. This method has the advantage of enabling comparisons with the exposure limit values and with previous studies applying the same methods.

In the present study, regardless of the type of waste, the highest bacterial concentrations were observed in the collectors' breathing zones during domestic waste collection (20,820 CFU/m^3). The concentrations reported during domestic waste collection were comparable to previously reported bioaerosol concentrations of 21,000 CFU/m^3 for collectors, 3400 CFU/m^3 for drivers and 470 CFU/m^3 in cabins (Nielsen et al., 1995). However, Park (collectors = 220,000 CFU/m^3; drivers = 18,000 CFU/m^3) and Krajewski (collectors = 267,000 CFU/m^3; drivers = 59,000 CFU/m^3) observed higher exposure levels for domestic waste workers (Park et al., 2011; Krajewski et al., 2002). Concerning recyclable materials and compost, respective median concentrations of 5600 CFU/m^3 and 50,300 CFU/m^3 were reported for collectors in a previous study (Lavoie et al., 2006). Our results (420–20,800 CFU/m^3) are not as high, but they confirm that domestic waste and compost collectors are exposed to bacterial concentrations near the recommended limit of 10,000 CFU/m^3 (Poulsen et al., 1995; Goyer et al., 2005; Marchand et al., 2017a; Marchand, 2021).

Concerning coliform bacteria, although most are not pathogenic, they can be good indicators of bioaerosol exposure. However, few studies specifically assess coliform bacteria in air. They are largely used for water and food quality evaluation and could also be useful for air quality assessment. Coliforms live in the intestines of warm-blooded animals (including humans) and their presence implies fecal pollution. Their presence in the air may pose a health risk to workers. No exposure limit is specifically proposed for coliforms in air, but an exposure limit of 1000 CFU/m^3 is proposed for total gram negative bacteria, which includes coliforms (Poulsen et al., 1995; Goyer et al., 2005; Marchand, 2021). Coliforms were measured in cabin air only, and the highest average concentration measured was in the recycling trucks (590 CFU/m^3). A study carried out in a wastewater treatment plant reported maximum concentrations of 260 CFU/m^3 (Szyłak-Szydłowski et al., 2016). In a study of air in Mexico, 1.2% of outdoor air and 2.7% of indoor air contained gram-negative bacteria (Rosas et al., 1997), lower than the 10% observed for the recyclable material collection trucks in this study. Inhaling bacteria can cause both respiratory and enteric

Table 8
Endotoxin concentrations of personal (collectors and drivers) and ambient air (cabins) measured during waste collection

Type of waste	Site	Endotoxin EU/m^3 (n) GM (GSD)
Recyclable	Cabins	(13) 5 (3.5)
	Drivers	(12) 7.5 (4.9)
	Collectors	(13) 20.9 (2.8)
Compost	Cabins	(4) 5.2 (1.6)
	Drivers	(4) 4.5 (2.2)
	Collectors	(4) 52 (4.4)
Domestic	Cabins	(8) 20 (1.6)
	Drivers	(9) 34 (2.6)
	Collectors	(7) 96 (3.2)

Table 9
Distribution of threshold toxicity levels (IC50) of collected air samples (N=19)

IC50	A549	HepG2
	Number of toxic samples	Number of toxic samples
10 mm^2/ml	2	15
5 mm^2/ml	2	3
2.5 mm^2/ml	2	0
1.25 mm^2/ml	1	1
0.625 mm^2/ml	0	0
N.D.	12	0

Table 7
Microorganism quantification and detection of personal (collectors and drivers) and ambient air (cabins) measured during waste collection

Waste type	Site	Total fungi	*Aspergillus* section *Fumigati*	*Aspergillus section Flavi*	*Asp/Pen*	*Legionella*	*E.coli*	*Enterococcus*
		ITS copies/m^3	18S copies/m^3	PEP copies/m^3	ITS copies/m^3	16S copies/m^3	grf-copies/m^3	16S copies/m^3
		GM (GSD)	GM (GSD)	GM (GSD)	GM (GSD)	GM (GSD)	GM (GSD)	GM (GSD)
Recyclable	Cabins (n=13)	22,000 (4.8)	190 (5.1)	13 (1.9)	3,300 (7.6)	13 (2.2)	8 (2.4)	380 (8.6)
	Drivers (n=12)	6,600 (5.0)	60 (2.1)	35 (1.4)	850 (5.8)	84 (2.6)	37 (1.8)	140 (4.8)
	Collectors (n=13)	44,000 (8.7)	100 (4.6)	50 (3.7)	5,700 (8.7)	86 (2.7)	35 (1.3)	250 (5.6)
Compost	Cabins (n=4)	870,000 (1.6)	310 (2.8)	14 (1,6)	6,700 (2.6)	11 (1.5)	6 (1.3)	94 (2.1)
	Drivers (n=4)	13,000 (1.2)	210 (2.9)	40 (1.2)	2,600 (2.5)	55 (2.1)	40 (1.2)	97 (1.9)
	Collectors (n=4)	320,000 (6.3)	530 (4.8)	90 (2.2)	40,000 (3.4)	200 (3.7)	53 (1.2)	540 (22.2)
Domestic	Cabins (n=8)	140,000 (2)	3,800 (2.1)	15 (1.6)	96,000 (11)	54 (3.8)	13 (3.8)	460 (3.8)
	Drivers (n=9)	70,000 (5.1)	990 (12)	74 (2.2)	21,000 (6.5)	37 (2.2)	54 (1.8)	680 (7)
	Collectors (n=7)	200,000 (5.6)	1,500 (4.9)	54 (1.9)	50,000 (5.1)	220 (4.4)	91 (5.3)	990 (7.5)

Grf: gene regulation function; ITS: internal transcribed spacer; PEP: aspergillopepsin

symptoms, as it is also possible for bacteria to enter the mouth and throat directly or through nasal secretions and be ingested (Ivens et al., 1999; Douwes et al., 2003).

