



Preliminary Assessment of Microbial Contamination from Winter Sampling Campaign in Portuguese Elementary School

Pedro Pena^{1,2}; Renata Cervantes^{1,2}; Carla Viegas^{1,2}

1 H&TRC—Health & Technology Research Center, ESTeSL—Escola Superior de Tecnologia e Saúde, Instituto Politécnico de Lisboa, 1990-096 Lisbon, Portugal;

2 NOVA National School of Public Health, Public Health Research Centre, Comprehensive Health Research Center, CHRC, REAL, CCAL, NOVA University Lisbon, Lisbon, Portugal

pedro.pena@estesl.ipl.pt



Improving **indoor air quality**
to bring about a **healthier future**
for our children



IAQ IN SCHOOLS AND HEALTH RISKS^{1,2}

- Microbial contamination risks health in indoor environments.
- Schools: high risk for respiratory/infectious diseases.
- Children in schools are especially vulnerable.
- They spend long hours in these settings.
- Indoor air quality (IAQ) management is critical.



ADDRESSING AIR QUALITY INDOORS

- Assessing airborne bacteria and fungi remain crucial for the prevention of respiratory health problems³
- Active air sampling remains the gold standard for compliance with legal frameworks on indoor air quality⁴
- Passive sampling methods, such as settled dust, are simple and cost-effective⁵
- Allow longer sampling periods, providing insights into prolonged exposure³



MAS-100

VS.



Settled dust collected through a UV-sterilized filters

Objective: assess bacterial and fungal contamination across rural and urban elementary school areas

SAMPLING CAMPAIGN

- Conducted in **10 schools** within the Metropolitan Lisbon Area.
- Samples collected during **cold season (N=10)**.
- Locations sampled: **classrooms (N=10)**
- **Sampling Method:** Settled dust collected using UV-sterilized coffee filters placed inside disinfected vacuum tubes.
- **Surfaces Sampled:** Shelves, plinths, and floors around students' desks and near the door in all sampled rooms.



FILTERS EXTRACTION AND INOCULATION

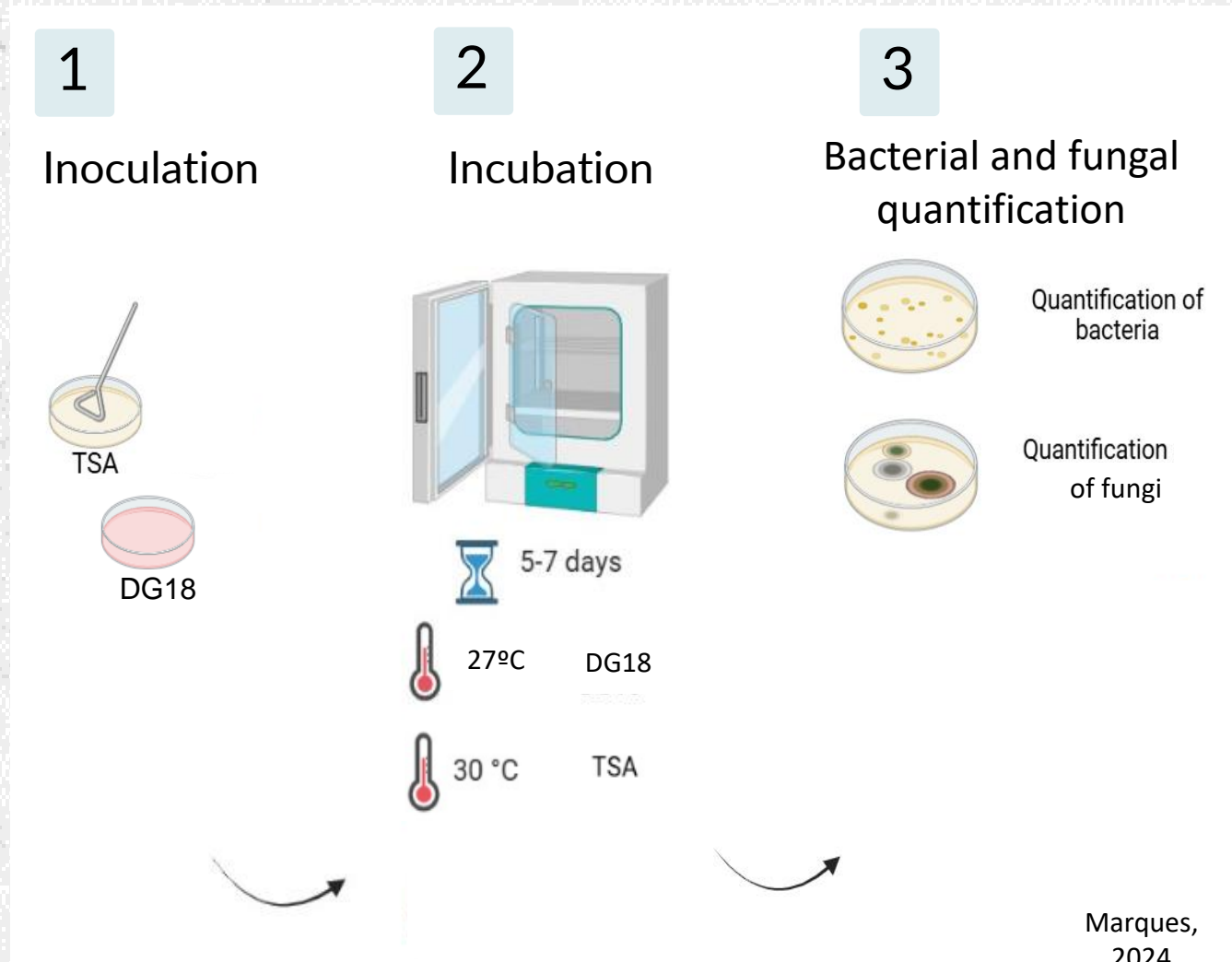
Extraction Method: Samples extracted using a NaCl + Tween 80 solution.

