

Biotage® Initiator+ Alstra™

Getting Started Guide for Peptide Synthesis



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CONTENTS

1 Quick Start

- 1 Set up User Accounts
- 1 Navigation
- 2 Set Up the Peptide Synthesis
- 4 Prepare the System
- 6 Run the Peptide Synthesis and Monitor the Progress
- 7 Unload the Reactor Vial
- 7 Search, Display, and Export Results
- 7 Empty the Waste and Scrubber Kit
- 7 Turn Off the System
- 7 More Information

8 UV Monitoring

- 8 UV Measurements
- 8 Set Up a UV Deprotection Operation
- 8 Results

9 Hints and Tips

- 9 Preparing Reagents for Peptide Synthesis
- 9 Potential Side Reactions

10 Instrument Overview

- 10 Robot Automation
- 10 Reactor Vials
- 11 Rack Tray and Rack Setup
- 11 Solvent Inlets
- 11 Heating and Agitation
- 11 UV Monitoring (Optional)
- 11 Audible Alarm
- 11 Inert Atmosphere
- 12 Waste and Scrubber Kit and Vacuum
- 12 Cleavage
- 12 Organic Synthesis Mode
- 12 Compliance

13 Software Overview

- 13 Software Modes
- 13 Peptide Synthesis Mode
- 16 More Information

17 Switch Between Peptide and Organic Synthesis

- 17 Switch from Peptide to Organic Synthesis
- 18 Switch from Organic to Peptide Synthesis
- 20 Connections

21 Maintenance

- 21 Weekly Cleaning of the Liquid Path
- 21 Monthly Cleaning of the Liquid Path
- 21 Clean the Drain Valves
- 22 Clean the Exterior of the System
- 23 Clean the Microwave Cavity, IR-Sensor, and Oscillating Mixing Unit
- 24 Clean the Needle Nose Assembly
- 24 Clean the Accessories
- 25 Release the Pressure
- 25 Empty the Waste and Scrubber Kit
- 25 Replace the Fuses
- 25 Replace the UV-Inlet Tube

27 General Information

- 27 Consumables and Accessories
- 27 Manufacturer
- 27 Contact Us

28 Notes

Quick Start

Warning:

- Observe general as well as specific safety regulations for the use of the system and its accessories and consumables at all times, in order to reduce the risk of personal injury, fire, and explosion; see the "Warning Summary" in the "Biotage® Initiator+ Installation and Safety" document (P/N 355976).

Set up User Accounts

A user can have system owner and/or chemist privilege:

- » The chemist privilege gives the user the possibility to have a default amino acid palette, get e-mail notifications, and have the user name in the experiment results. Note that you do not have to set up a user to be able to set up and run peptide synthesis experiments.
- » The system owner privilege gives the user access to system mode, i.e. the user can change system settings, manage users, configure a network connection, save logs on a USB memory device, and calibrate the robot. It is possible to password protect an account with system owner privilege.

Note: User accounts can only be set up by a user with system owner privilege. If your company does not have a user with system owner privilege, use the factory default account (factory password = 1234).

To add a new user:

- Log into the software's system mode:
 - If in peptide synthesis mode, press **Menu** and select **Main Menu** in the appearing menu.
 - Press **System** in the main menu.
 - Select your user account and press **OK**. If your account is password-protected, the **Input Password** dialog opens. Enter your password (use the factory password the first time) and press **OK**.
- Select the **Manage Users** tab and press **New**. The **User Editor** dialog opens.
- Press the **Name** text box and enter the user name.
- Press the **Password** text box and enter the password.
- If the system has been connected and configured to your network, it is possible to allow the user to request an e-mail when an experiment has been completed and when user intervention is required during a peptide synthesis, e.g. when a liquid needs to be replenished or a sample of the resin is scheduled to be removed. To enter the user's e-mail address, press the **E-mail** text box.
- Press the **Roles** text box and select the user privilege.

- To save the new account, press **Save**.
- To return to the main menu, press **Main Menu**.

Navigation

Enter peptide synthesis mode by pressing **Peptide Synthesis** in the main menu. This opens the synthesis wizard, which will guide you through the setup of a peptide synthesis.

Navigate through the seven steps of the wizard by pressing ► (next) or ◀ (previous), or by pressing the bullets in the top pane; see Figure 1. Each bullet represents a step in the wizard.

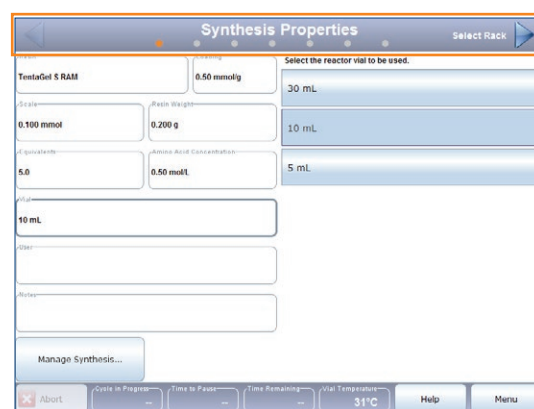


Figure 1. The top pane (highlighted) in the first step of the synthesis wizard.

Instructions are presented in each step of the wizard. On some pages you may need to scroll to see all information. Scroll by flicking a finger across the screen or by pressing the appearing green arrows, ▼ or ▲. The bottom pane contains information fields for monitoring the ongoing synthesis.

In all steps of the wizard, you can access more options via the **Menu** button (see Figure 2). The menu options are further

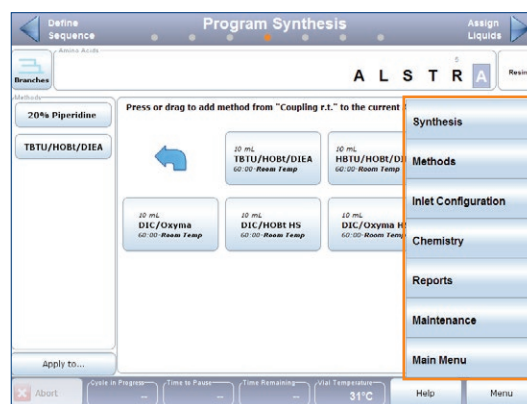


Figure 2. Menu options (highlighted) available in all wizard views.

described in “Software Overview” on page 13. For more detailed information you may refer to the online help by pressing the **Help** button in the bottom pane.

Navigate in the menu options and back to the synthesis wizard by pressing the desired view in the navigation path in the top pane (see Figure 3). You can also return to the wizard by pressing **Menu** and selecting **Synthesis** in the appearing menu.

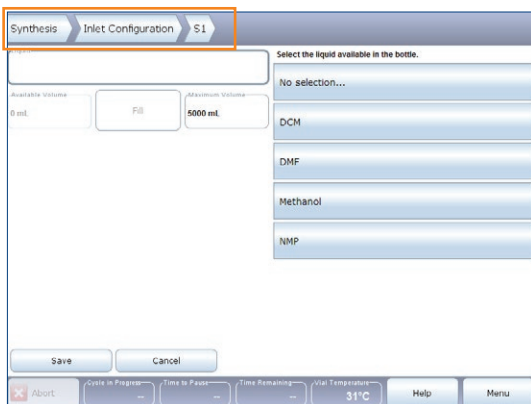


Figure 3. The navigation path (highlighted) in the top pane.

Set Up the Peptide Synthesis

Note: When heating a reaction mixture, ensure that the target temperature is at least 20°C below the boiling point of the solvent used (i.e. with a boiling point of 80°C, set the target temperature between room temperature and 60°C).

1. Enter peptide synthesis mode by pressing **Peptide Synthesis** in the main menu. This opens the wizard in the **Synthesis Properties** view; see Figure 4.

Press the text boxes to select resin, loading, scale, resin weight, equivalents of amino acids, amino acid concentration, and reactor vial size. It is optional to select user and enter notes.

Synthesis on 5 µmol – 2 mmol scale is possible. Scale is dependent on loading and swelling characteristics of the

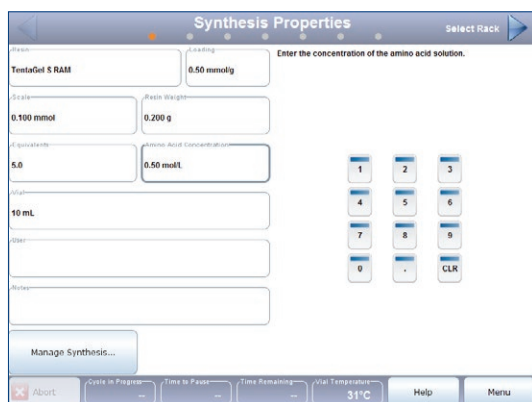


Figure 4. The Synthesis Properties view in the synthesis wizard.

chosen resin. Refer to “Hints and Tips” on page 9 or the online **Help** for more information on resin.

You can also import a peptide synthesis setup that has been saved on a USB memory device by connecting the device to the USB port at the front of the system and then pressing **Manage Synthesis....** See the online **Help** for more information.

2. Press ►. This opens the **Select Rack** view; see Figure 5. Press the amino acid rack setup you will use for the synthesis.

Note: The 32 x 30 mL rack setup is delivered with the system. The other rack setups are optional.

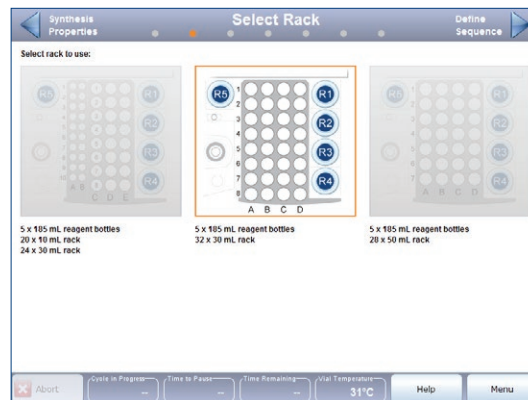


Figure 5. The Select Rack view in the synthesis wizard.

3. Press ►. This opens the **Define Sequence** view; see Figure 6. Enter the sequence you want to process; select the amino acids from the palettes. Toggle between palettes of standard, variant, and custom amino acids by pressing ◀ or ▶ in the **Selected Palette** pane.

The **Selected Amino Acid** pane shows details of the amino acid selected in the **Amino Acids** pane. If a variant is available, the **Variants** button will be enabled.

To copy, move (cut and paste), or delete an amino acid from the sequence, press and hold on the amino acid in the **Amino Acids** pane to open a menu with these options.

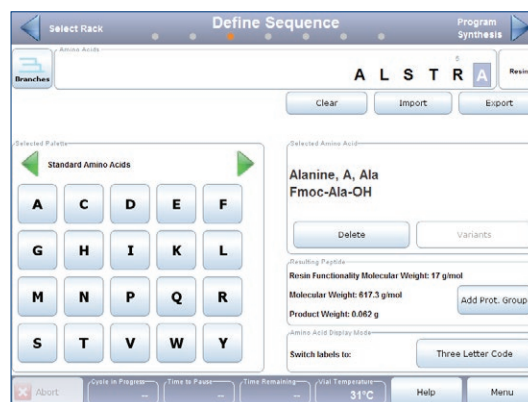


Figure 6. The Define Sequence view in the synthesis wizard.

The **Resulting Peptide** pane displays the calculated molecular and product weight. The product weight is calculated based on 100% yield of the synthesis. To add the molecular weight of one or more protecting groups, select the amino acid you want to add the group(s) to, press **Add Prot. Group**, and select the group(s) to add in the **✓** column.

To set up a cyclic, side-chain modified, or branched peptide, press **Branches**. See the online **Help** for more information.

You can also import a sequence (without cycles, side-chain modifications, branches, and protecting groups) via a USB memory device:

- Enter the sequence on your computer using Notepad or a similar text editing program.
- Type your sequence using one letter codes (upper case letters only, e.g. C) and/or three letter codes (first letter upper case, then lower case, e.g. Cys). Delimiters are required when using three letter codes; use space, comma, minus, tab, or new line.
Note: Lines beginning with “;” (semicolon) will be ignored and not imported. For more details, please refer to the online **Help**.
- Create an “initiator” folder on the root of the USB memory device, and in that folder create a sub-folder named “exported_sequence”.
- Save the text file (*.txt) in the “exported_sequence” folder.
- Eject the USB memory device from your computer.
- Connect the USB memory device to the USB port at the front of the system.
- Press **Import** and select the file you want to import in the dialog that opens. Unknown amino acids and amino acids with variants will be highlighted in red, and a dialog for handling them will open. See the online **Help** for more information.

