

Nov 14, 2025

Shoe sole dust sampling protocol

DOI

<https://dx.doi.org/10.17504/protocols.io.dm6gpmkb5gzp/v1>

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Protocol Citation: Martin Täubel, Sadia Marija Ferdous, Bridget Hegarty 2025. Shoe sole dust sampling protocol. **protocols.io** <https://dx.doi.org/10.17504/protocols.io.dm6gpmkb5gzp/v1>

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Protocol status: Working

We use this protocol and it's working

Created: November 13, 2025



Last Modified: November 14, 2025

Protocol Integer ID: 232237

Keywords: Shoe Sole Dust Sampling, Microbial Quantification, shoe sole dust samples for microbial quantification, shoe sole dust sampling protocol this protocol, sole dust sampling protocol, sole dust sample, collecting shoe, microbial data, microbial quantification, shoe, protocol, procedure

Funders Acknowledgements:

Finnish Institute for Health and Welfare

Dr Hegarty's startup funds from the Case School of Engineering at Case Western Reserve University

Abstract

This protocol describes the procedure for collecting shoe sole dust samples for microbial quantification. For example, as part of an epidemiological study, to collect microbial data during a walk of a specific length.

Guidelines

- An appropriate swab type should be selected based on the study.
- It is recommended to collect the shoe sole debris from both shoe soles onto separate swabs, i.e. to collect one sample for each shoe. It will be up to the researchers then to decide whether the two samples would be further processed separately, or pooled, or one sample will be bio-banked.
- The swab samples should be either frozen (-20°C or -80°C) or placed in stabilizing buffer prior to freezing, preventing microbial proliferation and DNA/RNA degradation. If a study was performed with field workers or in a setting that would allow freezing samples swiftly post sample collection (as was the case in our study), then no specific storage buffers would be required. On the other hand, if sampling was conducted by study participants and samples were to be shipped to the research laboratory, a swab sampling system with storage/stabilizing buffer would be recommended. There are various swab systems commercially available for environmental and/or human sample collection, including such with stabilizing solution that will allow sample shipment at ambient temperature.
- Downstream processing of samples collected in such way will typically include DNA extraction, potentially including bead beating, and high-throughput sequencing based microbiota characterization. However, viability and other molecular determinations will also be feasible, if appropriate storage solutions and swab materials are being used.

Cleaning of Shoe Soles:

- 1 Cleaning of shoe soles is done with brush, water, and mild detergent for approximately 1 minute per shoe sole.
- 2 After cleaning with detergent and rinsing with water, the shoe soles are then sprayed with 70% ethanol.
- 3 Time for drying should be allowed before using the shoes as 'sampling equipment'.

Swab Preparation:

- 4 An appropriate swab type should be selected based on the study.
- 5 The swab tip should be moistened with a sterile solution (e.g., sterile water, saline solution, or appropriate buffer solution). In our study we used sterilized water with 0.05% Tween 20.

Swabbing of Shoe Soles:

- 6 There is option to collect shoe sole samples immediately after cleaning of the shoes, to establish a baseline that can be corrected for in later analyses.
- 7 Swabbing of shoe sole debris will be done at the end of the sample accumulation period, as determined by the researchers organizing the study (e.g., after a single walk).
- 8 Swabbing will be performed after dipping the swab into the buffer (see explanation above), as if trying to clean the shoe sole, touching much of the surface on and between profiles.
- 9 Swabbing of the shoe sole is done for 15-20 seconds (per shoe), i.e. there will be two swabs per swabbing session (right and left shoe).
- 9.1 It is recommended to collect the shoe sole debris from both shoe soles onto separate swabs, i.e. to collect one sample for each shoe. It will be up to the researchers then to decide whether the two samples would be further processed separately, or pooled, or one sample will be bio-banked.



Storage:

- 10 After swabbing, the tip of the swab is transferred into a pre-coded sample storage vial, or directly into DNA extraction tubes, if the research is carried out in laboratory settings.
- 11 Many swab systems have a predefined breaking point to separate the swab as such from the stick; if using basic cotton swabs the tip should be cut off with sterile scissors that have been cleaned with 70% ethanol prior to use.
- 12 Samples should be stored at -20°C or -80°C within 60 minutes post sample collection, if in laboratory settings. If collected by study participants, samples may be stored in the chosen buffer and should be shipped to the research laboratory within days post sampling.
- 12.1 The swab samples should be either frozen (-20°C or -80°C) or placed in stabilizing buffer prior to freezing, preventing microbial proliferation and DNA/RNA degradation. If a study was performed with field workers or in a setting that would allow freezing samples swiftly post sample collection (as was the case in our study), then no specific storage buffers would be required. On the other hand, if sampling was conducted by study participants and samples were to be shipped to the research laboratory, a swab sampling system with storage/stabilizing buffer would be recommended. There are various swab systems commercially available for environmental and/or human sample collection, including such with stabilizing solution that will allow sample shipment at ambient temperature.

Downstream Process:

- 13 Downstream processing of samples collected in such way will typically include DNA extraction, gene quantification, and high-throughput sequencing based microbiota characterization. Viability and other molecular determinations will also be feasible, if appropriate storage solutions and swab materials are being used.
- 13.1 In our study the largest amount of baseline samples did not contain sufficient biomass for a successful PCR amplification and sequencing.