Extracted samples inoculated onto DG18 (Dichloran Glycerol) and Tryptic Soy Agar (TSA)

Incubation Conditions:

DG18: Incubated at 27°C for 4-6 days.

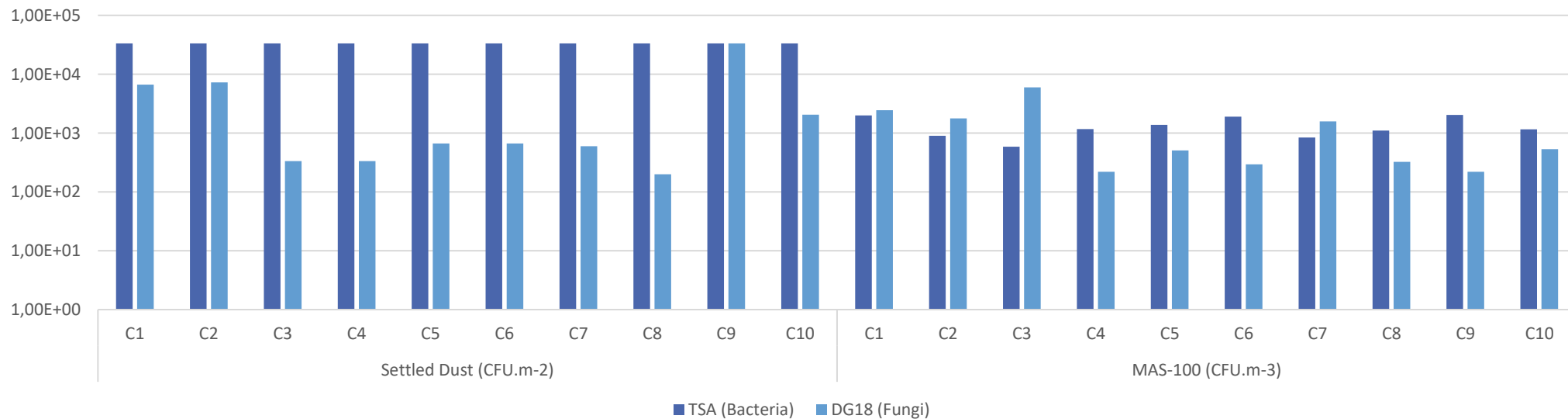
TSA: Incubated at 30°C for 7 days.



BACTERIAL VS. FUNGAL CONTAMINATION

Cold Season Contamination:

- The MAS-100 (CFU/m³) data shows high variability from one classroom to the next (e.g., TSA from 590 to 2040; DG18 from 220 to 5965).
- For Settled dust:
 - Bacteria (TSA): The settled dust (CFU/m²) values are uniformly high and identical (33.333) across all classrooms.
 - Fungi (DG18): The settled dust values show a different pattern of contamination than the active sampler.

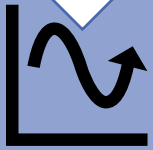


Bacterial vs. fungal contamination

- Bacteria (TSA): Uniformly High in All Classrooms
 - Settled dust shows a consistent, high-level bacterial reservoir on surfaces everywhere.
 - Active air sampling failed to identify this universal baseline problem.
- Fungi (DG18): The Case of Classroom C9
 - Active Air (Snapshot): Clean → "False sense of safety."
 - Settled Dust (Cumulative): Extreme contamination → "Massive hidden reservoir."



Highlights



Sampling methods employed are complementary tools, providing two essential pieces of the risk puzzle.^{6,7}



Relying solely on air snapshots provides a false sense of safety and an incomplete picture.



