

## LARGE AREA SURFACE SAMPLING METHOD USING THE INNOVAPREP® CONCENTRATING PIPETTE™

### PROBLEM

Traditional surface sampling methods provide notoriously poor results. Common surface sampling swabs limit users to very small surface areas and collection efficiency is often poor. Most swab kits are made for culture-based analysis requiring incubation of several hours or days and many pathogens of interest can be viable, though not culturable. Recovery of a surface sample from a swab for rapid molecular methods requires additional fluid for rinsing that often dilutes the target beyond the limit of detection of the assay.

### INTRODUCTION

NASA's Jet Propulsion Laboratory, California Institute of Technology, Biotechnology and Planetary Protection Group, Pasadena, CA, USA developed the following large area surface sampling method, for sampling bacteria and fungi on the International Space Station's surfaces, that uses the InnovaPrep Concentrating Pipette to concentrate the final sample into sub-milliliter volumes (see referenced publications below).

“culture-based analysis limits understanding of the diversity of microbes that grow and thrive on surfaces because only a small fraction of organisms in a given environment can be cultured under standard laboratory conditions. Molecular methods, such as quantitative polymerase chain reaction (qPCR) and targeted amplicon sequencing, which can identify and quantify both culturable and unculturable organisms provide a more thorough assessment of what is actually present and in what amounts.”

The method uses standard sterile nine-by-nine inch wetted polyester wipes to sample large area surfaces or objects up to one meter squared. The wipes are placed in a bottle with 200 mL of a buffer fluid, shaken or vortexed, and then the fluid is concentrated (in minutes) from 200 mL to about 250 µL using the Concentrating Pipette. The concentration factor for this operation is approximately 200 mL / 250 µL, or 800X, multiplied by the efficiency (usually 50-90%).

The InnovaPrep Concentrating Pipette Select (CP Select™) is an automated, bio-concentration device for modern microbiology. The CP Select uses dead-end filtration with single-use Concentrating Pipette tips (CPTs) to capture microorganisms from fluid sample matrices. A patented process called Wet Foam Elution is then used to recover the microorganisms into sample volumes that closely match the input volumes of modern molecular methods such as qPCR in minutes. The system's ease of use and ability to deliver exceptionally high concentration factors for sample recovery makes it an ideal approach for the concentration of microbial contamination on surface samples using modern molecular methods. The method is appropriate for monitoring bacteria, molds, spores, and viruses, including SARS-CoV-2, incidentally. The method is suitable for a variety of applications including food safety, outbreak investigations, disease monitoring including hospital-acquired infections, and animal health.

## **SAFETY**

Due to the potential presence of infectious pathogens in surface samples, users should work with their organization's occupational safety team to ensure that methods and safety measures are appropriate and approved. Unless working with samples known to be non-infectious, InnovaPrep recommends that CP Select operations be performed in a biosafety cabinet. For SARS-CoV-2, one source of information is published by the U.S. CDC: Interim Laboratory Biosafety Guidelines for Handling and Processing Specimens Associated with Coronavirus Disease 2019 (COVID-19). Guidelines are also available for other BSL 2 & BSL 3 pathogens from the CDC.

## **METHOD**

### STEP 1- Prepare Sample Wipes

- 9"x 9" ITW Texwipe (Mahwah, NJ)(or other sterile dry polyester-type material) folded two times.
- Soak each wipe in 15 mL of sterile molecular grade water for 30 min.
- Transfer to a sterile ziplock bag.

### STEP 2- Sampling Method

- Using gloved hands, remove an ITW Texwipe from a ziplock bag.
- Use the wipe to sample surfaces up to one square meter, including knobs, handles, crevices, or items (plastics, glass, metals, smooth wood, etc).
- After sample collection, transfer wipe to a 500 mL bottle containing 200 mL of sterile phosphate-buffered saline (PBS; pH 7.4).

### STEP 3- Sample Processing

- Shake or vortex the bottle with the wipe for 2 minutes.
- Concentrate the liquid sample using the InnovaPrep CP Select following manufacturer menu selection recommendations.
  - Pipette filter tips for the CP Select come in a range of six filter types and are chosen according to pore size for the organisms of interest. See Tip Selection Guide.
  - The final eluate can be analyzed using classical culture or rapid molecular methods of your choice.
  - Multiple samples can be pooled and concentrated at once without increasing the final sample volume or reducing concentration factors.

\*Store pre-concentrated or post-concentrated liquid samples at 4 °C if needed.

Note: The addition of 0.05% Tween 20 to the "soak water" and/or the "sample collection buffer" will provide improved recovery of SARS-CoV-2 and other target organisms.

## **REFERENCES**

2020 - Description of Chloramphenicol Resistant *Kineococcus Rubinsiae* sp. nov. Isolated from a Spacecraft Assembly Facility

2020 - Crewmember microbiome may influence microbial composition of ISS habitable surfaces

2020 - Characterization of Spacesuit Associated Microbial Communities and their Implications for NASA Missions

2019 - Characterization of the Total and Viable Bacterial and Fungal Communities Associated with the International Space Station Surfaces

2017 - Human presence impacts fungal diversity of inflated lunar/Mars analog habitat

2016 - Characterization of *Aspergillus Fumigatus* Isolates from Air and Surfaces of the International Space

2016 - Microbial succession in an inflated lunar/Mars analog habitat during a 30-day human occupation