

DNA Containment for Two-Step PCR

Alluvia™ System

A consumable / instrument system to dilute and transfer first step PCR products from a single PCR tube into multiple second step PCR wells for subsequent qPCR and/or gel electrophoresis while providing continuous sample containment.

- ❑ **Eliminates PCR contamination risk;**
- ❑ **Integrates with existing downstream qPCR and gel electrophoresis workflows;**
- ❑ **Integrates with standard lab equipment, consumables, and biochemistry.**



Advantages and Benefits of the Alluvia System:

Enables 2-Step Multiplex Nested PCR, which:

- ❑ **Improves sensitivity**, particularly when using precious samples, by multiplexing during the first step (less total sample needed);
- ❑ **Improves specificity** by using a second pair of nested primers.








Provides Consumable-Based PCR Product Containment, when:

- ❑ Biochemical methods are not sufficient (i.e. uracil/UNG/UDG is useful for only one round of 2-step PCR, and ineffective for methylation detection when utilizing bisulfite treatment);
- ❑ Facility and workflow-based containment strategies are cumbersome, costly, and are more user dependent and failure prone.

Applications Demonstrated:

- ❑ **DNA Methylation Analysis;**
- ❑ **Pathogen DNA Detection.**

Alluvia 2-Step PCR Integration

Sample Input	Second Step PCR Component Input	Second Step PCR Formats	Sample Transfer, Dilution & Aliquoting	Detection / Analysis	2-Step Nested PCR Example Assays
0.1ml or 0.2ml PCR tubes 	Pipette  Robotics	1x8 PLM  (4) 1x24 PLM  <i>(Prefilled with PCR reagents and primers)</i>	Alluvia Instrument 	qPCR  Gel Electrophoresis GLM 	DNA Methylation Analysis Pathogen Detection

Alluvia Consumables

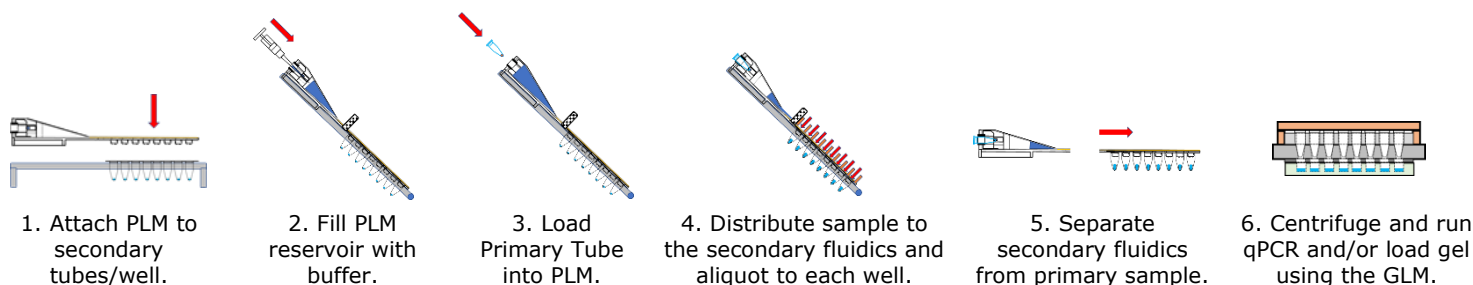
Plate Loading Manifolds (PLM's)

A set of consumables that completely contain the transfer and dilution of primary PCR product tubes into a variety of standard tube strip or well plate formats containing secondary PCR reactants.

Gel Loading Manifolds (GLM's)

Consumable adaptors and inserts that completely contain the transfer of PCR products from 8 tube strips into existing off-the-shelf disposable gel electrophoresis cartridges without the need of pipetting.

Workflow Summary:



Product Specifications

Alluvia Instrument

- User Interface: Integrated touchscreen
- Configurability: 8 tube strip and 96 well inserts (Additional in development)
- Total Process Time: 2-5 minutes
- Dimensions: 25x35x35cm (WxDxH)
- Power requirements: 110V/220V, 50/60 Hz
- Safety and emission certification: CE, ETL

Plate Loading Modules (PLM)

- Primary PCR Samples: 1 to 4
- Secondary PCR Conditions per Sample: 8, 24, 96...
- Primary PCR Dilution: 10x-250x
- Secondary PCR volumes: 10µl to 15µl

Gel Loading Module (GLM)

- DNA Containment: >99.9%
- DNA Transfer Reliability: >99%
- DNA Volume Transfer: >50% Efficiency

Two-Step PCR with Alluvia™

Contamination Containment - Improved Specificity & Sensitivity