To export a sequence, connect a USB memory device to the USB port at the front of the system and press **Export**. Enter the desired file name in the dialog that opens.

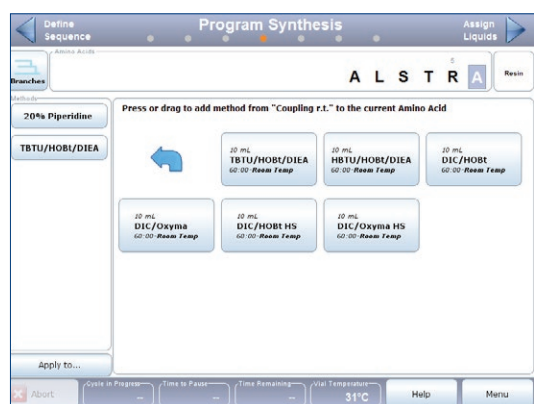


Figure 7. The Program Synthesis view in the synthesis wizard.

- Press **►**. This opens the **Program Synthesis** view where you define the methods for the current synthesis; see Figure 7. The system is delivered with a set of predefined methods. Custom folders and methods will also appear as options in the **Program Synthesis** view. Only methods relevant to the selected vial size are displayed.
- Select an amino acid by pressing it. Assign one or more methods to the selected amino acid by pressing one method at the time. Methods will appear in the **Methods** pane in the order you select them. You can also drag a method to a desired position in the **Methods** pane.

Press **⬅** to go up one level in the folder hierarchy, i.e. to the parent folder of the current.

To apply the selected method(s) to all or a selection of amino acids, press the **Apply to...** button and select **Apply to All**, **Apply Towards C** or **Apply Towards N**, where C and N refers to the C-terminus and N-terminus respectively.

To edit the order of the methods, press and drag the method you want to move to the desired place in the **Methods** pane.

To remove a method, drag it out of the **Methods** pane or press and hold on it until a menu appears and select **Remove**. The option **Remove All** will remove all methods from the **Methods** pane for the selected amino acid.

To edit a method for the current sequence or amino acid only, press the method you want to edit in the **Methods** pane. In the dialog that opens:

- Press the operation in the process lineup that you want to edit. When selected, it is highlighted in orange.
- Press the text box you want to edit and enter the data.
- For further edit options, press and hold on the operation you want to edit until a menu appears (see Figure 8). Refer to “Methods” on page 13 or the online **Help** for more information.
- Press **Save** to save the changes to the method and return to the **Program Synthesis** view.



Figure 8. In the method edit view, press and hold an operation to open a menu for options.

Note: In the **Program Synthesis** view, methods can only be edited for the current sequence. To create and save custom methods for reuse, see “Methods” on page 13 or the online **Help**.

6. Press ►. This opens the **Assign Liquids** view, displaying the selected rack, a table of the required liquids, the solvent inlet configuration, and a vessel information field where information on the selected vessel is displayed. See Figure 9.



Figure 9. The Assign Liquids view in the synthesis wizard.

7. Verify that the solvent inlet configuration is correct in the **Inlet Configuration** pane. If correct, go to step 8. To change the configuration:

- a. Press **Menu** and select **Inlet Configuration** in the appearing menu.
- b. Select the inlet you want to change.
- c. Press the **Liquid** text box and select the liquid that you want to assign to the bottle; see Figure 10. The system comes with predefined solvents and reagents. To add other liquids, see “Chemistry” on page 15, or the online **Help**.

Note: Always assign solvents to the S1 and S2 bottles.

Note: As the needle used for S2 and S3 is cleaned after each dispensation using the solvent in the S2 bottle, the S2 solvent has to be compatible with the S3 liquid, e.g. do not use DCM in S2 when using piperidine in S3.

Note: Assign activators, bases, and miscellaneous reagents, if used, to bottle S3 (normally 20% piperidine in DMF).

- d. Press the **Maximum Volume** text box and enter the maximum amount of liquid the bottle can hold.
- e. Press the **Available Volume** text box and enter the amount of the liquid that is present, or press the **Fill** button for a full bottle.
- f. Press **Save**. A dialog will remind you to go to the **Maintenance** view to prime the inlet. Pressing **OK** will reopen the **Inlet Configuration** view. For more information on priming, see “Prime the System” on page 5.

- g. To configure other solvent inlets, repeat steps b to f.
- h. Press **Synthesis** in the top pane or in the **Menu** to return to the synthesis wizard and verify the new inlet configuration in the **Assign Liquids** view.

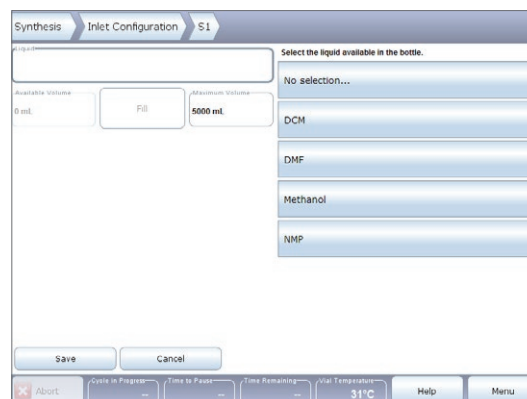


Figure 10. View to configure a solvent inlet.

8. Assign liquids to vessels and bottles on the rack tray either by pressing the button **Place Liquids...** and choose to place them by alphabetical or sequence order, or drag the liquids one by one from the table to the desired vessels and bottles. Liquids highlighted in orange are unassigned, while unmarked liquids are assigned.
9. Press ►. This opens the **Calculation Table** view (see Figure 11) showing details for the rack setup and preparation of amino acids, reagents, and solvents. You may need to scroll to view all data; press ▼ or ▲, or flick a finger up or down the screen.

Assign Liquids

Calculation Table

Running Synthesis

Resin

TentaGel S RAM

Loading

0.50 mmol/g

Quantity

0.200 g

Scale

0.100 mmol

Resin Functionality

Molecular Weight

17 g/mol

Molecular Weight

617.3 g/mol

Product Weight

0.052 g

Pos	Add	Chemical Name	Equivalents	Mol Mass [g/mol]	Mass [g]	Volume [mL]	Dissolve Volume [mL]	Concentration [mol/L]	Total Volume [mL]
A1	A	Fmoc-Ala-OH	5.0	311.3	0.327	1.842	0.5	2.1	
A2	L	Fmoc-Leu-OH	5.0	353.4	0.194	0.945	0.5	1.1	
B1	R	Fmoc-Arg(Pht)-OH	5.0	648.8	0.357	0.804	0.5	1.1	
C1	T	Fmoc-Thr(tBu)-OH	5.0	397.5	0.219	0.924	0.5	1.1	
D1	S	Fmoc-Ser(tBu)-OH	5.0	383.4	0.211	0.931	0.5	1.1	
R1		DIEA 2M in NMP	10.0	129.2	2.787	5.213	2.0	6.0	
DN		UNDEFINED	0.0	430.4	0.745	4.4	0.0	4.4	

Export to USB

Abort

Cycle in Progress

Time to Pass

Time Remaining

31°C

Help

Menu

Figure 11. The Calculation Table view in the synthesis wizard.

10. To get a printable calculation table, connect a USB memory device to the USB port at the front of the system and press **Export to USB**.

Prepare the System

Fill the Solvent Bottles

Fill the three solvent bottles with the liquids to be used. Ensure that no foreign matter (e.g. molecular sieve) is present in the bottles. If necessary, filter the liquids.

Prime the System

Prime each solvent inlet that you have assigned another liquid to than the one used in the previous experiment, or where the bottle has run dry and been replenished.

Notice: Handle chemical and liquid waste according to the Safety Data Sheets and to local/national guidelines on laboratory safety procedures.

1. If necessary, empty the waste reservoir.

Note: Before emptying the waste reservoir, the pressure should be released. See “Release the Pressure” on page 25.

2. Verify that the scrubber setup is suitable for the synthesis; see “Waste and Scrubber Kit and Vacuum” on page 12.
3. Ensure that the vacuum is turned on.
4. Press **Menu** and select **Maintenance** in the appearing menu.
5. Prime each solvent inlet that you have assigned a new liquid to or where the bottle has run dry and been replenished:
 - a. Ensure that the new liquid is filled up.
 - b. Start the priming by pressing **Prime S1, S2, S3, or All**.
 - c. If necessary, repeat steps a to b to prime another solvent inlet.

Note: Ensure that solvents have been assigned to the S1 and S2 bottles.

Note: Ensure to prime the S2 inlet before priming the S3 inlet as the system will use some of the solvent assigned to S2 to prime S3. Ensure that the liquids are compatible, e.g. do not use DCM in S2 when using piperidine in S3. For more details on priming, refer to the online **Help**.

Prepare the Amino Acid Rack(s) and Reagent Bottles

Refer to “Hints and Tips” on page 9 or the online **Help** for more information.

1. Print the calculation table created previously in step 10 on page 4.
2. Prepare the amino acids according to the calculation table:
 - a. Weigh the amino acids and pour them into empty vessels.
 - b. Add solvent and ensure that the amino acids are fully dissolved.
 - c. Ensure to load the amino acid vessels into the correct rack positions.
3. Fill the required reagents (activators, bases, and/or miscellaneous) into the reagent bottles or vessels according to the calculation table. If using vessels, ensure to load them into the correct positions on the amino acid rack(s).
4. If applicable, load empty vessels for preactivation or premixing into the correct positions on the amino acid rack(s).

Load the Amino Acid Rack(s) and Reagent Bottles

1. Load the prepared amino acid rack(s) onto the rack tray; see position(s) in the calculation table.
2. Load the prepared reagent bottles onto the rack tray; see positions in the calculation table.
3. Place the appropriate cover plates on the amino acid rack(s) and reagent bottles.

If the system is equipped with an inert gas manifold (optional), the cover plates should be placed so the nozzles on the manifold are depressed to enable inert gas to flow (see left image in Figure 12). Placing the cover plates in the opposite direction will cover the vials, but not allow the gas to flow as the nozzles will not be depressed (see right image in Figure 12).

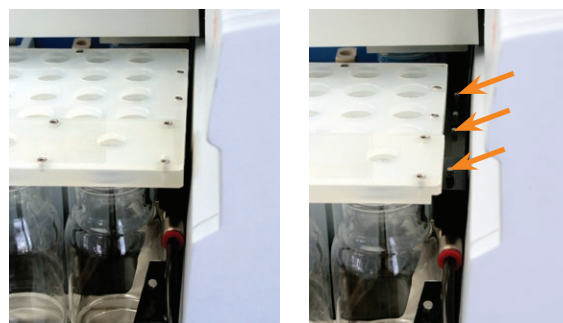
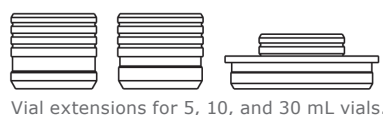


Figure 12. Left: Cover plates placed to depress gas nozzles on an Inert Gas Manifold, allowing gas to flow. Right: Cover plates placed to not depress gas nozzles, if no gas is required.

Prepare and Insert the Reactor Vial

1. Weigh the resin according to the calculation table and place it in a new reactor vial. If using the 5 mL vial, only fill the narrow part of the vial to about half way with resin (in solvated form) to obtain efficient mixing.
2. Insert a vial extension of the correct size into the vial.
3. If a 5 or 10 mL vial is selected, then ensure that a vial collar is inserted into the microwave cavity. If a 30 mL vial is selected, ensure to remove any vial collar.
4. Load the vial into the microwave cavity using the vial loading tool. Ensure to insert the vial correctly.

Note: Always use the vial loading tool when inserting reactor vials into the microwave cavity.



Vial extensions for 5, 10, and 30 mL vials.



Use a vial collar for 5 and 10 mL vials.



Amount of resin in the 5 mL vial.



Insert the vial using the vial loading tool.

Run the Peptide Synthesis and Monitor the Progress

1. Ensure that there is sufficient space in the waste reservoir before starting the synthesis.
2. Ensure that the vacuum is turned on.
3. If using inert gas, ensure that the gas is turned on and the inert gas valve is open. The valve can be opened at the **Peptide Synthesis** tab in system mode.
4. Press ► in the synthesis wizard. This opens the **Running Synthesis** view.
5. Press **Start Synthesis** to start the synthesis. The synthesis progress is now displayed; see Figure 13.

