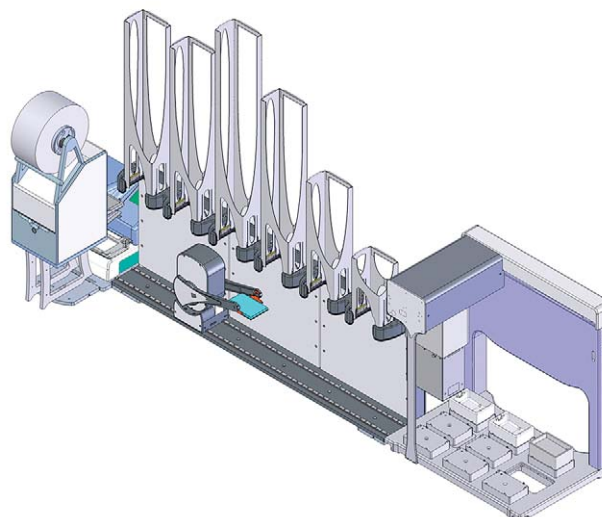
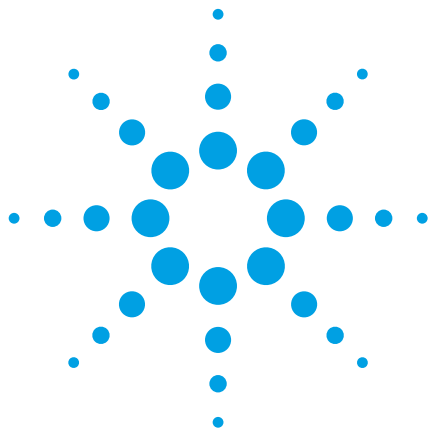


Agilent PCR Preparation Workstation

Application Bulletin



Agilent PCR Preparation Workstation layout consisting of an Agilent Bravo Automated Liquid Handling Platform (right), Agilent BenchCel Microplate Handling Workstation Series-R (center), Agilent PlateLoc Thermal Microplate Sealer (left), and a Thermo Multidrop (not shown)

Summary

- A flexible workstation providing high-throughput PCR preparation
- Up to 150 microplates can be processed in one run (with tip changing)
- Plate processing time is approximately 5 hours for a 50 microplate run, depending on protocol complexity

Introduction

The high demand for genomics applications and the considerable improvement in PCR thermocycler speed have created bottlenecks upstream and downstream of the PCR process. The PCR Preparation Workstation from Agilent Automation Solutions will speed up plate preparations and provide valuable walk-away time. The system is flexible, enabling you to modify protocols and procedures to accommodate your specialized needs. This application bulletin outlines a protocol for preparing PCR microplates using the Agilent PCR Preparation Workstation.

System Description

The workstation contains an Agilent Bravo Platform, Agilent BenchCel Workstation, and an Agilent PlateLoc Sealer. The Bravo gripper transports PCR microplates and tipboxes around the platform. Two chilled, multi-channel reservoirs keep the PCR master mixes and the exon solutions stable. The tip-wash station with sonication enables consumable conservation where possible, and the tip-waste station facilitates necessary tip changes. The Bravo pipette head can pick up single rows or columns of tips, providing great flexibility in reagent addition layouts. The BenchCel Workstation, with six stackers, stores microplates and tipboxes,

and transports microplates to the Thermo Multidrop and the PlateLoc Sealer for reagent addition and plate sealing, respectively. All tasks for each device of the system are scheduled by the Agilent VWorks Automation Control software for optimal throughput.