Workers' exposure to fungi was higher during the collection of compost and domestic waste than during the collection of recyclable materials. In addition, collectors were about four times more exposed than drivers. Regardless of the type of waste, drivers' and collectors' fungi concentrations measured in our study were in the same range as found by Madsen (GM: 5700 CFU/m^3) (Madsen et al., 2016). Other studies reported higher concentrations, ranging from 47,000 to 63,000 CFU/m^3 (Nielsen et al., 1995; Krajewski et al., 2002; Park et al., 2013). All collectors and drivers collecting domestic waste had exposures exceeding the fungal recommendation of 1000 CFU/m^3 (Marchand et al. 2017a, 2017b; SUVA, 2021; Marchand, 2021). In a previous study by Nielsen et al. detection of section *Fumigati* by culture was achievable in 20% of the samples taken from domestic waste workers (Nielsen et al., 1995). In this study, only 2% of the samples contained this section. *Penicillium* and *Aspergillus* species represented more than 80% of the culturable fungi isolated from cabin air. *Penicillium* has previously been identified as the dominant genus in landfill, wastewater treatment plants and during waste collection by other researchers (Kalwasińska et al., 2014; Viegas et al., 2014; Madsen et al., 2019).

4.2. Azole resistance screening

Azole resistance is an emerging phenomena that poses a real danger for immunocompromised individuals due to the increased risk of invasive aspergillosis, caused by *Aspergillus* section *Fumigati* ("Stop Neglecting Fungi" 2017; Resendiz et al., 2018). Although *A.* section *Fumigati* was not observed on the azole supplement media, the growth of fungi from air samples at EUCAST azole-breakpoint concentrations raises a red flag about the potential development of azole resistance in this environment and demands further investigation. In fact, a previous study reported section *Fumigati* isolates bearing the TR34/L98H mutation known for causing azole resistance in respiratory protective devices worn by workers in Portugal's waste-sorting industry (Gonçalves et al., 2021).

4.3. Molecular analysis of indicator microorganisms

Culturable microbiota has the drawback of representing only a very small percentage of the real microbial risk, since it is limited to culturable microorganisms that are able to develop under laboratory conditions (Cox et al., 2020). Viability is also an important parameter when studying infectious risks, which cannot be assessed using culturable microbiota. Overall, culture methods underestimate the immunological, toxicological and irritative health risks associated with bioaerosol exposure. For this reason, several researchers are now using molecular methods to assess microbial risk independently of culturability (Eduard and Halstensen, 2009; Viegas et al. 2012, 2014; Marchand et al. 2017a, 2017b; Mbareche et al., 2019). In addition, molecular biology detection methods make it possible to target certain microbial agents as risk indicators (*A.* section *Fumigati*, *A.* section *Flavi*, *Legionella, E. coli and Enterococcus*). These microorganisms, even in low concentrations, can represent a significant risk to the health of workers due to their infectious and toxicologic potentials.

Total fungi concentrations ranged from 6800 to 860,000 copies/m^3. These results indicate that 0.6%–7% of fungi were culturable when compared to the total flora obtained by molecular detection. This is not surprising, since culturable organisms have been reported to represent approximately 10% of the total microflora (Russell et al., 1999; Eduard, 2009). Fifty percent of the samples from the breathing zones of workers were over the 100,000 copies/m^3 threshold, proposed as an occupational limit by Eduard et al. (2012). In addition, the air measurements in the cabins for the three types of waste show that they were heavily contaminated by fungi, the highest concentration being observed in a compost truck (1,700,000 copies/m^3). These observations are consistent with the study by Madsen et al. which reported the contamination of cabins by airborne microorganisms from the outdoor unloading process and by workers entering the truck with their contaminated clothing (Madsen et al., 2016).

Aspergillus and *Penicillium* species represented the vast majority of the culturable microbiota measured in the present study. *Aspergillus* are well-known molds that can cause health problems due to their toxicity and allergenic and infection effects (Varga et al., 2015; Sabino et al., 2019). Species of the *Penicillium* genus have been reported by some to be the most frequent in waste collection environments (Madsen et al., 2020). Exposure to *Penicillium* can be associated with expiratory flow rate variability in people with asthma, and can induce both immediate and late asthma in sensitive individuals (Knutsen et al., 2012). Contrary to total fungi, the highest concentrations for the *Asp/Pen* group were observed during domestic waste collection.

Section *Fumigati* has the greatest clinical relevance, known for causing aspergillosis and other respiratory pathologies (Land et al., 1987; Malta-Vacas et al., 2012; Mousavi et al., 2016). Hypersensitivity pneumonitis (HP) caused by this section has previously been diagnosed in a waste worker (Hagemeyer et al., 2013). The *Fumigati* section is considered the most toxic among all the *Aspergillus* genus (Anyanwu et al., 2003; Piecková, 2012), due to its ability to produce a number of mycotoxins (Viegas et al., 2017b). Hence, it is recommended that exposure be reduced to a minimum in occupational environments (Marchand et al. 2017a, 2017b; Marchand, 2021). This section was detected in all air samples from the cabins, and domestic waste trucks were the most contaminated. A concentration of 145,000 copies/m^3 was observed in a driver's breathing zone when he was at the landfill to empty his load. This certainly demonstrates a health risk situation for the drivers who must go to this landfill site (Fig. S2), and wearing a respirator should be required. To our knowledge, no other studies have reported section *Fumigati* exposures by molecular methods during waste collection.

Although species from the *A.* section *Flavi* can cause invasive infections, their occupational health risk arises more from their ability to produce aflatoxins. These mycotoxins have significant health effects on humans and animals (Okoth et al., 2012; Varga et al., 2015). IARC Monographs have reported that naturally occurring aflatoxins are carcinogenic to humans (Group 1) (IARC, 1993, 2002). In the present study, *A.* section *Flavi* was detected in one out of four samples and the collectors were the most exposed group, with a maximum concentration of 3400 copies/m^3 measured in their breathing zones during recyclable material collection. Previously, in a study aiming to assess exposure to aflatoxin B1 in workers from the Portuguese waste industry (Viegas et al., 2012b, 2014), all the workers enrolled in the study showed detectable levels of AFB1 in blood samples. Values ranged from 2.5 ng mL^{-1} to 25.9 ng mL^{-1}, with a median value of 9.9 $\pm$ 5.4 ng mL^{-1}, and all of the controls showed values below the method's detection limit (Viegas et al., 2012b, 2014).