To stop the synthesis, press **Abort** and confirm the abort dialog(s). The settings for the current synthesis are saved. A pop up dialog appears where you can choose to clear the synthesis settings and set up a new synthesis, set up a new synthesis based on the aborted run, or show a report of the aborted run.

To pause a synthesis, press **Pause...** and select **Pause After Current Operation**, **Pause After Next Deprotection**, or **Pause After Next Coupling** in the appearing menu. A countdown timer will indicate the estimated time until pause.

When a synthesis is paused, it is possible to add and delete methods to/from unprocessed amino acids and edit unprocessed methods.

To undo a scheduled pause, press **Pause...** and select **Cancel Scheduled Pause**.

To resume synthesis, press **Resume**.

6. The progress of the synthesis is displayed in detail in the **Running Synthesis** view (see Figure 13):
The amino acid being processed is highlighted (blue) in the **Amino Acids** pane.

The methods assigned to the current amino acid are displayed in the **Methods** pane as a process lineup. The specific operations of the current method are displayed in the **Operations** pane.

The progress of the current method and its operations is illustrated with countdown timers and color codes. The countdown timers show the estimated time remaining for the method and for each of its operations. If the method contains a UV monitored Fmoc deprotection, “N/A” is displayed for both the method and the UV deprotection. The color codes are:

- » Green = completed
- » Blue = remaining to be performed

By default the settings of the operations will be shown in text as they are performed. You can hide the settings by pressing the button **Hide Details**.

When the system is performing microwave heating, a process graph with real time measurements of temperature and applied power is displayed.

For information on the data that is displayed during a UV monitored Fmoc deprotection, see “Results” on page 8.

You may need to scroll to view all information; press ▼ or ▲, or flick a finger up or down the screen.

The system’s running status is displayed in information fields in the bottom pane, which is available in all views in peptide synthesis mode. When the system is idle, the information fields show “--” and the **Abort** button is disabled.



Figure 13. The Running Synthesis view during a run.

The information fields are:

- » **Cycle in Progress:** States the number of the cycle in progress/the total number of cycles and, in brackets (), the amino acid being processed.
 - » **Time to Pause:** If **Pause** is activated, this field states the estimated time until the system pauses.
 - » **Time Remaining:** States the estimated time remaining for the synthesis in progress. If the synthesis contains a UV monitored Fmoc deprotection, “N/A” is displayed.
 - » **Vial Temperature:** States the current vial temperature. When microwaves are generated, (🔥) is displayed in this field.
7. When the synthesis is finished, a pop up dialog opens. You can choose to:
 - » **Start New Synthesis:** This choice brings you to the first step in the synthesis wizard. All settings from the previous synthesis are cleared.
 - » **Repeat Current Synthesis:** This choice brings you to the first step in the synthesis wizard. All settings from the previous synthesis are saved, and you can edit and/or run the synthesis again.
 - » **Display Report:** Shows a report of the finished synthesis. Press **Export to USB** to save the report as a PDF file on an inserted USB memory device.

Unload the Reactor Vial

Place the vial loading tool over the reactor vial and then gently press the vial eject lever to release the vial from the microwave cavity. Never remove the reactor vial by only using the vial eject lever.

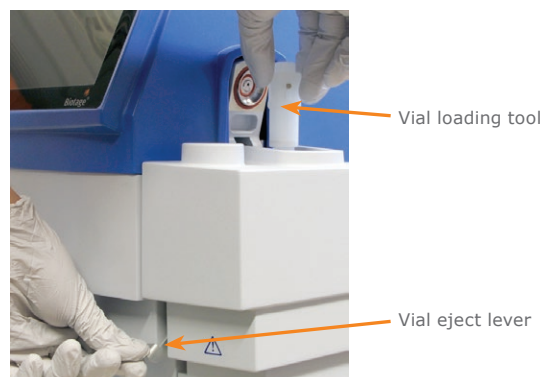


Figure 14. Unloading the reactor vial.

Search, Display, and Export Results

1. Press **Menu** and select **Reports** in the appearing menu.
2. Enter the desired search criteria by pressing the **Date**, **User**, and/or **Sequence** text box.
3. To search the results, press **Search Results**. All results that match the search criteria are listed.
4. To display a report, select the desired result (row). To scroll the list of results or the report, flick a finger up or down the screen or press ▼ or ▲.
5. To save the report as a PDF file on a USB memory device, connect the device to the USB port at the front of the system, press **Export to USB**, and enter the desired file name in the dialog that opens.

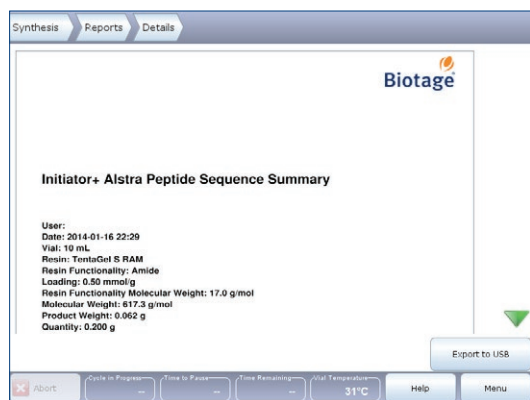


Figure 15. The Reports view after a completed synthesis.

Empty the Waste and Scrubber Kit

Notice: Handle chemical and liquid waste according to the Safety Data Sheets and to local/national guidelines on laboratory safety procedures.

The 500 mL scrubber bottle needs to be emptied and replenished after each synthesis. The 10 liter waste reservoir needs to be emptied when it is full. Before emptying or exchanging a scrubber bottle or waste reservoir, it is important that the vacuum is turned off and the pressure released. See steps 1 and 2 in “Turn Off the System” below.

Turn Off the System

1. Turn off the vacuum pump or, if your system is equipped with an external vacuum system, close the valve.
2. Release the pressure:
 - a. Press **Menu** and select **Maintenance** in the appearing menu.
 - b. Press **Empty Wash Station** repeatedly until equilibrium is achieved.
3. Turn off the software by pressing **Menu**, **Main Menu**, and then **Shut Down**.
4. When the message “Safe to power off” appears on the screen, turn it off. The mains switch is at the front of the system.

More Information

For more information and instructions, see “Software Overview” on page 13 or refer to the online **Help**.

UV Monitoring

If your system is equipped with a UV detector, it is possible to perform UV monitored Fmoc deprotections in the 10 or 30 mL vial using 20% piperidine in DMF or 20% piperidine in NMP. Based on the UV data, you can set the number of deprotection steps (iterations) to be performed, and adjust the next coupling method.

UV Measurements

The removal of the Fmoc group can be monitored by measuring the UV absorption of the dibenzofulvene-piperidine adduct formed during Fmoc deprotection with a piperidine solution.

The concentration of the adduct in DMF and NMP is known from the absorption. The maximum concentration at full deprotection is calculated from the scale given by the user. The two concentrations are compared and if the measured value falls within the set threshold, the deprotection is regarded as complete.

Example: The first measurement after 3 minutes gives a 97% deprotection and a second measurement after 10 minutes gives a 3% deprotection. The second deprotection iteration is below the threshold that is set to 5% and is regarded as complete. No more deprotection iterations will be performed.

Set Up a UV Deprotection Operation

1. Select the **Threshold** text box and enter the value in percent of the theoretical absorption after complete deprotection. The absorption will be measured after each deprotection iteration and when the absorption reaches or goes under this value, the deprotection will be considered completed.
2. Select the **Iterations** text box and enter the maximum number of deprotection iterations. If/when the threshold is reached, no further iterations will be performed.
3. Select the **UV Reference Liquid** text box and select the reference solvent, absorption=0 (zero), for the UV measurements. This should be the deprotection solvent, DMF or NMP.
4. Set the deprotection parameters for each iteration and what to do if the threshold is reached during the iteration:
 - a. Select an iteration (**A**) in the **Deprotection Iterations** list.
 - b. Select the **Time** text box and enter the reaction time.
 - c. Select the **Liquid** text box and select the deprotection liquid, 20% piperidine in DMF or 20% piperidine in NMP.
 - d. To extend the reaction time for the next coupling method if the threshold is reached, select the **Extend Next Coupling** text box and enter the additional reaction time.
 - e. To repeat the next coupling method if the threshold is reached, select the **Repeat Next Coupling** text box and select the number of additional times to run the method.

Figure 16. A. The deprotection parameters for each iteration and what to do if the threshold is reached during the iteration. B. The parameters for what to do if the threshold is not reached during any of the deprotection iterations.

5. Set the parameters for what to do if the threshold is not reached during any of the UV deprotection iterations:
 - a. Select the last item (**B**) in the **Deprotection Iterations** list.
 - b. To pause the synthesis after the last iteration, press the **Schedule Pause** text box so that **Yes** is displayed. Note that the resin is washed before the synthesis is paused.
 - c. To extend or repeat the next coupling method, select the corresponding text box and enter the additional reaction time or select the additional times to run the method.

Results

During the UV monitored Fmoc deprotection you will see a list of the iterations and their parameters and the outcome:

- = The threshold was not reached during the iteration.
- = The threshold was reached during the iteration.
- = The threshold was either reached during the first iteration or it was not reached during the last iteration.

The same information is presented in the synthesis report. The report also contains a diagram with the number of deprotection iterations performed on each amino acid. If the deprotection was successful, the bar is green. Otherwise it is red.

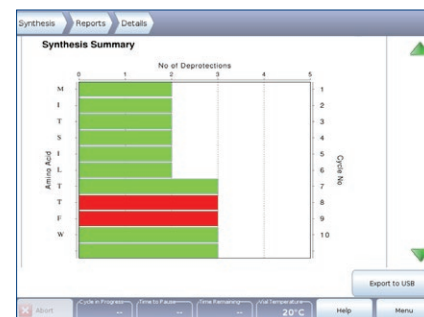


Figure 17. The report contains a diagram with the number of deprotection iterations performed on each amino acid.

Hints and Tips

Preparing Reagents for Peptide Synthesis

All reagents and amino acids need to be dissolved before use on the Biotage® Initiator+ Alstra™ system. Although we do not have a standard concentration, it is recommended to select the concentrations so that a minimum volume of 100 µL is aspirated and dispensed by the robot.

For amino acids, a concentration of 0.2–0.7 M is usually recommended for this system. In predefined Biotage methods, the recommended concentration is 0.2 M for the 5 mL vial, 0.5 M for the 10 mL vial, and 0.7 M for the 30 mL vial. At the higher concentration, the amino acid should be dissolved in NMP. Vigorous stirring may be needed to obtain complete dissolution. In cases where an amino acid is difficult to dissolve, a small addition of DMSO or NMP to the dry powder before adding the main solvent can be beneficial.

Note: There is a risk of oxidation of methionine if DMSO is used.

Scale is dependent on resin loading and type. When setting up a synthesis, two important aspects need to be considered:

- » The overall volume.
- » The resin to solvent ratio.

The minimum volumes for the vials (see Table 1) are important as they are the minimum volumes for efficient microwave heating and accurate temperature measurements. Do not use microwave heating if the minimum volumes are not reached. The maximum volumes are set to prevent spillage and to obtain efficient washing. All recommendations are based on pre-swollen resin.

The resin to solvent ratio is dependent on the resin type. We recommend that only resins with a bead size in the range of 100 to 200 mesh are used. It is very difficult to predict accurate solvation for all resins. The 5 mL reactor vial needs a higher solvent to resin ratio. As different resin and solvent combinations have different solvation characteristics, the user should manually check that the resin is adequately solvated by the volume that is dispensed by the planned methods, for the particular type of resin and solvent employed, before setting up the synthesis.

Example: If your method dispenses a total of 3 mL liquid, take the planned amount of resin (e.g. 0.2 g) and let it swell in the solvent of your choice so that it is properly solvated, empty the reactor vial of all extra liquid, and add the 3 mL of the solvent that is closest to the conditions of the method. Ensure that the resin–solvent mixture moves freely and that it is between the minimum and maximum volumes of the vial; please refer to Table 1 for guidelines.