Combining both methods is necessary to see both the instantaneous airborne threat and the historical exposure potential.

This comprehensive view is crucial for effective public health interventions and managing infection risks in schools⁸.

Next Steps

Contextual and operational information collection

- Cleaning procedures applied

Compare results from both seasons:

- MAS-100
- Personal air samplers
- Andersen 6-stage air sampler
- Passive samples (EDC, EDCT, Settled dust, Dust filers, Mops)

Risk assessment

Fill the gaps in IAQ policies, supporting regulators and exposure assessors on primary schools' IAQ improvement.

References

1. Heyder J. Deposition of inhaled particles in the human respiratory tract and consequences for regional targeting in respiratory drug delivery. *Proc Am Thorac Soc.* 2004;1(4):315–20.
2. Viegas C, Sousa P, Dias M, Caetano LA, Ribeiro E, Carolino E, Twarużek M, Kosicki R, Viegas S. Bioburden contamination and *Staphylococcus aureus* colonization associated with firefighter's ambulances. *Environ Res.* 2021 Jun;197:111125. doi: 10.1016/j.envres.2021.111125. Epub 2021 Apr 22. PMID: 33895113.
3. Viegas C, Peixoto C, Gomes B, Dias M, Cervantes R, Pena P, Slezakova K, Pereira MDC, Morais S, Carolino E, Twarużek M, Viegas S, Caetano LA. Assessment of Portuguese fitness centers: Bridging the knowledge gap on harmful microbial contamination with focus on fungi. *Environ Pollut.* 2024 Jun 1;350:123976. doi: 10.1016/j.envpol.2024.123976. Epub 2024 Apr 22. PMID: 38657893.
4. Napoli C, Marcotrigiano V, Montagna MT. Air sampling procedures to evaluate microbial contamination: a comparison between active and passive methods in operating theatres. *BMC Public Health.* 2 August 2012;12(1):594.
5. Viegas C, Dias M, Viegas S. Electrostatic Dust Cloth: A Useful Passive Sampling Method When Assessing Exposure to Fungi Demonstrated in Studies Developed in Portugal (2018–2021). *Pathogens.* Mars de 2022;11(3):345.
6. Fujiyoshi S, Tanaka D, Maruyama F. Transmission of Airborne Bacteria across Built Environments and Its Measurement Standards: A Review. *Front Microbiol* [Internet]. 2017 [cited on 23 Jun 2023]. Available on: <https://www.frontiersin.org/articles/10.3389/fmicb.2017.02336>
7. El Kased R, Gamaleldin N. Prevalence of Bacteria in Primary Schools. *J Pure Appl Microbiol.* 31 de dezembro de 2020;14:2627–36.
8. WHO. WHO Guidelines for Indoor Air Quality: Dampness and Mould. In: WHO Guidelines for Indoor Air Quality: Dampness and Mould [Internet]. World Health Organization; 2009 [cited on 10 July 2023]. Available on: <https://www.ncbi.nlm.nih.gov/books/NBK143944/>

Acknowledgments

H&TRC authors gratefully acknowledge FCT/MCTES UIDP/05608/2020 (<https://doi.org/10.54499/UIDP/05608/2020>) and UIDB/05608/2020 (<https://doi.org/10.54499/UIDB/05608/2020>). This work is also supported by national funds through FCT/MCTES/FSE/UE, 2023.01366.BD (<https://doi.org/10.54499/2023.01366.BD>); UI/BD/153746/2022 and CE3C unit UIDB/00329/2020 (<https://doi.org/10.54499/UIDB/00329/2020>); UI/BD/151431/2021 (<https://doi.org/10.54499/UI/BD/151431/2021>); and Instituto Politécnico de Lisboa, national support through IPL/2022/InChildhealth/BI/12M; and for funding the projects IPL/IDI&CA2024/WWTPSValor_ESTeSL and IPL/IDI&CA2024/MycoSOS_ESTeSL, and the Academy of Medical Sciences (SBF007/100130). InChildHealth is funded by the European Union (Grant Agreement No. 101056883) via the European Health and Digital Executive Agency (HaDEA). Additional funding is provided by the Swiss State Secretariat for Education, Research and Innovation (SERI; Grant 22.00324), UK Research and Innovation (UKRI; Grant 10040524), and the Australian National Health & Medical Research Council (NHMRC; Grants APP2017786 and APP2008813). The views expressed are those of the authors and do not necessarily reflect those of HaDEA, the EU, or any funding agency.



Funded by
the European Union



Project funded by



Schweizerische Eidgenossenschaft
Confédération suisse
Confederazione Svizzera
Confederaziun svizra

Swiss Confederation



UK Research
and Innovation

Federal Department of Economic Affairs,
Education and Research EAER
State Secretariat for Education,
Research and Innovation SERI





THANK YOU

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