Legionella may be responsible for respiratory pathologies. Even though legionellosis has been commonly linked with environmental exposures via contaminated water, compost and soil (Le Goff et al., 2010, Whiley and Bentham, 2011; Picard Masson et al. 2016; van Heijnsbergen et al., 2015), work environments are increasingly recognized as sources of *Legionella* infections (Principe et al., 2017). Among others, *Legionella* has been reported in composting environments, in a metal recycling plant, a wastewater treatment plant, a dentist's office, healthcare facilities, automotive industries and a recycling plant (Marchand, et al. 2017a, 2017b; Picard-Masson et al., 2016; Ali et al., 2010; van Heijnsbergen et al., 2015; Principe et al., 2017). Many countries have adopted public health policies to fight *Legionella*

infections but there is no occupational exposure limit for air samples and further efforts need to be made to protect workers (Principe et al., 2017). In the present study, 67% of the samples were positive for *Legionella*, with concentrations reaching 1300 copies/m^3. This is considerably lower than what has been measured in composting plants, where concentrations up to 185,000 copies/m^3 have been detected (Bonifait et al., 2017).

Although most strains of *E. coli* and *Enterococcus* cause only minor infections, their presence in water is indicative of the possible appearance of more pathogenic microorganisms (Price and Wildeboer, 2017). These two microorganisms are widely used for water quality assessment. They could also be relevant microbial indicators for the evaluation of occupational exposure to bioaerosols in some working environments. In this study, *E. coli* and *Enterococcus* exposure levels were in the same range for compost, recyclable material and domestic waste, with GM lower than 100 copies/m^3 for *E. coli* and 1000 copies/m^3 for *Enterococcus*. Collectors' exposure levels were higher than the recommended level of 1000 CFU/m^3 for gram negative bacteria, even though they represented only a fraction of them (*E.coli*: 1800 copies/m^3 and *Enterococcus*: 7400 copies/m^3). Few studies have performed analyses of inhalable *E. coli* and *Enterococcus* in the waste industry. In their study, Krajewski et al. identified the presence of *E. coli* in nine of the ten air samples taken on workers during waste collection (Krajewski et al., 2002); this study's occurrence is above 20% (5/24) for *E. coli* and 75% for *Enterococcus*, detected in the collectors' breathing zones.

4.4. Endotoxin and mycotoxin exposure

The Dutch expert committee on exposure recommended a health-based occupational limit value for endotoxins of 90 EU/m^3 based on respiratory symptoms (DECOS. Endotoxins-Health-based recommended occupational exposure limits., 2010). According to what has been reported by Madsen et al. (2020), in their literature review, the majority of waste collection workers would be exposed to endotoxin concentrations of around 10 EU/m^3. In our study, the endotoxin concentrations ranged from 1.0 to 450 EU/m^3 with a geometric mean of 35 EU/m^3. These results are in accordance with Wouters et al. for the exposure of collectors and drivers (GM: 29 to 66 EU/m^3) during organic, residual and mixed waste collections (Wouters et al., 2002). However, Krajewski et al. measured average concentrations for domestic waste workers that are four to ten times higher than the levels measured in the present study (Krajewski et al., 2002). Even though the average concentration observed is lower than the recommended limit, 13% (10/74) of the sample concentrations exceed that limit. This represents a risk to the health of exposed workers, since it is generally admitted that exposures should be controlled so that less than 5% of exposures exceed the recommended limit.

Despite the fungal contamination found, no sample from cabin air showed contamination by mycotoxins. The reason for these findings might be due to the SASS filter not having small enough pore sizes to retain the particles that carry mycotoxins (smaller diameter); this may have influenced the results obtained, with no mycotoxins being detected (Brasel et al., 2005; Viegas et al., 2014). Previous studies aiming to detect airborne mycotoxins demonstrated that it is necessary to use filters with very small pore sizes, namely 0.2 μm, to retain mycotoxins (Pasanen et al., 1993; Johanning et al., 2002; Viegas et al., 2017b).

4.5. Cytotoxicity assessment

The primary routes of bioaerosol exposure to humans are believed to be inhalation and inadvertent ingestion of contaminated dust. Human lung epithelial (A549) and human hepatocyte carcinoma (HepG2) cells were chosen to assess the cytotoxic effect of air samples from waste truck cabins, because of their characteristics and relevant predictive value from *in vitro* models for different exposure routes, such as inhalation and ingestion. Our results showed that ambient air samples from waste truck cabins could, to varying extents, inhibit the metabolic activity of both cell types. Previous studies using the MTT assay with these cell lines propose oxidative damage and apoptosis as the possible underlying cytotoxic mechanism (An et al., 2016; Taroncher et al., 2020). Despite their limitations, A549 and HepG2 cell lines are among the most widely used for lung and liver, respectively (Nikolic et al., 2018). Of note, the purpose of this study was not to fully validate A549 nor HepG2 as tools for lung and liver toxicity, but more to use them to identify the different toxicities of inhalable pollutants present in the air of waste truck cabins.

4.6. Worker exposure assessment

The present study suggests that occupational health risks from bioaerosols are greater during domestic waste collection when compared to recyclable material and compost collections, for most of the microbial contaminants. Our results show that collectors are the most exposed and could be at risk of developing health effects related to microbial exposure. This was expected, since collectors are in close contact with waste and the aerosolization of microorganisms could occur during loading and compacting garbage in the truck. Although garbage collectors are the most exposed, this study found that one domestic truck driver was exposed to the highest measured concentrations of mold, endotoxins, *Asp/Pen* group, *Aspergillus* sections *Fumigati and Flavi* and *E. coli*. The landfill used by this driver is in the open air and the contents of his truck were unloaded onto a pile of garbage (Fig. S2). This task could be responsible for the highest exposure, but since the other drivers using the same landfill did not present the same levels, a more exhaustive evaluation would be necessary to validate if unloading at this type of landfill represents a higher risk.

Because of the intensity of the activity itself, the high temperatures and humid environment during the summer, and the need to communicate in a noisy environment, it is very difficult to wear personal respiratory protective equipment during waste collection. However, it should be recommended that protective equipment be worn in specific situations that have the potential to generate high bioaerosol concentrations, such as when dumping waste at the landfill and when trucks are cleaned. Additionally, measures to regularly clean the trucks' cabins in order to reduce long-term contamination risks should be implemented. Respiratory protection must be used during cleaning.