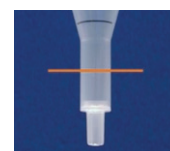
Vial Size	Scale (mmol)	Possible/Recommended Volume (resin + liquid)
5 mL	PEG based: 0.005–0.010 Polystyrene: 0.005–0.040	0.6–3.5 mL/0.6–1.2 mL
10 mL	PEG based: 0.05–0.40 Polystyrene: 0.05–1.00*	3.5–10.0 mL/3.5–10.0 mL
30 mL	PEG based: 0.50–0.75 Polystyrene: 0.5–2.0†	4.5–20.0 mL/10.0–20.0 mL

* For UV monitored Fmoc deprotections the scale is 0.05–0.5.

† For UV monitored Fmoc deprotections the scale is 0.5–1.0.

Table 1. Vial selection and scale.

Note: In order to obtain efficient mixing for the 5 mL vial, it is important to only fill the narrow part of the vial to about half way with resin (in solvated form) and the remainder with solvent. See the image to the right.



Potential Side Reactions

As solid-phase peptide synthesis (SPPS) at elevated temperatures can enhance some side reactions of certain residues and sequences, care must be taken when synthesizing peptides containing these residues and sequences. Below is a list of known side reactions that may occur for certain residues, as well as some recommendations to circumvent side reactions:


- » Epimerization of His residue – couple His derivatives at room temperature or below 50°C. Coupling with a weaker base or non-basic couplings may also help to suppress the epimerization.
- » Epimerization of Cys residue – avoid preactivation and use base free activation. Couple at room temperature or below 50°C.
- » δ-lactamisation of Arg residue – couple at room temperature or below 50°C.
- » Aspartimide formation – use 5% piperazine as deprotection base after incorporation of Asp-XX residue (XX = Gly, Asn, Ser, Thr, Gln, Arg (Pmc)). Longer deprotection times may be needed as piperazine is a weaker base than piperidine.
- » Diketopiperazine formation on the second coupling when using C-terminal proline and a resin with an ester linker – use a trityl-based resin.

Note: We do not recommend the use of microwave heating during the Fmoc deprotection step due to the potential risk of promoting side reactions such as aspartimide formation and epimerization. All deprotections should be performed at room temperature.

Instrument Overview



Figure 18. A = touch screen, B = USB port, C = mains switch, D = vial eject lever, E = microwave cavity and needle wash station (behind the cavity), F = holder for the vial collar, G = cavity cover, H = waste tray, I = robot arm with dispensing needles, J = cover plate, K = amino acid rack, L = rack tray for amino acid rack(s) and reagent bottles, M = Alstra pump module, N = accessories holder, and O = solvent inlets, and

The Initiator+ Alstra system, designed for fully automated microwave assisted peptide synthesis, is equipped with a 10.4" touch screen used for experimental planning, instrument control, and reaction monitoring. A built-in synthesis wizard assists the programming. The system's running status is displayed in information fields in the bottom pane, which is visible regardless of view. If microwaves are generated,  is displayed. When the system is idle, the information fields show "--" and the **Abort** button is disabled.

Robot Automation

The needles on the vertical robot arm are used to dispense liquids into the reactor vial inside the microwave cavity and into vessels used for preactivation or premixing. Liquids can be aspirated from

the vessels and bottles located on the rack tray and from the three solvent bottles using three integrated digital syringe pumps.

After completion of a liquid operation, the robot arm moves the needles to the needle wash station. At the needle wash station, excess liquid is discarded and the needles are rinsed both externally and internally using system solvent (S₁). The needle wash station is emptied using vacuum.

Liquid Level Detection

The system can detect the liquid surface during aspiration. This ensures that the cleaning of the needles is minimized, as only the needle tips have to be cleaned. Liquid level detection can be turned on or off in system mode, at the **Peptide Synthesis** tab.

Vial Overfill Detection

The system can detect if the reactor vial is overfilled before dispensing liquid. Vial overfill detection can be turned on or off in system mode, at the **Peptide Synthesis** tab.

Reactor Vials

The system can process reaction volumes (resin + liquid) between 0.6 and 20 mL.

The following reactor vials are available:

- » 5 mL (0.6–3.5 mL reaction volume)
- » 10 mL (3.5–10.0 mL reaction volume)
- » 30 mL (4.5–20.0 mL reaction volume)

The reactor vials are marked with minimum and maximum fill volumes. For recommended volumes, see Table 1 on page 9. Do not exceed or fall below a vial's specified volume range (in brackets above) and only use new vials supplied by Biotage.

Load and Unload Reactor Vials

Always insert a vial extension of the correct size into the reactor vial before loading the vial into the microwave cavity using the vial loading tool. Ensure to insert the vial correctly and only use

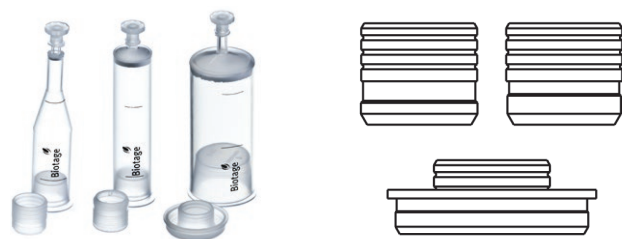


Figure 19. Left: Three reactor vials are available: 5, 10, and 30 mL. Right: Vial extensions for the 5, 10, and 30 mL vials.

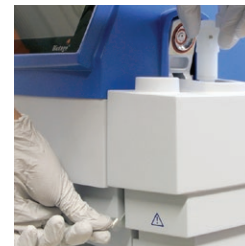


Figure 20. Left: Vial loading tool for inserting and removing reactor vials safely and a vial collar. Right: Never unload a reactor vial by only using the vial eject lever.

vial extensions supplied by Biotage. For 5 mL and 10 mL vials, use a vial collar inserted into the microwave cavity.

Unload a reactor vial by placing the vial loading tool over the reactor vial and then gently pressing the vial eject lever to release the vial from the microwave cavity.

If the temperature inside the reactor vial is above 59°C when the lid is opened, the text “Warning, hot vial!” is displayed in the bottom pane. Wait until the temperature drops to 59°C or lower before unloading the vial.

Rack Tray and Rack Setup

The rack tray can hold five reagent bottles (maximum load volume is 185 mL each) and one or two amino acid racks.

The following three amino acid rack setups can be used (from left):

- » 20 x 10 mL and 24 x 30 mL
- » 32 x 30 mL (delivered with the system)
- » 28 x 50 mL



Figure 21. Amino acid racks used with the Initiator+ Alstra system.

The reagent bottles are specially designed for the system and can only be ordered from Biotage.

Solvent Inlets

The system is equipped with three solvent inlets found on the right hand side of the system. Three GL45 laboratory glass bottles (brown) are delivered with the system; one 5 liters, one 2 liters, and one 1 liter.

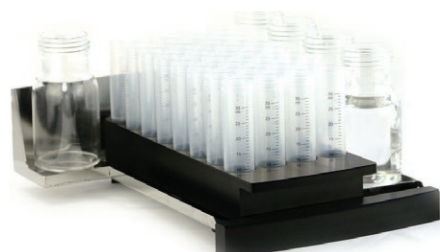


Figure 22. Rack tray with the default rack setup, 32 x 30 mL, delivered with the system.

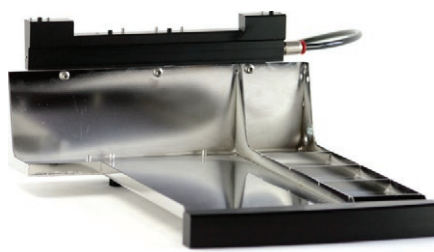


Figure 23. Rack tray equipped with the Inert Gas Manifold.



Figure 24. Three solvent inlets are available on the right hand side of the system.

Heating and Agitation

When the reactor vial has been inserted into the microwave cavity and the cavity lid has been closed, high-frequency microwaves (2.45 GHz), generated by the magnetron, heat the reaction mixture. Microwave heated peptide syntheses can be performed at temperatures between 40°C and 100°C.

During the heating process, the reactor vial is agitated by an oscillating mixer, which ensures homogeneous heat distribution.

Interval mixing is available for wash operations and reactions at room temperature.

UV Monitoring (Optional)

If the system is equipped with a UV detector, it is possible to optimize the synthesis by performing Fmoc deprotections that are monitored and controlled by the UV detector. Based on the UV data, you can set the number of deprotection steps to be performed, and adjust the next coupling method (extend the reaction time and/or add coupling steps).

Audible Alarm

Operations that have to be performed manually by the user, e.g. removing a sample of the resin, can be prompted by an audible alarm.

Inert Atmosphere

If the system is connected to an inert gas supply, the microwave cavity can be flushed with inert gas during the heating and draining (emptying) process. Ensure that the regulator is set to 0.5 bar. For more information, please see the “Biotage® Initiator+ Installation and Safety” document (P/N 355976).

Cover plates for amino acid racks and reagent bottles prevent spillage and contamination. Cover plates for the default setup are delivered with the system. Additional foil septa, additional

cover plates, and cover plates for the optional rack setups can be ordered from Biotage.

Inert Gas Manifold (Optional)

If the system has been equipped with an Inert Gas Manifold (sold separately), decomposition of amino acids and reagents can be reduced by using inert gas in combination with cover plates.

Waste and Scrubber Kit and Vacuum

A central vacuum system, or a vacuum pump (sold separately, P/N 356330), is required for emptying the reactor vial and the needle wash station. In order to protect the vacuum system, we recommend that a waste and scrubber kit is used to stop volatiles and condensation from entering and damaging the vacuum system.

Set up the waste and scrubber kit as described in the instructions supplied with the kit. Consider the following when choosing scrubber setup:

- » If acidic fumes are generated, water saturated with sodium carbonate can be used in the scrubber bottle.
- » If basic fumes are generated, a water solution of 10% citric acid can be used in the scrubber bottle.
- » If aggressive volatile solvents (such as dichloromethane, THF, dioxane, or acetonitrile) are used, the scrubber bottle (empty) will need to be cooled with dry ice and either acetone or ethanol to be efficient.

The 500 mL scrubber bottle needs to be emptied and replenished after each synthesis. We recommend that no more than 100 mL of scrubber solution is used.

Always ensure that there is sufficient space in the waste reservoir before starting a synthesis.

Ensure to turn off the vacuum and release the pressure (see page 25) before opening the scrubber bottle or the waste reservoir.



Figure 25. The waste & scrubber kit contains a 10 liter waste reservoir, a 0.5 liter scrubber bottle, and the tubing shown above.

Cleavage

Reactor vial caps and plugs, which can be used for cleavage, are delivered with the system. Additional sets of reactor vial caps and plugs can be ordered separately.



Figure 26. From left: cap for 30 mL vials, cap for 5 and 10 mL vials, and plug for all vial sizes.

Organic Synthesis Mode

By modifying the system, it is possible to perform organic synthesis on it. Please refer to “Switch Between Peptide and Organic Synthesis” on page 17 and the “Biotage® Initiator+ Getting Started Guide for Organic Synthesis” (P/N 355975).

Biotage® Initiator Peptide Workstation

Biotage® Initiator Peptide Workstation is an optional accessory for manual microwave peptide synthesis and cleavage using any Initiator system in organic synthesis mode. The workstation can be used to perform various types of chemistry including solution phase and solid phase peptide synthesis, and organic and PNA synthesis.

Initiator Peptide Workstation consists of a specially designed microwave peptide vial and wash station. Peptide synthesis is performed in the microwave peptide vial (glass vial) under atmospheric conditions at temperatures up to 100°C using any Initiator system. The vial contents are quickly filtered and washed using the wash station connected to a vacuum pump.



Compliance

Only genuine Biotage consumables and accessories must be used in the system. Some of them are listed on page 27. To order consumables and accessories, see contact information on the back of this document or visit our website www.biotage.com.

Software Overview

Software Modes

- » **Organic Synthesis:** Perform organic synthesis experiments, and view and manage the results. Please refer to “Switch Between Peptide and Organic Synthesis” on page 17.
- » **Peptide Synthesis:** Perform peptide synthesis experiments, export and import user-defined data (folders with methods and liquids), and view and manage the results.
- » **System:** Change system settings, manage users, configure a network connection, save logs on a USB memory device, and calibrate the robot. Only users with system owner privilege can log into system mode.
- » **Service:** Service can only be performed by an authorized Biotage service engineer.

