AssayMAP Protein Sample Prep Workbench

In-Solution Digestion: Single Plate v1.2 Quick Start Guide

This guide is intended for users who have been trained in the proper use of the AssayMAP Bravo Platform and understand the safety guidelines in the *Bravo Platform Safety and Installation Guide*. The procedures in this guide require the Protein Sample Prep Workbench and VWorks Automation Control software. See the user guide to verify the required software versions.

Step 1. Design your run

Use the **Reagent Volume Calculator** for In-Solution Digestion: Single Plate v1.2 to:

- Determine reagent volume preparation requirements.
- Ensure the labware selections are consistent with volume requirements.

For in-depth assay development guidelines, see the *In-Solution Digestion: Single Plate v1.2 User Guide* in the Literature Library of the Protein Sample Prep Workbench.

Step 2. Prepare sample and reagent plates

To minimize evaporation, fill the labware immediately before run time or keep them covered until you run the protocol.

CAUTION

A small reagent volume excess is required in all labware types to ensure proper volume transfer.

Use the Reagent Volume Calculator to automatically include excess volume, or look up the recommended value for each allowable labware type in the *AssayMAP Labware Reference Guide*, which is available in the Literature Library page of the workbench.

Step 3. Prepare the system

To prepare the system:

1. Check the levels of the wash station source and waste carboys, and fill or empty as required.
2. If you have not already done so, turn on the AssayMAP Bravo Platform and accessories, and start the Protein Sample Prep Workbench.
3. Open the **Utility Library**, and then open the **System Startup/Shutdown** utility  .
4. Click **Run Startup** to prepare the system for the run.

WARNING

The Bravo head and tie bar will move during the Bravo Startup protocol. To prevent injury, keep clear of the device while it is in motion.

5 During the Startup protocol, verify that all the wash station chimneys have liquid flowing through them. If liquid is not flowing through the chimneys, see the *96 Channel Wash Station Maintenance Guide* for troubleshooting guidelines.

Step 4. Prepare the tip seating station

If pipette tips are required for the run, fill the 96AM Cartridge & Tip Seating Station with the appropriate number and configuration of pipette tips to match the samples to be processed. To prepare the pipette tips, run the **Pipette Tip Transfer** utility.

Step 5. Run the application

To run the In-Solution Digestion: Single Plate application:

1 Open the **In-Solution Digestion: Single Plate v1.2** app.

2 In the **A: Select Method** area, click ... and select a method to use as written or to modify as required. Click **Load Values** to display all the settings and the Deck Layout associated with the selected method.

Note: By default, methods are stored in the C:\VWorks Workspace\Methods\AM In Solution Digestion Single Plate v1.0 folder.

To run a saved method exactly as written, go to step 7. Otherwise, proceed to step 3 to make changes.

3 Under **B. Input Sample Settings**, verify the settings, or make any required changes to meet the requirements of your run. For more details, see "Application overview" on page 3.

4 Under **C. Input Addition Step Settings**, verify the settings, or make any required changes to meet the requirements of your run. For more details, see "Application overview" on page 3.

5 Under **D. Update Deck Layout**, click **Update Deck Layout** to refresh the deck so that it reflects any changes that you made to the method.

6 To save your edited method, enter the name in the **Save Method** area. Select **Overwrite if Name Exists,** if applicable, and then click **Save**.

WARNING **The probes of the Bravo 96AM Head are sharp and can scratch you if they brush across your hand. A probe scratch can expose you to any contaminants remaining on the probes. Be careful to avoid touching the probes.**

7 Place the items at the assigned deck locations, as shown in the **Deck Layout** of the app interface:

- Place the filled sample plate at deck location 4, and place the filled reagent plates at their respective deck locations.
- If lids are specified in the method, ensure that the lids are on the corresponding plates.
- If pipette tips are required for the run, place the prepared 96AM Cartridge & Tip Seating Station at deck location 2.

CAUTION **Incorrect labware selections can cause a hardware collision, resulting in equipment damage. Ensure that the labware selected in the method matches the physical labware present on the Bravo deck.**

8 Ensure that all labware are properly seated on the deck.

CAUTION **Improperly seated labware can cause a hardware collision, resulting in equipment damage. Ensure that all labware are properly seated within the alignment features of their respective platepads.**

9 Click **Run Protocol** to start the run.

Step 6. Clean up after each run

To clean up after the run:

1 Remove used labware from the deck.

2 Discard the leftover reagents appropriately.

3 *Optional*. To conduct stringent washing of the syringes, run the **Syringe Wash** utility  .

Step 7. Shut down at end of day

To shut down at the end of the day:

1 Open the **System Startup/Shutdown** utility  .

2 Remove everything from the deck except the 96AM Wash Station (deck location 1) and the 96AM Cartridge & Tip Seating Station (deck location 2), and then click **Run Shutdown**.

3 After the Shutdown protocol has completed, turn off the power at the AssayMAP Bravo Platform and the accessories.

4 Close the Protein Sample Prep Workbench software.

Application overview

The application includes up to four generic liquid-handling steps that successively transfer a reagent from a reagent plate at deck location 5, 6, 8, and 9, into the Sample plate at deck location 4. The following figure shows the interface, and the following tables provide the settings and step descriptions.