4.7. Limitations

This project has some limitations, such as a restricted sample size, but it does not prevent the obvious observation of waste collectors and drivers being exposed to bioaerosols at certain times. Because this study assesses the worst-case scenario, the risk may not be the same during other times of the year.

5. Conclusions

This study is one of the first in the field of occupational health to use the quantification and detection of microorganisms by ddPCR, which allows absolute quantification of the workers' exposure to total bioaerosols. This multi-technique approach, based on the use of different sampling methods (personal, environment), and different assays (culturing, molecular biology, endotoxin, mycotoxin and cytotoxicity) made it possible to analyze a broader spectrum of the microbial biota present and of the health risk it poses to the waste collection industry. This approach is increasingly used and industrial hygienists should rely on these new strategies to obtain an accurate assessment of microbial risk.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

This work was supported by the Institut de Recherche Robert-Sauvé en Santé et en Sécurité du Travail (Grant #2018–0016). H&TRC authors gratefully acknowledge the national support of FCT/MCTES through the UIDB/05608/2020 and UIDP/05608/2020. Fabiola Salambaga is grateful to the Département de santé environnementale et santé au travail of the School of Public Health of Université de Montréal for her Master's grant. We would like to thank all the workers and waste management companies that participated in this study.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envres.2022.113597.

References

Anyanwu, E., Campbell, A.W., Jones, J., Ehiri, J.E., Akpan, A.I., 2003. The neurological significance of abnormal natural killer cell activity in chronic toxigenic mold exposures. Sci. World J. 3, 1128–1137. https://doi.org/10.1100/tsw.2003.98.

Altenburger, R., Scholze, M., Busch, W., Escher, B.I., Jakobs, G., Krauss, M., Krüger, J., et al., 2018. Mixture effects in samples of multiple contaminants – an inter-laboratory study with manifold bioassays. Environ. Int. 114 (May), 95–106. https://doi.org/10.1016/j.envint.2018.02.013.

Ali, S., Phillips, C.A., Phillips, P.S., Bates, M., 2010. Isolation and identification of Legionella pneumophila from material reclamation facilities. Int. J. Environ. Health Res. 20 (5), 367–377. http://www.tandfonline.com/doi/full/10.1080/09603123.2010.484859.

An, J., Hu, J., Shang, Y., Zhong, Y., Zhang, X., Yu, Z., 2016. The cytotoxicity of organophosphate flame retardants on HepG2, A549 and Caco-2 cells. J. Environ. Sci. Health. Part A 51 (11), 980–988. https://doi.org/10.1080/10934529.2016.1191819.

Bonifait, L., Marchand, G., Veillette, M., M'Bareche, H., Dubuis, M.-E., Pépin, C., Cloutier, Y., et al., 2017. Workers' exposure to bioaerosols from three different types of composting facilities. J. Occup. Environ. Hyg. 14 (10), 815–822. https://doi.org/10.1080/15459624.2017.1335054.

Brasel, T.L., Douglas, D.R., Wilson, S.C., Straus, D.C., 2005. Detection of airborne stachybotrys chartarum macrocyclic trichothecene mycotoxins on particulates smaller than conidia. Appl. Environ. Microbiol. 71 (1), 114–122. https://journals.asm.org/doi/10.1128/AEM.71.1.114-122.2005.

Cheneval, E., Busque, M.-A., Ostiguy, C., Lavoie, J., Bourbonnais, R., Labrèche, F., Zayed, J., 2015. Les emplois verts au Québec: définition et appréciation de leurs risques chimiques ou biologiques potentiels pour la santé des travailleurs. IRSST. https://www.irsst.qc.ca/media/documents/PubIRSST/R-875.pdf?v=2021-12-13.

Cox, J., M'bareche, H., Lindsley, W.G., Duchaine, C., 2020. Field sampling of indoor bioaerosols. Aerosol science and technology. J. Am. Assoc. Aerosol Res. 2020 https://doi.org/10.1080/02786826.2019.1688759.

DECOS. Endotoxins-Health-based recommended occupational exposure limits., 2010. The Hague. Health Council of the Netherlands, publication No. 2010/04OSH.

Douwes, J., Thorne, P., Pearce, N., Heederik, D., 2003. Bioaerosol health effects and exposure assessment: progress and prospects. Ann. Occup. Hyg. 47 (3), 187–200. https://doi.org/10.1093/annhyg/meg032.

Eduard, W., 2007. Nordic Expert Group for Criteria Documentation of Health Risks from Chemicals, Arbetslivsinstitutet (Sweden), 2007. 139. Fungal spores. National Institute for Working Life, Stockholm.

Eduard, W., 2009. Fungal spores: a critical review of the toxicological and epidemiological evidence as a basis for occupational exposure limit setting. Crit. Rev. Toxicol. 39 (10), 799–864. https://doi.org/10.3109/10408440903307333.

Eduard, W., Halstensen, A.S., 2009. Quantitative exposure assessment of organic dust. Scand. J. Work Environ. Health Suppl (7), 30–35.

Eduard, W., Heederik, D., Duchaine, C., Green, B.J., 2012. Bioaerosol exposure assessment in the workplace: the past, present and recent advances. J. Environ. Monit. 14 (2), 334. https://doi.org/10.1039/C2EM10717A.

Gniadek, A., Krzyściak, P., Twarużek, M., Macura, A., 2017. Occurrence of fungi and cytotoxicity of the species: Aspergillus ochraceus, Aspergillus Niger and Aspergillus flavus isolated from the air of hospital wards. Int. J. Occup. Med. Environ. Health 30 (2), 231–239. https://doi.org/10.13075/ijomeh.1896.00841.

Gonçalves, P., Melo, A., Dias, M., Almeida, B., Caetano, L.A., Veríssimo, C., Viegas, C., et al., 2021. Azole-resistant Aspergillus fumigatus harboring the TR34/L98H mutation: first report in Portugal in environmental samples. Microorganisms 9 (57). https://doi.org/10.3390/microorganisms9010057.

Goyer, N., Lavoie, J., Lazure, L., Marchand, G., 2005. Les bioaérosols en milieu de travail guide d'évaluation, de contrôle et de prévention. IRSST. https://www.irsst.qc.ca/media/documents/PubIRSST/T-23.pdf?v=2021-12-13.

