



SAMPLING STRATEGY: MICROORGANISMS DETECTION BY CULTURE

Coriolis micro AIR SAMPLER

/ PURPOSE

Assessment of total cultivable microorganisms (Bacteria, fungi) from indoor areas (hospital, buildings, offices...).

/ MATERIALS

- 1. Sampler: Coriolis μ (including at least one air intake, one cane)
- 2. Sampling Cones: Bertin sterile cones & caps (Ref. 05237-1-101)
- 3. Sampling media: Bertin collection liquid doses – 15 mL (Ref. 05237-1-202)
- 4. Rubbing alcohol 70%.
- 5. Refrigerant packs, if necessary for keeping samples cool during shipment.

NOTE: Prepare the number of sterile air intake, sterile cones and collection liquid doses according with your sampling schedule.

/ SAMPLING STRATEGY

1. Select at least three sites, one each to represent complaint area, a non complaint area (otherwise as similar as possible to complaint area), and outdoors.

2. Set the Coriolis μ in a trolley or in a tripod at 1 m or 1.20 m from the floor.

3. In turn at each site, sample simultaneously for fungi and bacteria. Typical sampling time is 4 minutes (a volume of 1 m³ sampled). Before moving to the next site, repeat twice to obtain triplicate, using a new sterile cone and collection liquid dose at each sampling.

NOTE: Start the sampling in lightly contaminated area and move on to heavily contaminated area. The same sampling time could be used in heavily or lightly contaminated areas.

/ SAMPLING

1. Set the Coriolis μ with a sterile cane and sterile air intake.

2. Load a collection liquid dose into a sterile cone (previously properly labeled with the sample identification) and screw on the air intake. Keep the cone cap in the sterile bag.

NOTE: Take care to prevent contamination of collection liquid during loading and unloading. Do not touch inside the cone or the air intake.

3. Sample at 250 L/min – 4 minutes (=1 m³).

4. At the end of sampling, unscrew the cone from the air intake and screw firmly the cap on the cone and store the sample in a cool box for transportation and/or until analysis

5. Repeat twice to obtain triplicate using new sterile collection liquid doses into new sterile cones.

6. Before to move to new area sampling, remove the air intake. Move on to the new area of sampling and start new triplicate sampling with a new sterile air intake, sterile collection liquid doses and sterile cones.

NOTE: It is also possible to carefully and thoroughly wipe the air intake with rubbing alcohol. Allow to dry.

/ ANALYSIS

Keep samples in a cool box and ship as quickly as possible to a laboratory for growth culture analysis (enumeration and identification), preferably within 24 hours and provide with the samples sample identification and all pertinent information (e.g. air flow rate, time and location).

NOTE: Keep samples cool but protect them from freezing.



SAMPLING STRATEGY: MICROORGANISMS DETECTION BY PCR

Coriolis micro AIR SAMPLER

/ ANALYSIS

Growth culture analysis: Growth culture analysis is used to quantify and characterize airborne cultural bacteria and fungi. A portion of the liquid sample is transferred to a culture medium on which the microorganisms are grown into colonies for counting. A broad spectrum of media is generally used.

Culture media recommended:

For total cultivable bacteria assessment: Trypticase soy agar (TSA)

For total cultivable fungi assessment: Malt extract agar (MEA)

The plates are prepared according to sampler manufacturer's recommendations.

NOTE: Other media may be used, if appropriate, e.g., dichloran glycerol agar (DG18) for xerophilic molds, R2A agar for heterotrophic bacteria, and rose bengal agar for slow-growing fungi such as Stachybotrys.

1. Before analysis, accurately measure the final volume of collection liquid in the cone in an aseptic area and record it.

NOTE: The liquid sample could be aseptically transferred to a sterilized container for analyses.

2. Vortex 5-10s second the liquid sample in the Coriolis cone (or in the transferred container) and remove e.g. 0.5mL with a sterile pipette. Spread this volume onto an appropriate agar plate. Repeat once or twice to obtain duplicate or triplicate by each culture medium.

NOTE: The sample can be diluted before spreading in agar media if the samples come from heavily contaminated areas.

3. Incubate the agar plates at the appropriate temperature.

4. After the incubation step, count the number of colony by plate

5. Express the result in CFU/m³ according the following formula:

$$\text{Total cultivable bacteria (CFU/m}^3\text{)} = [(X / V_s) \cdot V_t] / V_a$$

$$\text{Total cultivable fungi (CFU/m}^3\text{)} = [(X / V_s) \cdot V_t] / V_a$$

X = Number of colonies counted in a agar plate

V_s = Volume of sample spread onto the agar plate

V_t = Total volume of sample measured after the sampling

V_a = Volume of air collected.

V_a can be calculated using the formula: V_a [m³] = Air flow rate [L/min] x Time sampling [min] / 1000L.

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