

From Microbial Characterisation to Mechanistic Dose–Response

Contributing an Integrated Framework in Waste Sorting Industry

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The Challenge: Complex Occupational Exposures



Workers in waste sorting facility exposed to complex environmental hazards

⚠ Occupational Health Challenges ^{1,2}

🦠 Complex Fungal Contamination

Workers exposed to complex mixtures of fungal contaminants across sorting plants and collection trucks.

🧪 Mycotoxin Exposure Routes

Multiple exposure pathways including inhalation, dermal contact, and inadvertent ingestion.

❓ Poorly Characterized Risks

Limited understanding from exposure science perspective despite waste management being essential.

🛡 Protection Gaps

Current protective measures may be insufficient given contamination complexity.




Critical Need

Systematic research is essential to characterise exposure patterns, understand health implications, and develop evidence-based protection strategies for waste industry workers.




10+ Years of Systematic Research: Key Findings

Research Evolution Across Waste Management Environments

Early Phase (2014-2017)

-  Initial fungal characterization in Portuguese sorting plants revealed consistent *Aspergillus* contamination patterns.
-  High counts of *Fumigati*, *Flavi*, and *Nigri* sections alongside *Penicillium* sp. documented.
-  Recognition of allergenicity and mycotoxin production capacity under favourable conditions.

Analytical Advancement (2017-2022)

-  LC-MS/MS multi-mycotoxin analysis implementation sharpened exposure characterisation significantly.
-  Waste collection trucks showed 53.1% mycotoxin contamination in settled dust samples.
-  Mechanical protection gloves demonstrated 89.6% contamination rates with multiple toxins present.

Cumulative Research Impact

10+

Years of Systematic Investigation

4

Primary Environments Studied

3

Countries with Evidence Base

11

Peer-Reviewed Publications

Fungal Characterisation: Portuguese Sorting Plants

Early Research Findings

Portuguese Waste Sorting Plants (2014-2015)

Initial fungal characterisation revealed consistently high counts of pathogenic species with significant implications for worker health and mycotoxin production potential.

Aspergillus section Fumigati

Consistently high counts with **allergenic potential** and capacity for mycotoxin production under favorable conditions

Aspergillus sections Flavi & Nigri

Significant presence of **aflatoxin-producing species** creating complex exposure scenarios

Penicillium sp.

Widespread contamination with **multiple mycotoxin producers** across sorting facilities

Key Publications

Viegas et al. (2014, 2015) established the foundation for understanding occupational fungal exposure in waste management, demonstrating the scale of contamination challenge.



Journal of Toxicology and Environmental Health, Part A: Current Issues

Publication details, including instructions for authors and subscription information:
<http://www.tandfonline.com/loi/uteh20>

Assessment of Fungal Contamination in Waste Sorting and Incineration—Case Study in Portugal

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Fungal burden in waste industry: an occupational risk to be solved

Carla Viegas · Tiago Faria · Mateus dos Santos · Elisabete Carolino · Anita Quintal Gomes · Raquel Sabino · Susana Viegas

Mycotoxin Analysis: LC-MS/MS Revelations

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Research article

Microbial contamination in waste collection: Unveiling this Portuguese occupational exposure scenario


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Assessment of the microbial contamination of mechanical protection gloves used on waste sorting industry: A contribution for the risk characterization

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Advanced Analytical Findings

LC-MS/MS Multi-Mycotoxin Analysis

Comprehensive analytical approach revealing multiple exposure pathways beyond traditional inhalation routes, with significant contamination detected across different protective equipment types.

Waste Collection Trucks

53.1% positive samples with mycophenolic acid most frequent (up to 170.1 ng/g)

Mechanical Protection Gloves

89.6% positive samples with multiple toxins including roquefortine C and griseofulvin

Multiple Exposure Routes

Dermal contact and inadvertent ingestion pathways identified beyond inhalation

Key Mycotoxins Detected

Sterigmatocystin, deoxynivalenol, zearalenone, mycophenolic acid, roquefortine C, and griseofulvin across different matrices.