Hagemeyer, O., Bünger, J., van Kampen, V., Raulf-Heimsoth, M., Drath, C., Merget, R., Brüning, Th, et al., 2013. Occupational Allergic Respiratory Diseases in Garbage Workers: Relevance of Molds and Actinomycetes. In Neurobiology of Respiration. Dordrecht: Springer, Netherlands. https://doi.org/10.1007/978-94-007-6627-3_42. Mieczyslaw Pokorski, ed. 313–320.

Hernandez, A.F., Buha, A., Constantin, C., Wallace, D.R., Sarigiannis, D., Neagu, M., Biljana, et al., 2019. Critical assessment and integration of separate lines of evidence for risk assessment of chemical mixtures. Arch. Toxicol. 93 (10), 2741–2757. https://doi.org/10.1007/s00204-019-02547-x.

Hindson, B.J., Ness, K.D., Masquelier, D.A., Belgrader, P., Heredia, N.J., Makarewicz, A.J., Bright, I.J., et al., 2011. High-throughput droplet digital PCR system for absolute quantitation of DNA copy number. Anal. Chem. 83 (22), 8604–8610.

Huttunen, K., Rintala, H., Hirvonen, M.R., Vepsäläinen, A., Hyvärinen, A., Meklin, T., Toivola, M., et al., 2008. Indoor air particles and bioaerosols before and after renovation of moisture-damaged buildings: the effect on biological activity and microbial flora. Environ. Res. 107 (3), 291–298. https://doi.org/10.1016/j.envres.2008.02.008.

IARC, 1993. Monographs on the Evaluation of Carcinogenic Risks to Humans: Some Naturally Occurring Substances: Food Items and Constituents, Heterocyclic Aromatic Amines and Mycotoxins, vol. 56. France: International Agency for Research on Cancer, Lyon, pp. 1–599.

IARC, 2002. Traditional herbal medicines, some mycotoxins, naphthalene and styrene. IARC Monographs on the evaluation of carcinogenic risks to humans. In: International Agency for Research on Cancer, vol. 82. France, Lyon, pp. 1–556.

INRS, 2017. Prélèvement des Aérosols par le dispositif CIP-10. http://www.inrs.fr/dms/inrs/PDF/metropol-prelevement-cip10.pdf.

Ivens, U.I., Breum, N.O., Ebbehøj, N., Nielsen, B.H., Poulsen, O.M., Würtz, H., 1999. Exposure-response relationship between gastrointestinal problems among waste collectors and bioaerosol exposure. Scand. J. Work Environ. Health 25 (3), 238–245. https://doi.org/10.5271/sjweh.430.

Johanning, E., Gareis, M., Nielsen, K., Dietrich, R., Märtlbauer, E., 2002. Airborne mycotoxin sampling and screening analysis. In: Proceedings: Indoor Air: 6.

Kalwasińska, A., Burkowska, A., Brzezinska, M.S., 2014. Exposure of workers of municipal landfill site to bacterial and fungal aerosol: exposure of workers of municipal landfill site. Clean - Soil, Air, Water 42 (10), 1337–1343. https://doi.org/10.1002/CLEN.201300385.

Knutsen, A.P., Bush, R.K., Demain, J.G., Denning, D.W., Dixit, Fairs A., Greenberger, P.A., et al., 2012. Fungi and allergic lower respiratory tract diseases. J. Allergy Clin. Immunol. 129 (2), 280–291. https://doi.org/10.1016/j.jaci.2011.12.970.

Krajewski, J.A., Tarkowski, S., Cyprowski, M., Szarapińska-Kwaszewska, J., Dudkiewicz, B., 2002. Occupational exposure to organic dust associated with municipal waste collection and management. Int. J. Occup. Med. Environ. Health 15 (3), 289–301. PMID: 12462456.

Land, C., Hult, K., Fuchs, R., Hagelberg, S., Lundstrom, H., 1987. Tremorgenic mycotoxins from Aspergillus fumigatus as a possible occupational health problem in sawmills. Appl. Environ. Microbiol. 193, 262–275. https://doi.org/10.1128/aem.53.4.787-790.1987.

Lavoie, J., Dunkerley, C.J., Kosatsky, T., Dufresne, A., 2006. Exposure to aerosolized bacteria and fungi among collectors of commercial, mixed residential, recyclable and compostable waste. Sci. Total Environ. 370 (1), 23–28. https://doi.org/10.1016/j.scitotenv.2006.05.016.

Le Goff, O., Bru-Adan, V., Bacheley, H., Godon, J.J., Wéry, N., 2010. The microbial signature of aerosols produced during the thermophilic phase of composting: diversity in compost bioaerosols,. J. Appl. Microbiol. 108 (1), 325–340. https://doi.org/10.1111/j.1365-2672.2009.04427.x.

Mack, S.M., Madl, A.K., Pinkerton, K.E., 2019. Respiratory Health Effects of Exposure to Ambient Particulate Matter and Bioaerosols. In Comprehensive Physiology. American Cancer Society, pp. 1–20. https://doi.org/10.1002/cphy.c180040.

Malmros, P., Sigsgaard, T., Bach, B., 1992. Occupational health problems due to garbage sorting. Waste Manag. Res. 10 (3), 227–234. https://10.1016/0734-242X(92)90101-P.

Malta-Vacas, J., Viegas, S., Sabino, R., Viegas, C., 2012. Fungal and microbial volatile organic compounds exposure assessment in a waste sorting plant. J. Toxicol. Environ. Health Part A 75 (22–23), 1410–1417. https://doi.org/10.1080/15287394.2012.721175.

Madsen, A.M., Frederiksen, M.W., Jacobsen, M.H., Tendal, K., 2020. Towards a risk evaluation of workers' exposure to handborne and airborne microbial species as exemplified with waste collection workers. Environ. Res. 183 (April), 109177. https://doi.org/10.1016/j.envres.2020.109177.

Madsen, A.M., Matthiesen, C.B., Frederiksen, M.W., Frederiksen, M., Frankel, M., Spilak, M., Gunnarsen, L., et al., 2012. Sampling, extraction and measurement of bacteria, endotoxin, fungi and inflammatory potential of settling indoor dust. J. Environ. Monit. 14 (12), 3230. https://doi.org/10.1039/c2em30699a.