Peptide Synthesis Mode

The software in peptide synthesis mode consists of a step-by-step wizard for peptide synthesis and a menu with further options. The wizard guides the user through the steps for peptide synthesis as described in the “Quick Start” section on page 1. Navigate through the seven steps of the wizard by pressing ► (next) or ◀ (previous), or by pressing the bullets in the top pane. Each bullet represents a step in the wizard.

In addition to the wizard steps there is a **Menu** that is always accessible in the bottom pane; see Figure 27. The **Menu** options are: **Synthesis**, **Methods**, **Inlet Configuration**, **Chemistry**, **Reports**, **Maintenance**, and **Main Menu**.

When defining methods and chemicals, viewing reports, or performing maintenance tasks (**Menu** options), navigation is performed by pressing the desired view in the navigation path shown in the top pane (see Figure 28). The view displayed is the

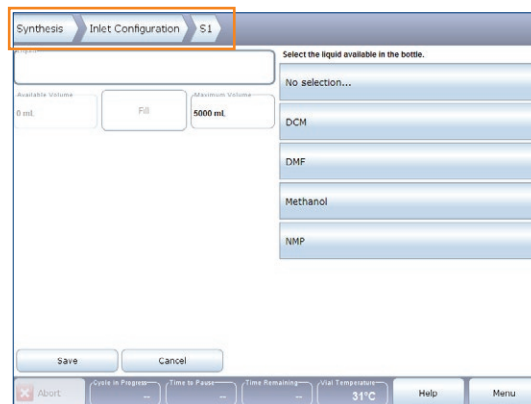


Figure 28. Pressing Synthesis in the navigation path (highlighted) in the top pane brings you back to the current wizard step.

rightmost. To return to the synthesis wizard, press **Synthesis** in the navigation path.

Synthesis

The **Synthesis** menu option brings you back to the current step in the synthesis wizard.

Methods

The **Methods** view is a method editor where you can set up, edit, and save frequently used methods. The software comes with a number of predefined methods. If desired, these can be copied and edited to your preferences.

Folders and methods created in the **Methods** view appear as selectable methods in the **Program Synthesis** view in the synthesis wizard. To only show methods designed for a certain vial size in the **Methods** view, press the **Show Methods for Vial** text box and select the desired vial size.

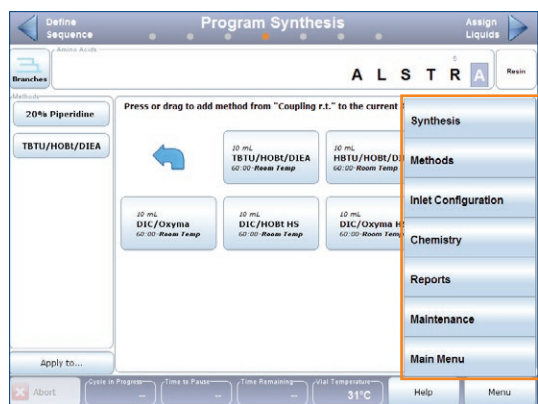


Figure 27. Menu options available in all wizard views.

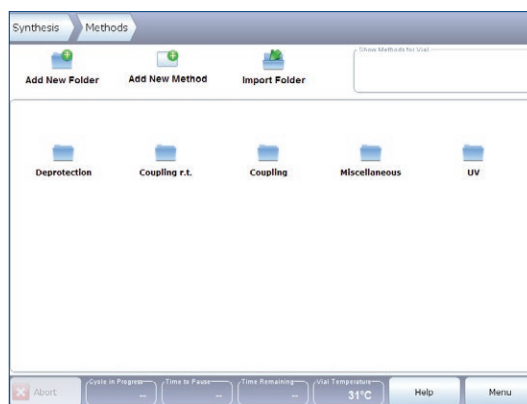





Figure 29. In the Methods view, you can set up, edit, and save frequently used methods and import and export entire folders.

Create a new folder by pressing . A folder can be copied, cut, renamed, and deleted by pressing and holding on the folder until a menu appears (see the image to the right).


Create a new method by pressing . A method can be copied, cut, and deleted by pressing and holding on the method until a menu appears.

Paste a folder or method by pressing and holding on an empty space in a folder or on a method until a menu appears.

To export a folder, insert a USB memory device, press and hold on the folder until a menu appears, select **Export Folder** in the menu, and enter the desired file name in the dialog that opens.

To import a folder, insert the USB memory device, press , and select the method folder you want to import. **Note:** The import includes the folder and all its sub-folders and methods, and all liquids used in the methods.

To edit a method, open it by pressing it. A method consists of one or more operations. To add, copy, move, or delete an operation, press and hold on it until a menu appears. See Figure 30.

To add a new operation at the end of the method, press . The following operations are available:

- » **Fill:** Used to dispense liquids into the reactor vial or a preactivation or premixing vessel. Enter a fixed volume for solvents. For coupling reagents and amino acids, you can enter a fixed volume or an **Equivalents Factor** that determines the molar relationship between the amino acid and the reagent. See the online **Help** for more information.
- » **Empty:** Used to drain the reactor vial using vacuum. Either specify the empty time or let the system detect when the vial is empty.
- » **Reaction:** Used for reactions at room temperature (RT) or at temperatures between 40°C and 100°C. When heating, ensure that the target temperature is at least 20°C below the boiling point of the solvent used in the



reaction (i.e. with a boiling point of 80°C, set the target temperature between room temperature and 60°C).

Note: The time parameter specifies the length of reaction time at the target temperature. The reactor vial is agitated continuously when heated and continuously or in intervals when at room temperature.

- » **Wash:** Used to wash the reactor vial. This operation is made up of a fill and an empty operation. The reactor vial can be agitated continuously or in intervals during the wash.
- » **Manual:** Used for operations that will be performed by the user, e.g. removing a sample of the resin. The operation will be prompted by a user-defined message and, if desired, an audible alarm. Select whether the cavity lid is to be opened or not before the operation.
- » **Premix Reaction:** Used to mix liquids that have been added into a premixing vessel. Enter the number of mixing iterations and the time to pause after all mixing iterations.
- » **Fill From Premix:** Used to dispense liquids into the reactor vial from a premixing vessel. Enter the number of times the premixing vessel is to be washed after the dispensation.
- » **UV Deprotection:** Used to perform a Fmoc deprotection that is monitored and controlled by the UV detector. Based on the UV data, you can set the number of deprotection steps to be performed, and adjust the next coupling method (extend the reaction time and/or add coupling steps). Please refer to the online **Help** for more information. Note that this operation is only available on systems equipped with a UV detector and for the 10 mL and 30 mL reactor vials.

If the system is connected to an inert gas supply, the microwave cavity can be flushed with inert gas during the heating and draining (emptying) process.

An operation highlighted in orange is open for editing. Modify a parameter by pressing the corresponding text box and entering/selecting the desired setting. To save changes, press **Save**.

Note: Ensure that the method is designed for the reactor vial size you are using; see the setting in the **Vial** text box.



Figure 30. Press and hold on an operation in the Edit Methods view to open a menu for options.

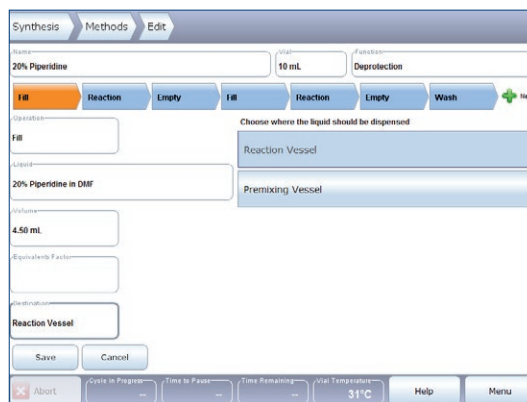


Figure 31. Press the text boxes to edit the selected operation, which is highlighted in orange in the Edit Methods view.

Inlet Configuration

In the **Inlet Configuration** view, you can specify the contents of the three solvent bottles (S1–S3). See Figure 32.

When a method is run, the software references the inlet configuration to determine which bottle contains the liquid used in a fill or wash operation. Therefore, it is important that the inlets are configured accurately. The solvent inlets are found on the system's right hand side. Ensure that the bottles are set up correctly according to the configuration as presented in the calculation table.

Note: Always assign solvents to the S1 and S2 bottles.

Note: As the needle used for S2 and S3 is cleaned after each dispensation using the solvent in the S2 bottle, the S2 solvent has to be compatible with the S3 liquid, e.g. do not use DCM in S2 when using piperidine in S3.

Note: Assign activators, bases, and miscellaneous reagents, if used, to bottle S3 (normally 20% piperidine in DMF).

Note: Prime each solvent inlet that you have assigned another liquid to than the one used in the previous experiment, or where the bottle has run dry and been replenished. See “Prime the System” on page 5 or the online **Help**.

Note: Ensure that the correct volume is entered each time a bottle is replenished. The system will issue a warning when you need to replenish a liquid.

Chemistry

The **Chemistry** view presents five options: **Solvents**, **Palettes and Amino Acids**, **Resins**, **Protecting Groups**, and **Activators, Bases and Miscellaneous**.

Solvents

In the **Solvent** view (see Figure 33), you can view the selection of solvents available. The system comes with a number of predefined solvents to choose from. These are locked (🔒) and cannot be modified or deleted. To add a solvent, press **New**. The parameters are: name, aspiration and dispensation rates, and

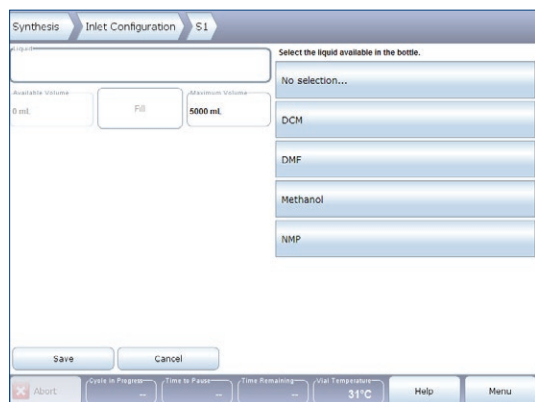


Figure 32. View to configure a solvent inlet.

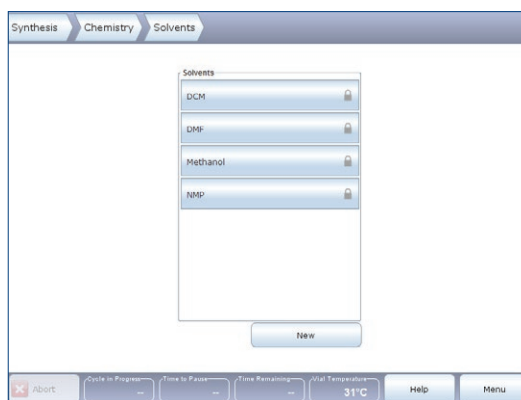


Figure 33. The Solvents view.

number of needle washes after a dispensation. We recommend 50 mL/min and no washes for solvents. Note that solvents assigned to S1 and S2 are always aspirated and dispensed at 50 mL/min.

A solvent can be copied or deleted by pressing and holding on it until a menu appears and then selecting **Copy** or **Delete**. To paste, press and hold an area in the **Solvents** pane until a menu appears, and then select **Paste**. The solvents are sorted in alphabetical order.

To edit a solvent, open it by pressing it and modify the relevant parameters.

Palettes and Amino Acids

In the **Palettes and Amino Acids** view (see Figure 34), you can create new amino acids and customized groups of amino acids (palettes) for the ones most frequently used. You can also copy, edit, move, and delete amino acids and rename and delete palettes. The system comes with two predefined palettes:

- » **Standard Amino Acids:** 20 standard amino acids. These cannot be modified, but can be copied to another palette.
- » **Variant Amino Acids:** Variants of standard amino acids.

The **Standard Amino Acids** palette is the default palette when setting up peptide synthesis. To change the default palette for

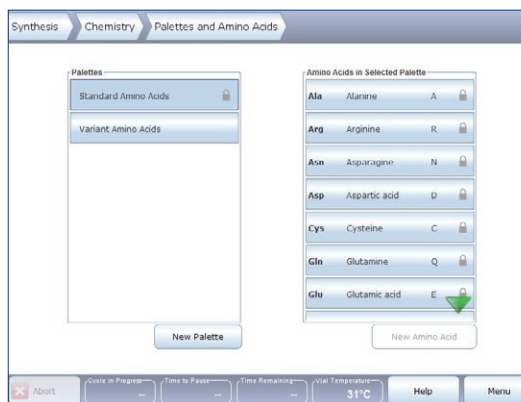


Figure 34. The Variant Amino Acids palette selected in the Palettes and Amino Acids view.

your user account, press and hold on the desired palette until a menu appears, select **Select as Default...** in the menu, and then select your user account in the dialog that opens.

