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Sample Concentration by Tangential Flow Filtration

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Protocol status: Working

Sample Preservation protocol from MaCuMBA project.

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Materials

STEP MATERIALS

 0.1 M NaOH

 0.1 M NaOH

Protocol materials

 0.1 M NaOH

Before start

Equipment



- Vivaflow Cartridge 0.2 μm (PES) VF20P7
- Masterflex Pump 6-600 rpm (ref Bioblock F39671)



- Rotor 3 "cylinders" (ref F39110)
- 1 high throughput head (ref F40103) (can be replaced with quick load head)
- Replace tube provided by a stronger tube with two connectors (see picture)
- Bottle 6 L
- Bottle 1 L
- Conical tube 50 mL
- Masterflex Tygon tubing size 16
- Plastique pipettes (1 mL)
- Clamps with a screw (to control retentate1 speed)
- Clamps

Solutions

- Rinsing solution for regenerated cellulose membranes: 250 ml of 0.1 M NaOH
- Storage solution: Deionised water with 10 % ethanol.
- Deionized water
- Filtered seawater

Setup

- 1 Get Vivaflow cartridge out of storage and as shown in Figure 1 below (Courtesy of C. Brussaard, NIOZ).

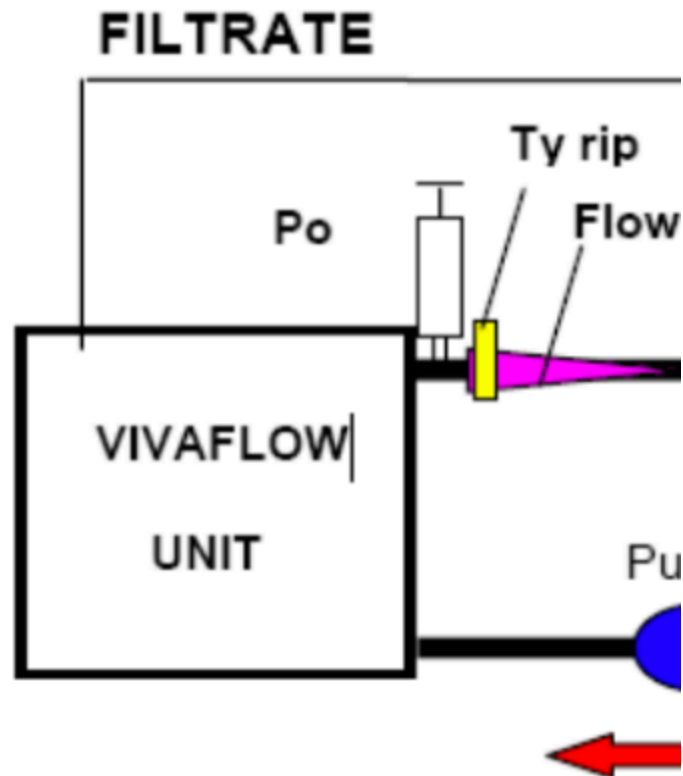


FIG 1. FLUSHING SET UP

Washing

- 2 Remove the clamps and set the pump to maximum speed. Manometer should be at about 2.5 bars (with a new cassette sometimes the manometer get stuck, if the value is too low)

there is a leak in the system). Rinse the cartridge with about 250 mL of distilled water (longer if the cartridge has been stored in ethanol).

🧪 250 μ L Distilled (MilliQ) Water

Sample Concentration

- 3 Replace MilliQ water by the sample in the 6L bottle. Rinse cartridge with about 250 mL of the sample. Put the retention line into the sample bottle (Figure 2 below)

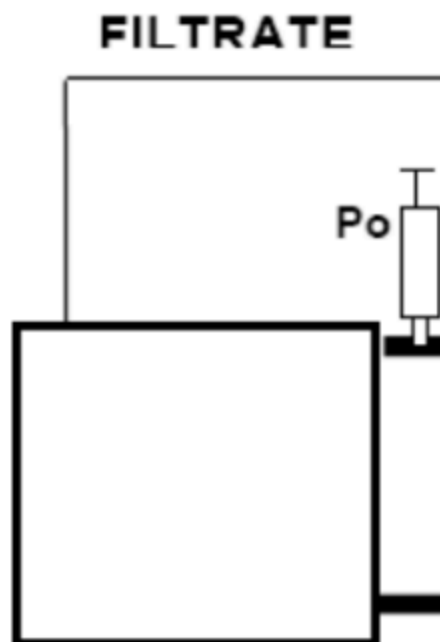


FIG 2 CONCENTRATING

Put a screwing clamp on the retention line to increase filtrate flow so that Manometer gets up to 2.5 bars. Now concentrate sample until about 250 mL, which takes about one hour for the 6L sample bottle. Afterward, transfer to a smaller container (first to a 250 mL bottle then finally to a 50 mL tube). Continue to concentrate being very careful and lowering the pump speed. When the final volume is about 10 mL, clamp filtrate tube and



recirculate slowly (no change of volume should take place). Last, leave the filtrate tube clamped, get the feed line out of the sample in order to get back the total volume of concentrated sample

Store Sample

- 4 Store concentrated sample.

Cleaning the Equipment

- 5 Go back to the configuration as shown in Figure 1 above. In order to clean first rinse 1 min with filtered seawater, followed by 1 min with distilled water. Now rinse with 50 mL NaOH 0.1 M. Put all three tubes (feed, retention, filtrate) in a bottle containing NaOH 0.1 M. Now recirculate for 20 min (to get rid of everything on the cartridge filter). Finally, rinse on more time with 250 mL of distilled water (Fig. 1). Stop the pump and clamp all three tubes. Store at 4°C (For a storage longer than one day, store with 10% ethanol).

 0.1 M NaOH