Madsen, A.M., Frederiksen, M.W., Kurdi, I.M., Sommer, S., Flensmark, E., Tendal, K., 2019. Expanded cardboard waste sorting and occupational exposure to microbial species. Waste Manag. 87 (March), 345–356. https://doi.org/10.1016/j.wasman.2019.02.018.

Madsen, A.M., Alwan, T., Ørberg, A., Uhrbrand, K., Jørgensen, M.B., 2016. Waste workers' exposure to airborne fungal and bacterial species in the truck cab and during waste collection. Ann. Occup. Hyg. 60 (6), 651–668. https://doi.org/10.1093/annhyg/mew021.

Marchand, G., 2021. Chapitre 8: microorganismes et leurs sous-produits. In: Hygiène du travail (Ed.), du diagnostic à la maîtrise des facteurs de risques. Montréal: TC Média livres inc, pp. 165–196, 2é édition, AQHSST.
Marchand, G., Bonifait, L., Veillette, M., Pépin, C., Beaudet, Y., Légaré, É. Lavoie L., et al., 2017a. Évaluation des bioaérosols et des composés gazeux émis lors des compostages de résidus agroalimentaires et résidentiels. IRSST. http://www.irsst.qc.ca/media/documents/PubIRSST/R-960.pdf.
Marchand, G., Gardette, M., Nguyen, K., Amano, V., Neesham-Grenon, E., Debia, M., 2017b. Assessment of workers' exposure to grain dust and bioaerosols during the loading of vessels' hold: an example at a port in the province of Québec. Ann. Work Expos. Health 61 (7), 836–843. https://doi.org/10.1093/annweh/wxx045.
Marchand, G., 2009. Analyse des endotoxines. Méthode analytique MA-332. Montréal: IRSST(institut de recherche Robert Sauvé en santé et sécurité du travail). http://www.irsst.qc.ca/media/documents/pubirsst/M-332.pdf.
Mbareche, H., Veillette, M., Bilodeau, G.J., Duchaine, C., 2019. Fungal aerosols at dairy farms using molecular and culture techniques. Sci. Total Environ. 653 (February), 253–263. https://doi.org/10.1016/j.scitotenv.2018.10.345.
McKean, C., Tang, L., Tang, M., Billam, M., Wang, Z., Theodorakis, C.W., Kendall, R.J., et al., 2006. Comparative acute and combinative toxicity of aflatoxin B1 and fumonisin B1 in animals and human cells. Food Chem. Toxicol.: Int. J. Publ. Brit. Indus. Biol. Res. Assoc. 44 (6), 868–876. https://doi.org/10.1016/j.fct.2005.11.011.
Mousavi, B., Hedayati, M.T., Hedayati, N., Ilkit, M., Syedmousavi, S., 2016. Aspergillus species in indoor environments and their possible occupational and public health hazards. Curr. Med. Mycol. 2 (1), 36–42. https://doi.org/10.18869/acadpub.cmm.2.1.36.
Ncube, F., Ncube, E.J., Voyi, K., 2017. Bioaerosols, noise, and ultraviolet radiation exposures for municipal solid waste handlers. J. Environ. Publ. Health 3081638. https://doi.org/10.1155/2017/3081638.
Nikolic, M., Sustersic, T., Filipovic, N., 2018. In vitro models and on-chip systems: biomaterial interaction studies with tissues generated using lung epithelial and liver metabolic cell lines. Front. Bioeng. Biotechnol. 3 (6), 120. https://doi.org/10.3389/fbioe.2018.00120.
Nielsen, E.M., Nielsen, B.H., Breum, N.O., 1995. Occupational bioaerosol exposure during collection of household waste. Ann. Agric. Environ. Med. 2, 53–59.
Okoth, S., Nyongesa, B., Ayugi, V., Kang'ethe, E., Korhonen, H., Joutsjoki, V., 2012. Toxigenic potential of Aspergillus species occurring on maize kernels from two agro-ecological zones in Kenya. Toxins 4 (11), 991–1007. https://doi.org/10.3390/toxins4110991.
Park, D., Ryu, S., Kim, S., Byun, H., Yoon, C., Lee, K., 2013. Airborne bacteria and fungi associated with waste-handling work. Int. J. Occup. Environ. Health 19 (4), 311–318. https://doi.org/10.1179/2049396713Y.0000000036.
Park, D.U., Ryu, S.H., Kim, S.B., Yoon, C.S., 2011. An assessment of dust, endotoxin, and microorganism exposure during waste collection and sorting. J. Air Waste Manag. Assoc. 61 (4), 461–468. https://doi.org/10.3155/1047-3289.61.4.461.
Pasanen, A.L., Nikulin, M., Tuomainen, M., Berg, S., Parikka, P., Hintikka, E.L., 1993. Laboratory experiments on membrane filter sampling of airborne mycotoxins produced by stachybotrys atra corda. Atmos. Environ. Part A. Gener. Top. 27 (1), 9–13. https://doi.org/10.1016/0960-1686(93)90065-7.
Picard-Masson, M., Lajoie, E., Lord, J., Lalancette, C., Marchand, G., Levac, E., Lemieux, M.A., et al., 2016. Two related occupational cases of Legionella longbeachae infection, Quebec, Canada. Emerg. Infect. Dis. 22 (7), 1289–1291. https://doi.org/10.3201/eid2207.160184.
Piecková, E., 2012. Adverse health effects of indoor moulds/štetni zdravstveni učinci plijesni unutarnjih prostora. Arh. Hig. Rada. Toksikol. 63, 545–549. https://doi.org/10.2478/10004-1254-63-2012-2221.
Price, R.G., Wildeboer, D., 2017. E. coli as an indicator of contamination and health risk in environmental waters. *Escherichia coli* - Eec. Adv. Physiol. Pathog. Biotechnol. Appl. https://doi.org/10.5772/67330.
Poole, C.J.M., Wong, M., 2013. Allergic bronchopulmonary aspergillosis in garden waste (compost) collectors–occupational implications. Occup. Med. 63 (7), 517–519. https://doi.org/10.1093/occmed/kqt097.
Poulsen, O.M., Breum, N.O., Ebbehøj, N., Hansen, A.M., Ivens, U.I., van Lelieveld, D., Malmros, P., et al., 1995. Sorting and recycling of domestic waste. Review of occupational health problems and their possible causes. Sci. Total Environ. 168 (1), 33–56. https://doi.org/10.1016/0048-9697(95)04521-2.
Principe, L., Tomao, P., Visca, P., 2017. Legionellosis in the occupational setting. Environ. Res. 152, 485–495. https://doi.org/10.1016/j.envres.2016.09.018.
Rački, N., Dreo, T., Gutierrez-Aguirre, I., Blejec, A., Ravnikar, M., 2014. Reverse transcriptase droplet digital PCR shows high resilience to PCR inhibitors from plant, soil and water samples. Plant Methods 10 (1), 42. https://doi.org/10.1186/s13007-014-0042-6.
Research International inc., SASS® 3100 Dry Air Sampler Data Sheet. Access 2022-04-12.
Resendiz, S., Lagrou, A.K., Meis, J.F., Chowdhary, A., Lockhart, S.R., 2018. Verweij, P.E., the ISHAM/ECMM Aspergillus resistance surveillance working group triazole resistance surveillance in Aspergillus fumigatus. Med. Mycol. 56 (Suppl. l_1), S83–S92. https://doi.org/10.1093/mmy/myx144.
Roca-Barcelo, A., Douglas, P., Fecht, D., Sterrantino, A.F., Williams, B., Blangiardo, M., Gulliver, J., et al., 2020. Risk of respiratory hospital admission associated with modelled concentrations of Aspergillus fumigatus from composting facilities in England. Environ. Res. (January), 108949. https://doi.org/10.1016/j.envres.2019.108949.