Protection Equipment: Unexpected Contamination

FRPD Findings

Substantial Fungal Diversity

Interior layers contained *Aspergillus* sections *Fumigati*, *Flavi*, *Nigri*, and *Circumdati* despite filtration

Molecular Detection

Fumigati DNA detected where no growth obtained by culture, highlighting limitations of culture-based methods

Mycotoxin Detection

Mycotoxins rarely detected in FRPD compared to **89.6% positive** mechanical gloves

Protection Efficacy

Questions raised about actual protective capacity against complex fungal exposures in waste environments

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Waste Management

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Are workers from waste sorting industry really protected by wearing Filtering Respiratory Protective Devices? The gap between the myth and reality



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RESEARCH PAPER



Microbial Occupational Exposure in e-waste Recycling: Biological Hazards, Their Sources, and Potential Toxic Effects

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Protection Equipment: Unexpected Contamination



microorganisms



Article

Azole-Resistant *Aspergillus fumigatus* Harboring the TR₃₄/L98H Mutation: First Report in Portugal in Environmental Samples

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Resistance Concerns

Antifungal Resistance

Isolates showed susceptibility to medical azoles at EUCAST breakpoints, indicating environmental selection pressure

TR₃₄/L98H Mutation

First report of azole-resistant *Aspergillus fumigatus* in Portuguese environmental samples from waste facilities

Clinical Implications

Environmental resistance patterns may compromise treatment options for occupationally-acquired infections

Global Health Concern

Waste industry as potential reservoir for antifungal-resistant strains with broader public health implications

International Evidence: Global Patterns

International Research Expansion

Norwegian E-Waste Facilities

Mycotoxins detected in FRPD and settled dust, linked to inadequate protective equipment replacement protocols.

Canadian Waste Collectors

Fumigati at critical concentrations in breathing zones with cytotoxic effects in most air samples.

Automation Independence

Widespread toxigenic fungi are reported regardless of facility automation levels, with seasonal variation patterns.

Comprehensive assessment of occupational exposure to microbial contamination in waste sorting facilities from Norway

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

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Research Gaps: What We Still Don't Know

? Critical Knowledge Gaps



Co-exposure Effects

Complex interactions between multiple fungal species and mycotoxins remain uncharacterised



Exposure-Dose Relationships

Links between external exposure and internal biological dose are still unknown



Health-Based Limits

Absence of evidence-based exposure limits for complex fungal mixtures



Current Research Limitations

Despite extensive characterization work, cytotoxicity persists even when individual toxins are below detection limits, suggesting complex mixture effects that current methodologies cannot fully capture.

→ Future Research Priorities

Integrated approaches combining environmental monitoring, human biomonitoring, and mechanistic dose-response studies are essential to establish evidence-based risk mitigation strategies.

TOHC-Waste: An Integrated Solution



TOHCWaste Project

Total Occupational Health in Waste

Integrated multi-tiered approach addressing identified knowledge gaps through comprehensive methodology combining environmental monitoring, human biomonitoring, and advanced in vitro models.

FRPD as Passive Samplers

Analysis via culture, molecular tools, and comprehensive multi-mycotoxin quantification

Human Biomonitoring

Linking external exposure to internal dose through biomarkers in urine and blood

Advanced In Vitro Models

A549 lung, HepG2 liver, kidney cell lines, plus 3D human airway epithelium co-culture

Chemical co-exposure

Bisphenols, PFAs, metals, endotoxins

Innovative Methodologies

Integration of environmental monitoring, biomonitoring, and New Approach Methodologies to generate the first comprehensive dataset linking exposure to biological effects.

TOHC-Waste Methodology: Multi-Tiered Approach



Tackling the Occupational Health Concerns in the Waste-Sorting Industries



Environmental Monitoring

- FRPD as passive samplers (time-integrated)
- Culture-based methods (culturomics)
- Molecular tools (next-generation sequencing)
- Multi-mycotoxin quantification (LC-MS/MS)

Human Biomonitoring

- Urine biomarkers (mycotoxin metabolites)
- Blood analysis (systemic exposure biomarkers)
- Exposure-dose links (external → internal dose)
- Temporal monitoring (longitudinal assessment)

In Vitro Models

- Cell lines: Calu-3 & A549 (lung), HepG2 (liver), HEK 293 (kidney)
- 3D co-culture: THP-1 macrophages + Calu-3
- Endpoints: oxidative stress, inflammation, barrier integrity

Expected Impact: Evidence-Based Solutions

Expected Outcomes

First Comprehensive Dataset

Linking fungal and mycotoxin exposure to early biological effects in waste workers

Evidence-Based Exposure Limits

Establishing health-based limits for complex fungal mixtures in occupational settings

Risk Mitigation Strategies

Practical guidance for protecting workers in waste management environments

Scientific Innovation

Integration of environmental monitoring, biomonitoring, and New Approach Methodologies for comprehensive risk assessment.

Circular Economy Benefits

Industry Standards

Supporting development of sector-specific occupational health guidelines

Worker Protection

Enhanced safety protocols for the growing circular economy workforce

Policy Development

Informing regulatory frameworks for sustainable waste management practices

Sustainable Future

Enabling safe expansion of circular economy initiatives through evidence-based occupational health protection.

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Thanks for your attention and looking forward to hear your questions.