To create a new palette, press **New Palette**. Rename or delete a palette by pressing and holding on it until a menu appears and then select **Rename...** or **Delete....**

To create a new amino acid, select a palette in the **Palettes** pane and press **New Amino Acid**.

To copy or move an amino acid from one palette to another, start by selecting the palette from which you want to copy or move it. In the list of amino acids, press and hold on the desired amino acid until a menu appears and then select **Copy** or **Cut**. To paste, select the palette you want to add the amino acid to, press and hold an area in the **Amino Acids** pane until a menu appears, and then select **Paste**.

It is also possible to move an amino acid from one palette to another by dragging the amino acid to the desired palette. The amino acids are sorted in alphabetical order.

To edit an amino acid, open it by pressing it and modify the relevant parameters.

Resins, Protecting Groups, and Activators, Bases and Miscellaneous

In the **Resins, Protecting Groups, and Activators, Bases and Miscellaneous** views, you can add, copy, edit, and delete resins, protecting groups, and reagents.

To add a new resin, protecting group, or reagent, press **New** in the corresponding view and add data to the relevant text boxes.

A resin, protecting group, or reagent can be copied or deleted by pressing and holding on it until a menu appears and then selecting **Copy** or **Delete**. To paste, press and hold an area in the list until a menu appears, and then select **Paste**. The lists are sorted in alphabetical order.

To edit a resin, protecting group or reagent, open it by pressing it and modify the relevant parameters.

Figure 35. View to edit a reagent in the Activators, Bases and Miscellaneous view.

Reagents that can be used for performing UV monitored Fmoc deprotections, on a system equipped with a UV detector, are locked (🔒) and cannot be modified or deleted.

Reports

In the **Reports** view, you can search, view, and delete the results of peptide syntheses. It is also possible to save reports as PDF files on a USB memory device.

For information on the data that is displayed for a UV monitored Fmoc deprotection, see “Results” on page 8.

Maintenance

In the **Maintenance** view (see Figure 36), procedures are available for troubleshooting and performing maintenance on the system, such as cleaning the liquid path, priming the system, washing the needles, emptying the reactor vial or wash station, and moving the robot arm, etc. In running mode, only a limited number of the procedures are available and they can only be accessed when the synthesis is paused.

Note that it is not possible to add, edit, or delete procedures in the **Maintenance** view.

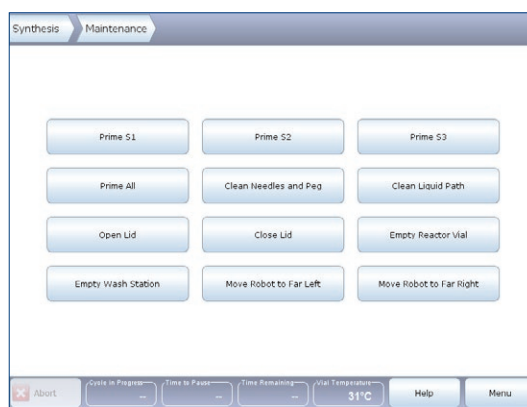


Figure 36. The Maintenance view with procedures for troubleshooting and performing maintenance on the system.

More Information

For more information and instructions, please see “Quick Start” on page 1 and the system’s online **Help**.

Switch Between Peptide and Organic Synthesis

Switch from Peptide to Organic Synthesis

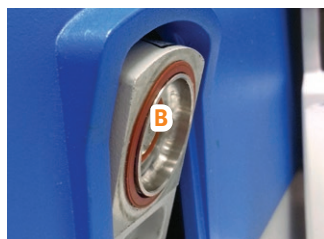
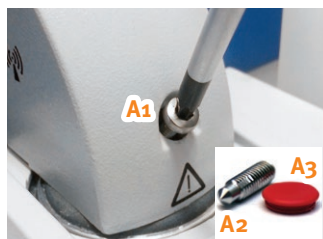
Warning:

- Failure to follow these instructions may result in personal injury and/or equipment damage.
- Ensure that the power cord, cables, and tubing connected to the system cannot come in contact with water or chemicals.

To be able to perform organic synthesis on an Initiator+ Alstra system, follow the procedure shown below.

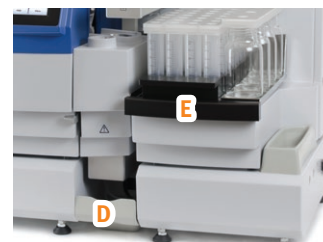
The following items supplied with the system are required: The T20 Torx and 2 mm hex screwdrivers, vent screw and plug, and the waste tray and cavity cover used for organic synthesis. You also need a pair of long tweezers.

1. Clean the liquid path as described in “Monthly Cleaning of the Liquid Path” on page 21.
2. Empty the system of liquid by priming with air:
 - a. Remove the solvent inlets from their bottles.
 - b. Ensure that the vacuum is switched on.
 - c. Press **Menu** and select **Maintenance** in the appearing menu.
 - d. Start the prime by pressing **Prime All**.
3. Replace the M4x16 screw on the cavity lid with the vent screw:
 - a. Press **Close Lid** and then **Move Robot to Far Right**. Wait until the robot arm is located to the right of the rack tray.
 - b. Remove the M4x16 screw (A1) located on the cavity lid using the T20 Torx screwdriver.
 - c. Insert and tighten the vent screw (A2) using the 2 mm hex screwdriver.
 - d. Cover the hole using the red plug (A3).
 - e. Press **Open Lid** and check that the pressure plate (B) is aligned with the cavity lid according to the image below.



4. Turn off the vacuum pump or, if your system is equipped with an external vacuum system, close the valve.
5. Release the pressure by pressing **Empty Wash Station** (in the **Maintenance** view) repeatedly until equilibrium is achieved.

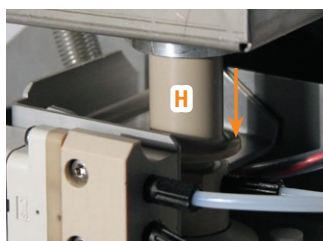
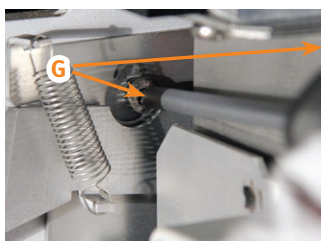
6. Press **Menu** and select **Main Menu** in the appearing menu.
7. In the main menu, press **Organic Synthesis**.
8. Read and confirm dialogs that open.
9. In the main menu, press **Shut Down**.
10. Read and confirm dialogs that open.
11. When the message “Safe to power off” appears on the screen, turn off the system and disconnect the power cord.
12. Remove the cavity cover (C) by lifting it and then pulling it toward you.
13. Remove and empty the waste tray (D) located underneath the microwave cavity.
14. Remove the rack tray (E) with racks and bottles.



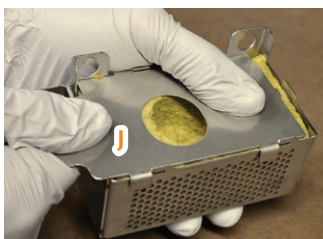
15. Remove the oscillating mixing unit using the T20 Torx screwdriver:
 - a. Disconnect the cable to the mixing unit from the **VORTEX** port at the rear of the Alstra module (see “Connections” on page 20).
 - b. Disconnect the drain tube from the **Vial** port below the microwave cavity (see “Connections” on page 20).
 - c. Disconnect the UV detector tube from the connection on the front of the Alstra module (see “Connections” on page 20).
 - d. Remove the vial eject tube (F) by pressing the eject lever and using a pair of tweezers.
 - e. Slightly loosen the two screws (G) holding the mixer unit to the cavity wall.
 - f. Unhook the mixer unit from the loosened screws (G) and then remove it by carefully lowering it straight down.

Note: Do not bend the mixer motor (H) as this will damage it.

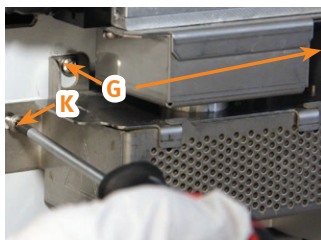




16. Disconnect the communication cables from the **PUMP** and **ROB** ports at the rear of the system (see “Connections” on page 20).
17. Mount the waste tray used for organic synthesis on the cavity wall using the T20 Torx screwdriver:
 - a. Ensure that the waste tray has a clean and intact waste tray insert (I) and is closed properly using the waste lid (J).
 - b. Hang the waste tray on the two screws (G) on the cavity wall.
 - c. Fasten the screw supplied with the waste tray in the lower left screw hole (K) and tighten the two upper screws (G).



18. Ensure that a pressurized air supply, > 60 l/min (2.1 cubic feet/min), 2.5 to 4.0 bar (0.25 to 0.40 MPa; 36 to 58 PSI), is connected to the pressurized air inlet at the rear of the system (above the power inlet on the Initiator+ unit).
19. Remove only the upper screw of the two screws holding the wash station, using the T20 Torx screwdriver. This allows the cavity cover to fit properly; see step 20.
20. Gently press the wash station back 5 mm toward the robot and place the cavity cover (L) used for organic synthesis in its position.



21. Connect the power cord and turn on the system.
22. Perform a reference run as described in the getting started guide for organic synthesis (P/N 355975).

Switch from Organic to Peptide Synthesis

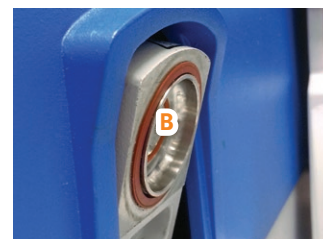
Warning:

- Failure to follow these instructions may result in personal injury and/or equipment damage.
- Ensure that the power cord, cables, and tubing connected to the system cannot come in contact with water or chemicals.

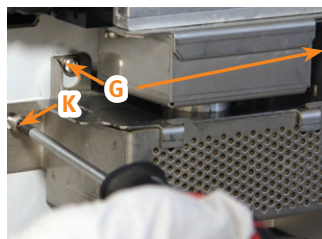
To switch back from organic to peptide synthesis, follow the procedure shown below.

The following items supplied with the system are required: The T20 Torx and 2 mm hex screwdrivers, M4x16 screw, oscillating mixing unit, vial eject tube, wash station screw, rack tray, and the waste tray and cavity cover used for peptide synthesis. You also need a small flat-blade and a T10 Torx screwdriver.

1. Replace the vent screw on the cavity lid with the M4x16 screw:
 - a. Press **Menu** and select **Maintenance** in the appearing menu.
 - b. Press **Close Lid** and then **Move Robot to Far Right**. Wait until the robot arm is located to the right of the rack tray.
 - c. Remove the red plug (A3) located on the cavity lid using a small flat-blade screwdriver.
 - d. Remove the vent screw behind the red plug, using the 2 mm hexagon spanner.
 - e. Insert and tighten the M4x16 screw using the T20 Torx screwdriver.
 - f. Press **Open Lid** and check that the pressure plate (B) is aligned with the cavity lid according to image below.



2. Press **Main Menu** in the right-hand panel.
3. In the main menu, press **Peptide Synthesis**.
4. Read and confirm dialogs that open.
5. In the main menu, press **Shut Down**.
6. Read and confirm dialogs that open.
7. When the message “Safe to power off” appears on the screen, turn off the system and disconnect the power cord.
8. Remove the cavity cover (L) by lifting it and then pulling it toward you.
9. Remove the waste tray using the T20 Torx screwdriver; remove the lower left screw (K), slightly loosen the two upper screws (G), and unhook the waste tray.
10. Connect the communication cables to the **PUMP** and **ROB** ports at the rear of the system (see “Connections” on page 20).



11. Mount the oscillating mixing unit using the T20 Torx screwdriver:

- Carefully lift the mixing unit straight up into its position in the microwave cavity and hang it on the two upper screws (G) on the cavity wall.

Note: Do not bend the mixer motor (H) as this will damage it.