Rosas, I., Salinas, E., Yela, A., Calva, E., Eslava, C., Cravioto, A., 1997. Escherichia coli in settled-dust and air samples collected in residential environments in Mexico city. Appl. Environ. Microbiol. 63 (10), 4093–4095. https://doi.org/10.1128/aem.63.10.4093-4095.1997.
Roy, R., Jan, R., Joshi, U., Bhor, R., Pai, K., Gursumeeran Satsangi, P., 2020. Characterization, pro-inflammatory response and cytotoxic profile of bioaerosols from urban and rural residential settings in Pune, India. Environ. Pollut. 264 (September), 114698. https://doi.org/10.1016/j.envpol.2020.114698.
Russell, C., Mitchell, J., Godish, T., 1999. Apparent viability of airborne mould spores/particles determined from culturable/viable and total mould spore sampling methods. Proc. Indoor Air 1999, 934–938.
Rylander, R., 2002. Endotoxin in the environment-exposure and effects. J. Endotoxin Res. 8 (4), 241–252. https://doi.org/10.1179/096805102125000452.
Sabino, R., Carolino, E., Veríssimo, C., Martinez, M., Clemons, K.V., Stevens, D.A., 2016. Antifungal susceptibility of 175 Aspergillus isolates from various clinical and environmental sources. Med. Mycol. 54 (7), 740–756. https://doi.org/10.1093/mmy/myw024.
Sabino, R., Veríssimo, C., Viegas, C., Viegas, S., Brandao, J., Alves-Correia, M., Borrego, L.M., et al., 2019. The role of occupational Aspergillus exposure in the development of diseases. Med. Mycol. 57 (Suppl. ment_2), S196–S205. https://doi.org/10.1093/mmy/myy090.
Samake, A., Uzu, G., Martins, J.M.F., Calas, A., Vince, E., Parat, S., Jaffrezo, J.L., 2017. The unexpected role of bioaerosols in the oxidative potential of PM. Sci. Rep. 7 (10978) https://doi.org/10.1038/s41598-017-11178-0.
Segura, P.A., Matthieu, M., Gagnon, C., Sauvé, S., 2009. Review of the occurrence of anti-infectives in contaminated wastewaters and natural and drinking waters. Environ. Health Perspect. 117 (5), 675–684. https://doi.org/10.1289/ehp.11776.
"Stop Neglecting Fungi", 2017. Nat. Microbiol. 2 (8), 1–2. https://doi.org/10.1038/nmicrobiol.2017.120.
Suva, Protection de la santé au poste de travail, 2021. Valeurs limites d'exposition aux postes de travail: valeurs VME/VLE (remarques), risques physiques, contraintes physiques [WWW Document]. https://www.suva.ch/fr-CH/materiel/directives-et-textes-de-lois/erlaeuterungen-zu-den-grenzwerten/. (Accessed 31 May 2022).
Swain, R.J., Kemp, S.J., Goldstraw, P., Tetley, T.D., Stevens, M.M., 2010. Assessment of cell line models of primary human cells by Raman spectral phenotyping. Biophys. J. 98, 1703–1711.
Sykes, P., Morris, R.H.K., Allen, J.A., Wildsmith, J.D., Jones, K.P., 2011. Workers' exposure to dust, endotoxin and β-(1–3) glucan at four large-scale composting facilities. Waste Manag. 31 (3), 423–430. https://doi.org/10.1016/j.wasman.2010.10.016.
Szyłak-Szydłowski, M., Kulig, A., Miaśkiewicz-Pęska, E., 2016. Seasonal changes in the concentrations of airborne bacteria emitted from a large wastewater treatment plant. Int. Biodeterior. Biodegrad. 115, 11–16. https://linkinghub.elsevier.com/retrieve/pii/S0964830516302311.
Taylor, S.C., Laperriere, G., Germain, H., 2017. Droplet digital PCR versus QPCR for gene expression analysis with low abundant targets: from variable nonsense to publication quality data. Sci. Rep. 7 (1), 1–8. https://doi.org/10.1038/s41598-017-02217-x.
Taylor, S.C., Carbonneau, J., Shelton, D.N., Boivin, G., 2015. Optimization of droplet digital PCR from RNA and DNA extracts with direct comparison to RT-QPCR: clinical implications for quantification of oseltamivir-resistant subpopulations. J. Virol Methods 224 (November), 58–66. https://doi.org/10.1016/j.jviromet.2015.08.014.
Taroncher, M., Pigni, M.C., Diana, M.N., Juan-García, A., Ruiz, M.J., 2020. Does low concentration mycotoxin exposure induce toxicity in HepG2 cells through oxidative stress? Toxicol. Mech. Methods 30 (6), 417–426. https://doi.org/10.1080/15376516.2020.1757000.
van Heijnsbergen, E., Schalk, J.A.C., Euser, S.M., Brandsema, P.S., den Boer, J.W., de Roda Husman, A.M., 2015. Confirmed and potential sources of Legionella reviewed. Environ. Sci. Technol. 49, 4797–4815. https://doi.org/10.1021/acs.est.5b00142.
Varga, J., Baranyi, N., Chandrasekaran, M., Vágvölgyi, C., Kocsubé, S., 2015. Mycotoxin producers in the Aspergillus genus : an update. Acta Biol. Szeged. 59 (2), 151–167. http://www2.sci.u-szeged.hu/ABS.
Verhaegen, B., De Reu, K., De Zutter, L., Verstraete, K., Heyndrickx, M., Van Coillie, E., 2016. Comparison of droplet digital PCR and QPCR for the quantification of shiga toxin-producing Escherichia coli in bovine feces. Toxins 8 (5), 157. https://doi.org/10.3390/toxins8050157.
Viegas, C., Caetano, L.A., Viegas, S., 2021a. Occupational exposure to Aspergillus section Fumigati: tackling the knowledge gap in Portugal. Environ. Res. 194 (March), 110674. https://doi.org/10.1016/j.envres.2020.110674.
Viegas, C., Sousa, P., Dias, M., Caetano, L.A., Ribeiro, E., Carolino, E., Twarużek, M., et al., 2021b. Bioburden contamination and Staphylococcus aureus colonization associated with firefighter's ambulances. Environmental Research 197 (April), 111125. https://doi.org/10.1016/j.envres.2021.111125.
Viegas, C., Caetano, L.A., Cox, J., Korkalainen, M., Haines, S.R., Dannemiller, K., Viegas, S., et al., 2020a. The Effects of Waste Sorting in Environmental Microbiome, THP-1 Cell Viability and Inflammatory Responses. Environmental Research 185 (June), 109450. https://doi.org/10.1016/j.envres.2020.109450.
Viegas, C., Dias, M., Almeida, B., Caetano, L.A., Carolino, E., Quintal-Gomes, A., Twarużek, M., et al., 2020b. Are Workers from Waste Sorting Industry Really Protected by Wearing Filtering Respiratory Protective Devices? The Gap between the Myth and Reality. Waste Manag. 102, 856–867. https://doi.org/10.1016/j.wasman.2019.12.001.
Viegas, C., Twarużek, M., Dias, M., Almeida, B., Carolino, E., Soszczyńska, E., Ałtyn, I., et al., 2020c. Cytotoxic Effect of Filtering Respiratory Protective Devices from the Waste Sorting Industry: Is in Vitro Toxicology Useful for Risk Characterization? Environmental Research 191 (December), 110134. https://doi.org/10.1016/j.envres.2020.110134.
Viegas, S., Caetano, L.A., Korkalainen, M., Faria, T., Pacífico, C., Carolino, E., Quintal-Gomes, A., et al., 2017a. Cytotoxic and Inflammatory Potential of Air Samples from Occupational Settings with Exposure to Organic Dust. Toxics 5 (1). https://doi.org/10.3390/toxics5010008.