Materials

Component List

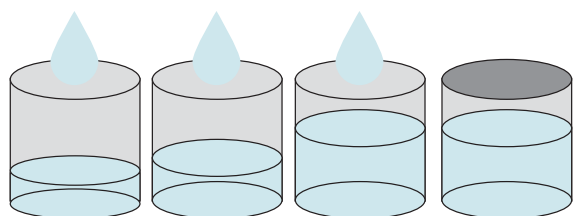
- BenchCel Workstation Series-R with 6 stackers
- Bravo Platform with gripper, 384ST disposable tip head, 2 chilled Reservoirs, sonic tip wash, and tip waste
- PlateLoc Sealer
- Thermo Multidrop

Labware List

- Microplate A: Eppendorf twin.tec 384 PCR
- Tipbox A: Agilent Tips 384 ST 70 μ L

Reagent List

- Reservoir A: Master-mix solution
- Reservoir B: Exon Solution



Diluent is added to DNA sample

Master-mix solution is

Exon solution is added

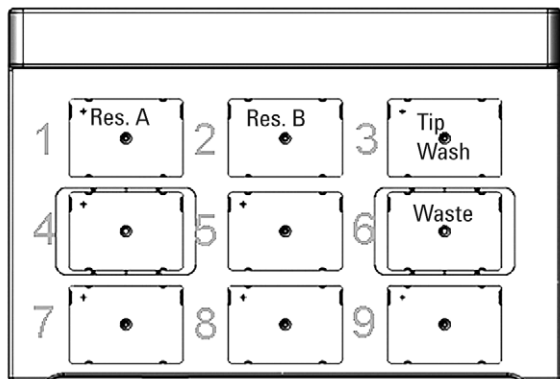
Plate is sealed and stored for further use

Overview of the PCR Preparation procedure: Diluent, Mastermix and Exon solution are added to PCR plates, the plates are sealed and stored for further use.

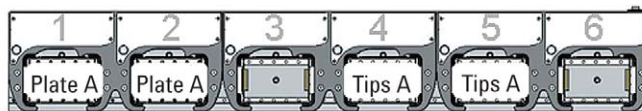


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Instrument Layout



The Agilent Bravo Platform (top view) has two chilled reservoirs (locations 1, 2) to keep solutions stable, a tip-wash station equipped with sonication (location 3), and a waste station suitable for all solid waste at location 6.



The Agilent BenchCel stacker layout (top view). Stackers 1 and 2 contain microplates to be processed (each rack can store up to 65 microplates). Stackers 4 and 5 contain tipboxes (each rack can store up to 30 tipboxes). Stackers 3 and 6 are used for moving microplates.

Protocol Workflow

1. Move tipbox A from BenchCel stacker 4 to Bravo location 7.
2. Move tipbox A from location 7 to 8.
3. Move microplate A from BenchCel stacker 1 to Multidrop device.
4. Dispense 25 μ L diluent into microplate A.
5. Move microplate A from Multidrop device to Bravo location 7.
6. Press on tips at location 8.
7. Aspirate 25 μ L master-mix from reservoir A and dispense into microplate A.
8. Wash tips at location 3.
9. Aspirate 25 μ L exon from reservoir B and dispense into microplate A.
10. Release tips at waste at Bravo location 6.
11. Move microplate A from Bravo location 7 to PlateLoc.
12. Seal microplate A.
13. Move microplate A from PlateLoc to BenchCel stacker 3.
14. Move tipbox from location 8 to 7.
15. Move tipbox A from Bravo location 7 to BenchCel stacker 6.

Conclusions

The Agilent PCR Preparation Workstation provides the throughput, flexibility, and walk-away time necessary to keep up with the demands of genomic applications. The Agilent BenchCel Workstation is well equipped to handle the wide variety of PCR microplates, and its range of motion allows the integration of up to three instruments. The speed and precision of the Agilent Bravo Automated Liquid Handling Platform enables the workstation to meet the needs of a wide range of PCR preparation demands, and its configurable tip usage enables many reagent layout options. The estimated throughput of the outlined protocol is 5 h for 50 microplates processed (depending on exact liquid handling procedures). Agilent VWorks software uses a simple drag-and-drop method to create the event-driven protocol for the entire process and includes features such as error checking, error recovery, event reporting, and user-access management.

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