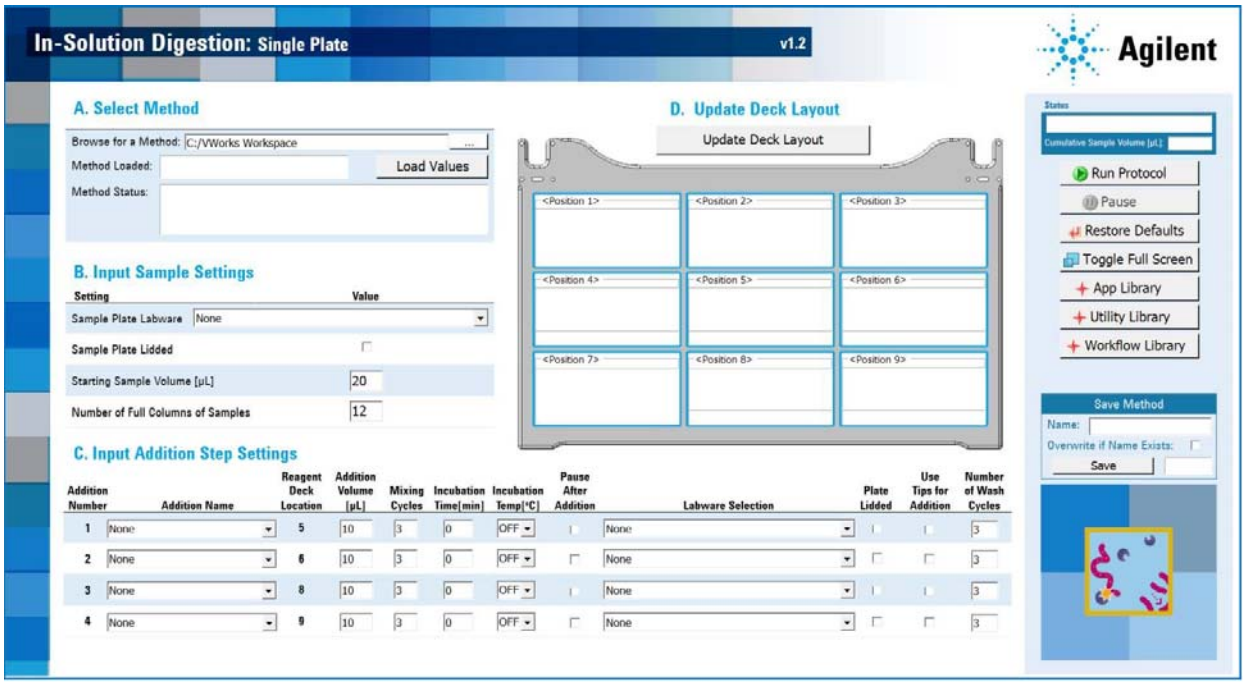

Table **Sample settings and step descriptions**

Sample setting	Description	Default value (range)
Sample Plate Labware	Specifies the type of labware that will be placed at deck location 4, which contains the sample to be digested.	–

Sample setting	Description	Default value (range)
Sample Plate Lidded	Specifies whether the Sample Plate has a lid. If selected, the Sample plate must have a lid present at the start of the run. Clear the check box if the Sample Plate will not have a lid.	Not selected
Starting Sample Volume (µL)	Specifies the volume of sample that is initially present in the Sample Plate at the beginning of the run.	20 µL (0–1000 µL)
Number of Full Columns of Samples	Specifies the number of full columns of samples in the digestion plate.	12 (1–12)

Table **Addition step settings and descriptions**

Setting	Description	Default value (range)
Addition Number	Indicates the order in which the addition steps will be executed.	These values are fixed.
Addition Name	Specifies a name used to uniquely identify the addition step. Select a name from the list or enter a new name.	–
Reagent Deck Location	Indicates the deck location from which liquid will be drawn when this step runs.	This value is fixed.
Addition Volume (µL)	Specifies the volume of each reagent that will be transferred from its respective reagent plate, into the Sample plate.	10 µL (0–1000 µL)
Mixing Cycles	Specifies the number of mixing cycles that will be used to homogenize the combined samples and reagents after reagent addition.	3 (0–30)
Incubation Time (min)	Specifies the amount of time that the Sample plate will be incubated (deck location 4) after receiving a reagent aliquot.	0 (0–300 min)
Incubation Temp (°C)	Specifies the temperature set point of the Peltier Thermal Station during the Sample plate incubation (deck location 4).	OFF (OFF, 4–75 °C)
Pause After Addition	Pauses the protocol after completing the tasks in the step and before starting the next addition step. See the *In-Solution Digestion: Single Plate v1.1 User Guide* for instructions on effectively using the Pause feature.	–
Labware Selection	Specifies the labware type that will be used for each addition step.	–
Plate Lidded	Specifies whether the labware has a lid. If selected, the corresponding plate must have a lid present at the start of the run. Clear the check box if the plate will not have a lid.	–
Use Tips for Addition	Specifies whether pipette tips (deck location 2) will be used instead of the bare probes to add this reagent into the Sample plate. To use tips, select the check box. To use the bare probes, clear the check box.	–
Number of Wash Cycles	Specifies the number of wash cycles after the reagent addition step has completed. If the Use Tips for Addition check box is selected, this step washes the pipette tips. If the Use Tips for Addition check box is cleared, the protocol washes the bare syringe probes.	3 (0 - 10)

Contacting Agilent Technologies

Web: https://www.agilent.com

Contact page: https://www.agilent.com/en/contact-us/page

Documentation feedback: documentation.automation@agilent.com

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