Viegas, C., Faria, T., de Oliveira, A.C., Caetano, L.A., Carolino, E., Quintal-Gomes, A., Twarużek, M., et al., 2017b. A New Approach to Assess Occupational Exposure to Airborne Fungal Contamination and Mycotoxins of Forklift Drivers in Waste Sorting Facilities. Mycotoxin Research 33 (4), 285–295. https://doi.org/10.1007/s12550-017-0288-8.

Viegas, C., Pinheiro, C., Sabino, R., Viegas, S., Brandão, J., Veríssimo, C., 2015. In: Environmental Mycology in Public Health: Fungi and Mycotoxins Risk Assessment and Management. Academic Press.

Viegas, C., Gomes, A.Q., Abegão, J., Sabino, R., Graça, T., Viegas, S., 2014. Assessment of fungal contamination in waste sorting and incineration - Case study in Portugal. J. Toxicol. Environ. Health. Part A 77 (1-3), 57–68. https://doi.org/10.1080/15287394.2014.865583.

Viegas, C., Viegas, S., Monteiro, A., Carolino, E., Sabino, R., Veríssimo, C., 2012. Comparison of indoor and outdoor fungi and particles in poultry units. Environ. Impact 62, 589–596. https://doi.org/10.2495/EID120511.

Viegas, S., Veiga, L., Figueredo, P., Almeida, A., Carolino, E., Sabino, R., Veríssimo, C., et al., 2012b. Occupational exposure to aflatoxin (AFB1) in poultry production. Journal of Toxicology and Environmental Health, Part A 75, 1330–1340. https://doi.org/10.3920/WMJ2012.1531.

Von Essen, S., Donham, K., 1999. Illness and Injury in Animal Confinement Workers. Occupational Medicine 14 (2), 337–350. PMID: 10329909.

Wéry, N., 2014. Bioaerosols from Composting Facilities- a Review. Frontiers in Cellular and Infection Microbiology 4 (42). https://doi.org/10.3389/fcimb.2014.00042.

Whiley, H., Bentham, R., 2011. Legionella longbeachae and Legionellosis. Emerging Infectious Diseases 17, 579–583. https://doi.org/10.3201/eid1704.100446.

Wouters, I.M., Spaan, S., Douwes, J., Doekes, G., Heederik, D., 2006. Overview of Personal Occupational Exposure Levels to Inhalable Dust, Endotoxin, Beta(1–>3)-Glucan and Fungal Extracellular Polysaccharides in the Waste Management Chain. The Annals of Occupational Hygiene 50 (1), 39–53. https://doi.org/10.1093/annhyg/mei047.

Wouters, I., Hilhorst, S., Kleppe, P., Doekes, G., Douwes, J., Peretz, C., Heederik, D.D., 2002. Upper airway inflammation and respiratory symptoms in domestic waste collectors. Occupat. Environ. Med. 59 (2), 106–112. https://doi.org/10.1136/oem.59.2.106.