Collect, extract, pool, and cultivate surface swab samples from built environments

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Method Article

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Abstract

This is a protocol to collect microbiome samples from built environment surfaces using swabs as the collection device, followed by extracting the biomass from swab heads to liquid buffer solutions for downstream processing. The protocol also describes the cultivation pipeline to capture a large fraction of the indoor microbiome diversity. Additionally, it provides steps to pool multiple samples together if needed.

Introduction

Reagents

Sample collection

1. COPAN Nylon Flocked Swabs (Copan Diagnostics, Murrieta, CA, USA, Cat# 502CS01)
2. Phosphate buffered saline
3. Tween 80
4. Falcon tube (Cat# 12-565-268)
5. Ice

Cultivation

1. Tryptic soy agar (TSA)
 - Tryptone (DOT Scientific, Burton, MI, USA, Cat# DST60065-500)
 - Soytone (BD Biosciences, Franklin Lakes, NJ, USA, Cat# 243620)
 - Sodium chloride (Fisher Scientific, Hampton, NH, USA, Cat# BP358-1)
 - Agar (Fisher Scientific, Hampton, NH, USA, Cat# BP1423-500)
2. Difco™ R2A agar (BD Biosciences, Franklin Lakes, NJ, USA, Cat# 218262)
3. Thermo Scientific™ Blood Agar (TSA with Sheep Blood) (Thermo Fisher Scientific, Waltham, MA, USA, Cat# R01198)
4. Alfa Aesar™ Itraconazole, 98% powder (Alfa Aesar, Haverhill, MA, USA, Cat# J66390)

Equipment

Sample collection

1. cooler

Swab extraction and sample aggregation

1. Vortex
2. Shaker
3. Centrifuge

Procedure

Sample collection

- 1) Premoisten 3 swabs (COPAN Diagnostics; Nylon Flocked Dry Swabs in Peel Pouches; Sterile; Cat# 502CS01) with 1.5 mL buffer solution (Phosphate buffered saline with 0.02% Tween 80; PBST) prefilled in a 15 mL falcon tube (Cat# 12-565-268).
- 2) Press out excess solution against the interior wall with a rotating motion.
- 3) Swab target surfaces 3 times for each sampling location with 3 swabs held together. Switch the order of the swabs as well as rotate the swabs each time during the sampling. Keep the swab pressure and swab speed relatively consistent all the time.
- 4) Place the swabs back into the falcon tube by breaking the swab at the breakpoint.
- 5) Collect 2 negative field controls, one at the beginning and one at the end of the sampling session.
- 6) Prepare 2 unopened tubes of PBST from the same lot as negative media controls.
- 7) Store the samples in a cooler filled with ice for up to 12 hours during the sample collection process.

Swab extraction and sample aggregation

Figure 1

- BSC is the abbreviation of biosafety cabinet

Cultivation

Figure 2

Troubleshooting

Time Taken

Anticipated Results

References

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Figures

Swab extraction and sample pooling

Figure 1

Swab extraction and sample aggregation

Cultivation pipeline

Figure 2

Cultivation pipeline to capture a large fraction of the indoor microbiome diversity.