

INTRODUCTION

The InnovaPrep Concentrating Pipette (CP) is an automated, rapid micro-particle concentrator developed for general microbiology use. The system performs rapid "mechanical enrichment" as a front-end to rapid microbial detection, replacing older, slower, labor-intensive methods such as centrifugation or culture-based enrichment steps. The one-pass method provides sample volume reduction and removal of soluble matrix-associated inhibitors for modern analysis methods such as PCR.

The concentration process uses tangentially-loaded, dead-end filtration to capture particles on the surface of a porous membrane filter within the InnovaPrep CP's single-use Concentrating Pipette Tip. After the sample has been processed and the particles have been trapped, InnovaPrep's patented Wet Foam Elution™ process is employed to sweep the particles off of the membrane surface into a very small liquid volume providing exceptional concentration factors.

This technology has proven to be indispensable for its ability to deliver concentrated samples of waterborne pathogens to rapid detection methods such as real-time polymerase chain reaction systems in a fraction of the time compared to other methods.

Examples of environmental water samples include but are not limited to: swimming pool samples for *Cryptosporidium* and/or fecal coliforms; recreational waters (including lakes, ponds, rivers, streams, and ocean beaches) where livestock or sewage run-off result in *E. coli* contamination; cooling tower waters where the growth of *Legionella* is of concern; irrigation waters where animal and human feces can cause *Salmonella* and *E. coli* contamination in crops; marine research; industry run-off monitoring; source water monitoring; effluent water monitoring, and many other applications.

COMMON WATERBORNE PATHOGENS

- · E. coli
- · Salmonella typhi (typhoid fever)
- · Legionella pneumophila (legionnaires disease)
- · Vibrio cholerae (cholera)
- $\cdot \textit{Shigella} \; (dysentery)$
- · Giardia lamblia (giardiasis)
- · Cryptosporidium (diarrheal illness cryptosporidiosis)

PROTOCOL OVERVIEW

A simple protocol for concentrating environmental samples using the InnovaPrep CP is provided below.

Many users have used this protocol with excellent success.

MATERIALS REQUIRED

- · InnovaPrep Concentrating Pipette
- · CP Elution Fluid (HC08000 or HC08001)
- · Concentrating Pipette Tip (CC08001, CC08018, CC08022, CC08020 or CC08003)

STEP 1 OPTIONAL PREFILTRATION

InnovaPrep has demonstrated the ability to concentrate bacteria from environmental waters with various levels of organic and non-organic material loading. The device is robust; however, membrane blinding can occur while processing samples with debris or a high particle load. It may be necessary to pre-filter environmental samples.

This can be accomplished with the use of optional Mesh Pipette Filter Sleeves (HC08016). The sleeves prevent large particulates from blinding the filter within the Concentrating Pipette tip while processing liquids. Half of the sleeve is made of a polyethylene mesh filter with pore sized holes 330 microns in diameter, and the other half is solid 3.0 mil (.075 mm) polyethylene. The pipette tip slides into the sterile sleeve and is inserted into the liquid sample fluid to be tested. The liquid will pass through the filter, while screening out the larger debris.

Mesh Pipette Filter Sleeve

STEP 2 SELECT ELUTION FLUID FORMULATION

Select the elution fluid formulation that is suitable for your desired analysis method.

ITEM #	FORMULATION	NOTES
HC08000	0.075% Tween 20 PBS	analysis by classical culture
HC08001	0.075% Tween 20 Tris	analysis by molecular methods

STEP 3 CONCENTRATION

Note that universal precautions are required for working with pathogenic samples.

- 1. Set up Concentrating Pipette (CP) as instructed in Section 4 of the CP Select User Guide.
- 2. Insert a Concentrating Pipette Tip (CPT) and select a menu protocol as instructed in Section 5.2 of the CP Select User Guide for the chosen CPT type.
- 3. Lower the CPT into the sample.
- 4. Press "Start Run" on the user screen.
- 5. When the entire sample has been processed the CP will stop.
- 6. Place a clean final sample container under the CPT. The menu screen will prompt you to press "Elute".
- 7. Press "Elute". The sample will dispense from the pipette tip into the sample container.
- 8. The sample is ready for subsequent sample preparation and analysis steps.

THE CONCENTRATING PIPETTE SELECT TIP SELECTION GUIDE

Single-use CPT's are available in the following configurations, sold in packages of 10 and 60.



PART NUMBER	FILTER TIP MEDIA TYPE	PORE SIZE	MEMBRANE SURFACE AREA	INPUT SAMPLE VOLUME (VARIES BY MATRIX)	FINAL CONCENTRATED SAMPLE VOLUME	FLOW RATE (VARIES BY MATRIX)		
CC08000	Flat membrane Polycarbonate Track Etch	0.4 μm	8.5cm ²	Up to 1 L	200-1000 μL	Up to 100mL/min.		
Recommended for concentrating bacteria, whole cells, spores, pollen, and parasites from food and beverage matrices per samples containing proteins.								
CC08001	Flat membrane Polyethersulfone	0.1μm	8.5cm ²	Up to 1 L	200-1000 μL	Up to 100mL/min.		
Recommended for concentrating bacteria, whole cells, spores, pollen, and parasites from environmental samples.								
CC08018	Hollow Fiber Polysulfone (PVP)	0.45 μm	98cm²	Up to 5 L	150-1000 μL	Up to 200mL/min.		
Recommended for concentrating bacteria, whole cells, spores, pollen, and parsites from high fouling matrices.								
CC08022	Hollow Fiber Polysulfone (PVP)	0.2μm	98cm²	Up to 5 L	150-1000 μL	Up to 200mL/min.		
Recommended for concentrating bacteria, whole cells, spores, pollen, and parasites from all sample types.								
CC08020	Hollow Fiber Polysulfone (PVP)	0.05μm	98cm²	Up to 3 L	150-1000 μL	Up to 90mL/min.		
CC08003	Hollow Fiber Polysulfone (PVP)	Ultra- filtration	98cm²	Up to 500 mL	150-1000 μL	25mL/min.		
Recommended for virus, DNA, and prion concentration. The addition of Tween 20 to the sample is								

Recommended for virus, DNA, and prion concentration. The addition of Tween 20 to the sample is recommended to aid recovery efficiency (add 10% solution of Tween 20 in a 1/100 ratio to the sample).