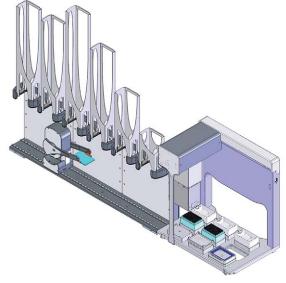


Agilent PCR Clean-up Workstation Application Bulletin



Agilent PCR Cleanup Workstation consisting of an Agilent BenchCel Microplate Handling Workstation (left) and an Agilent Bravo Automated Liquid Handling Platform (right).

Summary

- A flexible workstation providing high-throughput PCR clean up
- Up to 15 PCR microplates can be processed without user intervention
- Processing time is approximately 20 min per microplate (depending on the exact procedure)

Introduction

The high demand for genomics applications and the considerable improvement in PCR thermocycler speed have created bottlenecks upstream and downstream of PCR processes. To address this demand, the PCR CleanUp Workstation from Agilent Automation Solutions is custom tailored around the Promega Wizard SV96 kit, one of the most successful PCR kits on the market.

System Description

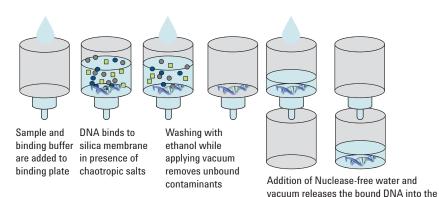
collection plate

The combination of the Agilent Bravo Automated Liquid Handling Platform and the Agilent BenchCel Microplate Handling Workstation provides a flexible, efficient and compact approach for automated PCR clean up. The BenchCel robot moves the labware to and from the Bravo deck. The Bravo gripper assembles and disassembles the Vacuum Filtration Station and transports microplates to and from the station.

Reservoirs and two sets of tipboxes are placed manually on the Bravo deck at the beginning of a run. A waste station on the Bravo deck can accommodate both tipboxes and microplates.

The Agilent VWorks Automation Control software uses a simple drag-and-drop method to create the event-driven protocol for the entire process. The VWorks software includes error checking, error recovery, event reporting, and user-access management.

This application bulletin outlines a protocol for the Promega Wizzard SV96 kit using the Agilent PCR Cleanup Workstation. This workstation setup can process 15 kits in 96-well plate format running without manual intervention. The estimated throughput time is 20 minutes per microplate, depending on the exact procedures.



Overview of the PCR cleanup procedure using the Promega Wizzard SV96 kit



Materials

Component List

- Agilent BenchCel Workstation (R-series with 6 stackers)
- Agilent Bravo Platform with gripper, 96LT disposable-tip head, 3 x Reservoir, Tip Waste, Vacuum Filtration Station
- Agilent VWorks Automaton Control software

Labware List

- · Microplate A: PCR sample plate
- Microplate B: Promega Wizard SV 96 binding plates
- Microplate C: Promega Wizard SV 96 elution plates
- Tipbox A: Agilent Tips 96 LT 200 μL

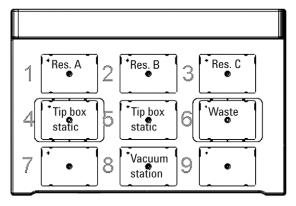
Reagent List

- Reservoir A: Binding solution
- Reservoir B: Ethanol (85%)
- Reservoir C: Nuclease-free water

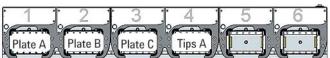
Protocol Workflow

- Move tipbox A from BenchCel stacker 4 to Bravo location 7.
- 2. Move tipbox A from location 7 to 9.
- 3. Move microplate B from BenchCel stacker 2 to Bravo location 7.
- 4. Move microplate B from location 7 to 8.
- 5. Move microplate A from BenchCel stacker 1 to Bravo location 7.
- 6. Press on fresh tips at location 9.
- 7. Aspirate 100 µL binding solution from reservoir A and dispense into microplate A.
- 8. Perform mixing with tips.
- 9. Transfer 200 μL from microplate A to microplate B.
- 10. Remove used tips at location 9.
- 11. Move tipbox A from location 9 to waste at location 6.
- 12. Move microplate A from location 7 to waste at location 6.
- 13. Apply vacuum at location 8 for 30 s.
- 14. Press on fresh tips at location 4.
- 15. Aspirate 200 µL from reservoir B and dispense into microplate B. Wait 1 min.

Instrument Layout



The Agilent Bravo deck (top view) with three reagent reservoirs (locations 1 to 3), a waste station (location 6), and a Vacuum Filtration Station (location 8). Two tipboxes are manually placed at locations 4 and 5.



The BenchCel 6 stacker top view with three different microplate types (stackers 1 to 3) and up to 30 tipboxes (stacker 4).

- 16. Apply vacuum at location 8 for 30 s.
- Loop: Repeat steps 15 and 16 a total of 3 times.
- 17. Apply vacuum at location 8 for 4 min. Wait 1 min.
- 18. Remove used tips at location 4.
- 19. Move microplate B and collar from location 8 to 9.
- 20. Move microplate C from BenchCel stacker 3 to Bravo location 7.
- 21. Move microplate C from location 7 to 8.
- 22. Move microplate B and collar from location 9 to 8.
- 23. Press on fresh tips at location 5.
- 24. Aspirate 100 μL water from reservoir C and dispense into microplate B.
- 25. Wait 1 min. Apply vacuum for 30 s.
- 26. Remove used tips at location 5.
- 27. Move microplate B from location 8 to waste at location 6.
- 28. Move collar from location 8 to 9.
- 29. Move microplate C from location 8 to 7.
- 30. Move collar from location 9 to 8.
- 31. Move microplate C from location 7 to BenchCel stacker 6.

Conclusions

The Agilent PCR Cleanup Workstation provides the throughput, flexibility, and walk-away time necessary to meet the demands of genomic applications. The BenchCel Microplate Handling Workstation handles the wide variety of PCR microplates on the market and is capable of storing and delivering a number of different labware types (microplates, tipboxes) simultaneously. The speed and precision of the BenchCel Workstation and the Bravo Automated Liquid Handling Platform can meet the challenge of a wide range of PCR cleanup demands. The Bravo Platform tip usage is configurable to support many reagent layout options.

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