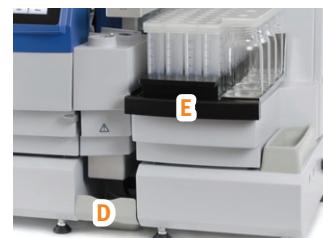
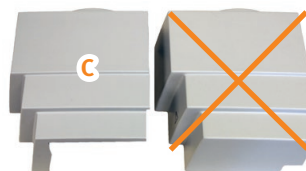
- Fasten the two upper screws (G).
- Connect the drain tube ($\varnothing = 1/8"$) to the **Vial** port below the cavity (see "Connections" on page 20).
- Connect the cable to the mixing unit to the **VORTEX** port at the rear of the Alstra module (see page 20).
- Connect the UV detector tube ($\varnothing = 1/16"$) to the connection on the front of the Alstra module (see page 20).

14. Fasten the upper of the two screws holding the wash station, using the T20 Torx screwdriver.

15. Place the cavity cover used for peptide synthesis (C) in its position.

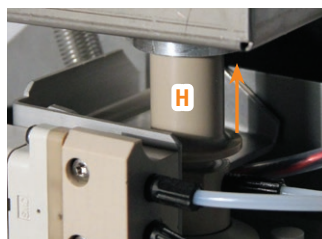
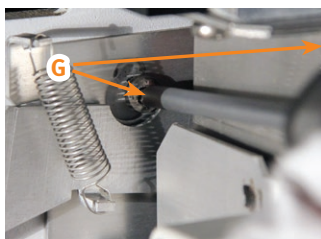
16. Place the waste tray (D) in its position underneath the microwave cavity.

17. Place the rack tray (E) in its position. Ensure it is positioned correctly.



18. Connect the power cord and turn on the system.

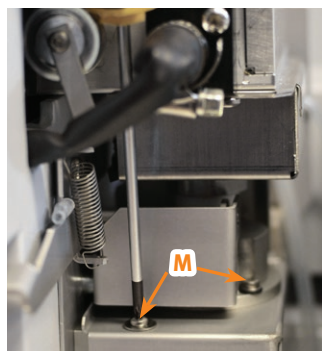
Note: If the position of the wash station should need to be recalibrated after the switch, log into the system mode, select the **Robot Calibration Wizard** tab, and follow the instructions that appear on the screen.



12. Insert the vial eject tube (F) into the microwave cavity with the larger opening downward.

13. Ensure that the vial eject tube (F) is centered in the microwave cavity. If not, calibrate the oscillating mixing unit:

- Slightly loosen the screws (M) holding the mixing unit to the microwave cavity, using a T10 Torx screwdriver.
- Center the vial eject tube (F) by moving the mixing unit.
- Fasten the screws holding the mixing unit.



Connections

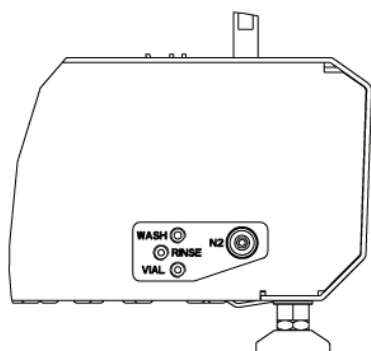


Figure 37. Connections on the front inside right of the Alstra module.

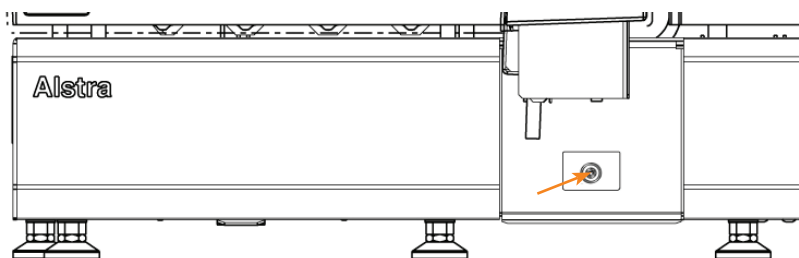


Figure 38. UV connection on the front of the Alstra module.

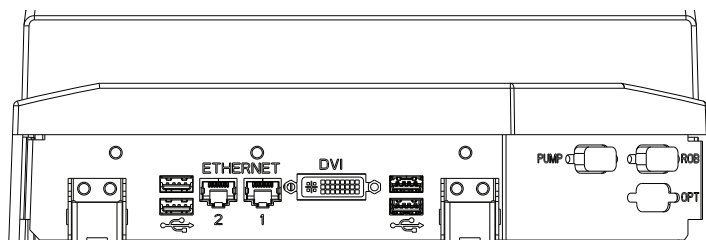


Figure 39. Connections at the rear of the system.

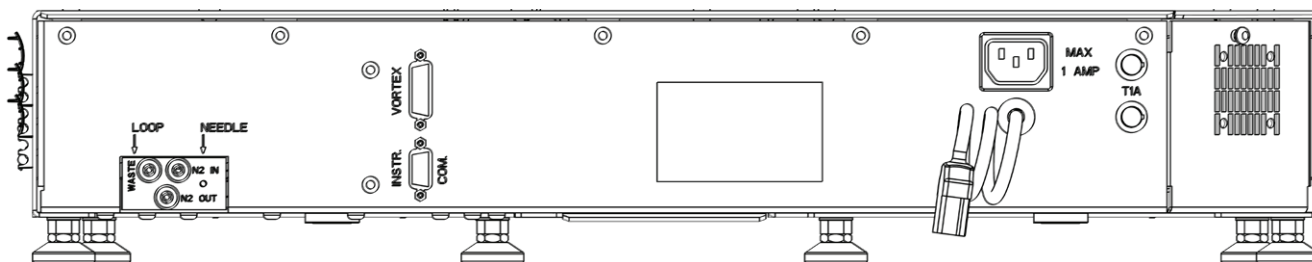


Figure 40. Connections at the rear of the Alstra module.

Maintenance

Weekly Cleaning of the Liquid Path

Notice: Handle chemical and liquid waste according to the Safety Data Sheets and to local/national guidelines on laboratory safety procedures.

We recommend that the liquid path (including waste path, valves, oscillating mixing unit, needles, and UV detector, if installed) is cleaned at least once every week.

Required: 25 mL of ethanol or similar, a reagent bottle, an empty 10 mL reactor vial, a 10 mL vial extension, a vial collar, and the vial loading tool.

1. Ensure that the vacuum is turned on.
2. Press **Menu** and select **Maintenance** in the appearing menu.
3. Ensure that the robot arm is located to the right of the rack tray. If you need to move the robot arm, press **Move Robot to Far Right**. Wait until the robot arm is in the correct position.
4. Ensure that a vial collar is inserted into the microwave cavity.
5. Insert an empty 10 mL reactor vial with the appropriate vial extension into the cavity using the vial loading tool.
6. If the system is equipped with a UV detector, fill a reagent bottle with at least 25 mL of ethanol or similar. Otherwise, fill it with at least 17 mL.
7. Place the reagent bottle in position **R4** on the rack tray.
8. Press **Clean Liquid Path**. Wait until the task is finished.
9. Remove the reagent bottle.
10. Place the vial loading tool over the reactor vial and then gently press the vial eject lever to release the vial; see Figure 14 on page 7.

Monthly Cleaning of the Liquid Path

Notice: Handle chemical and liquid waste according to the Safety Data Sheets and to local/national guidelines on laboratory safety procedures.

We recommend that the liquid path and S3 inlet is cleaned at least once every month and whenever the system is not going to be used for a period of time. This procedure is also to be used if the S3 inlet runs dry.

Required: 80 mL of ethanol or similar, a reagent bottle, an empty 10 mL reactor vial, a 10 mL vial extension, a vial collar, and the vial loading tool. If priming of inlet S2 is unnecessary (see step 5 below), then 55 mL of ethanol or similar is adequate.

1. Perform the clean liquid path procedure as described in “Weekly Cleaning of the Liquid Path”.
2. Ensure that the vacuum is turned on.

3. Press **Menu** and select **Inlet Configuration** in the appearing menu.
4. Press **S2**.
5. If ethanol or similar is already assigned to inlet S2, verify that at least 5 mL is available, press **Cancel**, and proceed to step 11 below.
6. Assign ethanol or similar to inlet S2 and enter at least 30 mL as the available volume.
7. Press **Save** and then **Go to Maintenance** in the appearing dialog.
8. Fill the bottle assigned to inlet S2 with the correct volume of liquid.
9. Press **Prime S2** and wait until the task is finished.
10. Press **Menu** and select **Inlet Configuration** in the appearing menu.
11. Press **S3**.
12. Assign ethanol or similar to inlet S3 and enter at least 25 mL as the available volume.
13. Press **Save** and then **Go to Maintenance** in the appearing dialog.
14. Fill the bottle assigned to inlet S3 with the correct volume of liquid.
15. Press **Prime S3** and wait until the task is finished.

Clean the Drain Valves

Notice: Handle chemical and liquid waste according to the Safety Data Sheets and to local/national guidelines on laboratory safety procedures.

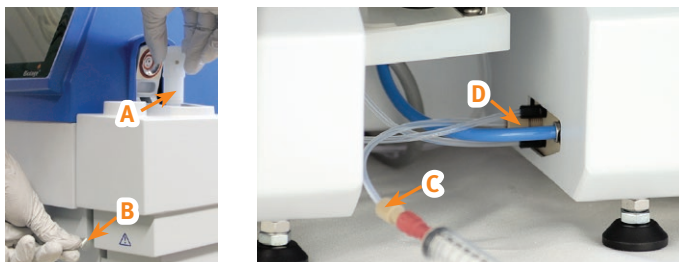
If the wash station or a reactor vial inside the microwave cavity is not emptied properly or is constantly drained, the problem may be resolved by cleaning the drain valves (by flooding them).

Note: If the cleaning is not performed according to the instructions below, the drain valves may break.

Required: 20 mL of ethanol or similar, the vial loading tool, and the injection maintenance kit (syringe and injection tube, P/N 411888) supplied with the system.

1. If the system is processing and cleaning of the drain valves is required, press **Pause...** and select the appropriate pause option in the appearing menu (**Pause After Current Operation**, **Pause After Next Deprotection**, or **Pause After Next Coupling**).
2. Press **Menu** and select **Maintenance** in the appearing menu.
3. Press **Move Robot to Far Right**. Wait until the robot arm is located to the right of the rack tray.

- Remove any vial located in the microwave cavity by placing the vial loading tool (A) over the vial and then gently pressing the vial eject lever (B) to release the vial.
- Remove and empty the waste tray located below the microwave cavity.
- Fasten the injection tube (C) to the **Rinse** port (D) below the microwave cavity.



- Put the waste tray back in place.
- Fill the syringe with 10 mL of ethanol or similar and connect it to the injection tube.
- Log into the software's system mode:
 - If in peptide synthesis mode, press **Menu** and select **Main Menu** in the appearing menu.
 - Press **System** in the main menu. All user accounts with system owner privilege are listed in the **Select User** dialog.
 - Select your user account and press **OK**. If you do not have an account with system owner privilege, please contact your system supervisor.
 - If your account is password-protected, the **Input Password** dialog opens. Enter your password and press **OK**.
- Select the **Peptide Synthesis** tab.
- Press to close the lid.

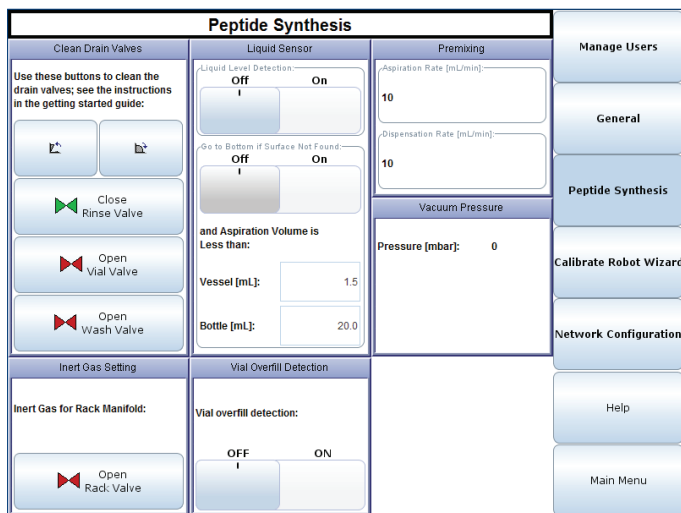


Figure 41. The Peptide Synthesis tab in system mode.

- Press **Open Vial Valve**.
- Inject the syringe contents.

Note: The syringe contents will be pushed through the vial valve and into the microwave cavity. The cavity will be flooded and then drained into the waste tray below the cavity.
- If necessary, empty the waste tray.
- Press to open the lid.
- Press **Close Rinse Valve** and **Close Vial Valve**.
- Remove the syringe from the injection tube and fill it with 10 mL of ethanol or similar.
- Reconnect the syringe to the injection tube.
- Press **Open Rinse Valve** and **Open Wash Valve**.
- Inject 5 mL of the syringe contents.
- Ensure that the vacuum is turned on for the rest of the steps.
- Press **Close Rinse Valve** to empty the wash station.
- When the wash station has been emptied, press **Open Rinse Valve**.
- Inject the rest of the syringe contents.
- Press **Close Rinse Valve** to empty the wash station.
- When the wash station has been emptied, press **Close Wash Valve** and **Open Rinse Valve**.
- Remove and empty the waste tray.
- Remove the syringe and the injection tube.
- Put the waste tray back in place.
- If necessary, clean the microwave cavity as described on page 23.

Clean the Exterior of the System

Warning:

- Ensure that the system is turned off and the power cord is disconnected before cleaning.

If the touch screen has been contaminated by chemicals, it must be cleaned immediately.

- Shut down the system:
 - If in peptide synthesis mode, press **Menu** and select **Main Menu** in the appearing menu.
 - If in system mode, press **Main Menu**.
 - In the main menu, press **Shut Down**.
- When the message "Safe to power off" appears on the screen, turn off the system and disconnect the power cord.
- Clean the touch screen and the exterior of the system, using a soft and clean cloth. The cloth can be dry or lightly dampened with a neutral detergent or alcohol.
- When the system has been cleaned, connect the power cord and turn on the system.

Clean the Microwave Cavity, IR-Sensor, and Oscillating Mixing Unit

Warning:

- Do not attempt to operate the system if the microwave cavity contains trapped objects or moisture. There is a risk of damage to the system and microwave leakage.
- Ensure that the cavity cover, cavity lid seals, and oscillating mixing unit are in position when the system is processing.

Notice: Handle chemical and liquid waste according to the Safety Data Sheets and to local/national guidelines on laboratory safety procedures.

The microwave cavity, IR-sensor, and oscillating mixing unit must be cleaned after the occurrence of a vial leakage.

Required: The T20 Torx screwdriver supplied with the system, the vial loading tool, a pair of flat-nosed pliers, a soft lens cleaning tissue (or similar), cotton swabs, soft and clean cloths, an emery cloth, pressurized air, water and/or alcohol, and ethanol or similar. The cleaning solution is dependent on the residues inside the cavity.

Note: If the cavity air guide and/or the cavity lid seals are broken or distorted, they have to be replaced.

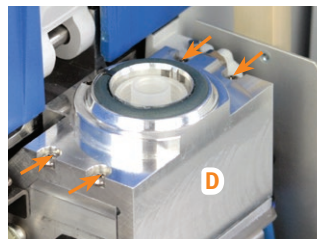
I. Shut down the system and disconnect the power cord:

- If the system is processing and you need to clean the microwave cavity at once, press **Abort** in the bottom pane and then **Yes** in the dialog that opens to abort the task in progress.
- In the **Synthesis Aborted** dialog, press **OK** to move the robot arm to the wash station.
- Press **Menu** and select **Main Menu** in the appearing menu.
- Press **Shut Down** in the main menu.
- When the message “Safe to power off” appears on the screen, turn off the system and disconnect the power cord.

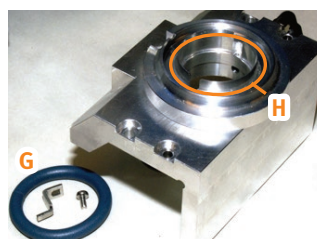
II. Clean the microwave cavity, IR-sensor, and mixing unit:

- Remove any reactor vial located in the microwave cavity; place the vial loading tool (A) over the vial and then gently press the vial eject lever (B) to release the vial.
- Remove the cavity cover (C) by lifting it and then pulling it toward you.

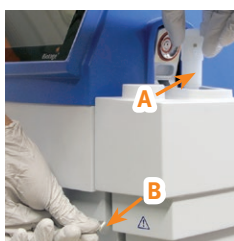
- Remove the service lid (D) by removing the four screws and disconnecting the inert gas tubing (E) by pushing in the collar (F) against the fitting and pulling the tubing out.



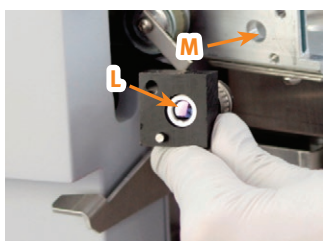
- Remove the cavity lid seal (G) by removing the screw and carefully pulling out the lid seal.
- Clean the cavity lid seal (G) with ethanol or mild soapy water. Do not use aromatic or chlorinated solvents.
Note: If the cavity lid seal is broken or distorted, it has to be replaced.
- Clean the service lid (D) using a cloth lightly dampened with ethanol or mild soapy water.
- Clean the seal slot (H) using an emery cloth.
- Ensure that the service lid (D) and all its parts are dry and that the two service lid seals (I), on the back of the service lid, are in place. If a seal is broken or distorted, contact Biotage® 1-Point Support™.
- Verify that the seal slot (H) is dry and then put the cavity lid seal (G) back in place.
Note: Do not tighten the screw too hard.



- Remove the cavity air guide (J) and clean it using a cloth. If the cavity air guide is broken or distorted, replace it.
- Remove the burst wall (K) and clean it using a cloth. If the burst wall is broken or distorted, replace it.



17. Remove the IR-sensor (L) from the microwave cavity by removing the screw.
18. Clean the IR-sensor (L) using a soft lens cleaning tissue (or similar) lightly dampened with distilled water, alcohol, or a camera lens cleaner. Do not scratch the surface! If the IR-sensor is broken, contact Biotage 1-Point Support.
19. If possible, vacuum the microwave cavity. Otherwise remove as much as possible of the spill with a soft and clean cloth.
20. Clean and dry the cavity, including the IR-housing (M), using pressurized air, a cloth, and cotton swabs.
21. Ensure that all parts are dry and that the two service lid seals (N), on the side of the cavity wall, are in place. If a seal is broken or distorted, contact Biotage 1-Point Support.



22. Reassemble the IR-sensor, burst wall, cavity air guide, service lid, and inert gas tubing.
Note: Ensure to insert the cavity air guide correctly with the hole facing the IR-housing.
23. Clean the oscillating mixing unit by gently spraying ethanol or similar from a spray can into the microwave cavity; see the image to the left below. Waste will collect in the waste tray (O) below the cavity.
24. If necessary, gently spray the area below the oscillating mixing unit.
25. Empty and dry the waste tray (O), and put it back in place.



26. If the inside of the cavity lid needs to be cleaned:
 - a. Connect the power cord and turn on the system.
 - b. In the main menu, press **Peptide Synthesis**.
 - c. Press **Menu** and select **Maintenance** in the appearing menu.
 - d. Press **Close Lid** and then **Move Robot to Far Right**. Wait until the robot arm is located to the right of the rack tray.

- e. Remove the M4x16 screw located on the cavity lid using the T20 Torx screwdriver.
- f. Press **Open Lid** and gently remove the pressure plate (P) from the lid using a pair of flat-nosed pliers.
- g. Clean the lid and pressure plate using a cloth lightly dampened with a solvent suitable for the residues. If the cavity lid seal (Q) is broken or distorted, replace it.
- h. Put the pressure plate back in place. Ensure that the plate is positioned correctly.
- i. Press **Close Lid** and put the M4x16 screw back in place.
- j. Press **Open Lid** and check that the pressure plate (P) is aligned with the cavity lid according to the image to the right below.



IV. Put the cavity cover back in place:

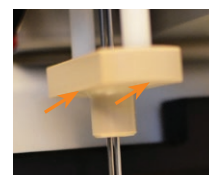
27. Put the cavity cover (C) back in place.
28. If applicable, connect the power cord and turn on the system.

Clean the Needle Nose Assembly

The needle nose assembly, located at the bottom of the vertical robot arm, may need cleaning.

Dip the tip of the needle nose assembly in a container containing a cleaning solution appropriate for the residues and dry with a dry cloth.

If necessary, you can remove the needle nose assembly by removing the two screws (see the image) and then clean it. Wipe it dry with a dry cloth and put it back in place.



Clean the Accessories

When necessary, clean the rack tray, racks, cover plates, vial extensions, and vial loading tool using a laboratory dishwasher with an alkaline detergent.

Note: Clean the cover plates using a dish washer program for plastic with a maximum cleaning temperature of 60°C and no drying. Empty the cover plates of liquid using inert gas or pressurized air.

To clean without using a laboratory dishwasher, use soap, water and/or acetone. Note that cover plates should not be dried in temperatures above 75°C.

Release the Pressure

Before emptying or exchanging a scrubber bottle or waste reservoir, it is important to release the pressure. See also “Empty the Waste and Scrubber Kit” below.

1. Turn off the vacuum pump or, if your system is equipped with an external vacuum system, close the valve.
2. Press **Menu** and select **Maintenance** in the appearing menu.
3. Press **Empty Wash Station** repeatedly until no pressure remains.

Empty the Waste and Scrubber Kit

The 500 mL scrubber bottle needs to be emptied and replenished after each synthesis. We recommend that no more than 100 mL of scrubber solution is used. The 10 liter waste reservoir needs to be emptied when it is full.

Before emptying, remember to turn off the vacuum and release the pressure as described in “Release the Pressure” above.

Replace the Fuses

Warning:

- Ensure that the system is turned off and the power cord is disconnected before replacing fuses.
- Use only correct replacement fuses supplied by Biotage. Incorrect fuses create a potential fire hazard. See the labels at the rear of the system.

Required: Clean, non-abrasive, dry cloths, and ethanol.

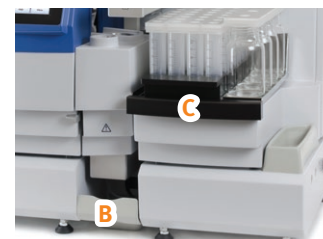
1. Turn off the system and disconnect the power cord.
2. Remove the fuse holders at the rear of the system; two fuses (T8A) are available on the Initiator+ unit and two (T1A) on the Alstra module.
3. Clean the new fuses using a cloth lightly dampened with ethanol and wipe them dry with a dry cloth.
Note: Do not touch the metal surfaces with your hands after the fuses have been cleaned.
4. Replace the blown fuse(s) and put the fuse holders back in place.

Replace the UV-Inlet Tube

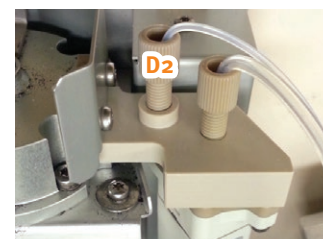
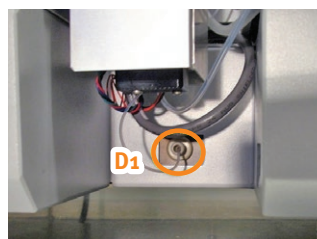
If a reactor vial inside the microwave cavity is not emptied properly when performing a UV monitored Fmoc deprotection, the UV-inlet tube is probably clogged. Try to clean it by cleaning the liquid path; see “Weekly Cleaning of the Liquid Path” on page 21. If cleaning does not solve the problem, replace the UV-inlet tube.

Required: The T20 Torx screwdriver supplied with the system and a new UV-inlet tube (P/N 356500SP).

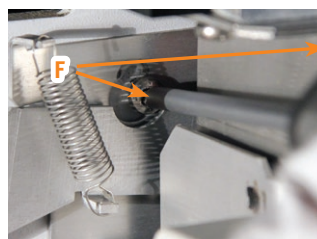
1. Ensure that the reactor vial is empty. If not, press **Menu**, select **Maintenance**, and press **Empty Reactor Vial**.
2. Remove the cavity cover (A) by lifting it and then pulling it toward you.
3. Remove and empty the waste tray (B) located underneath the microwave cavity.
4. Remove the rack tray (C) with racks and bottles.



5. Collect the liquid in the UV-inlet tube (D1) by disconnecting the tube from the connection on the front of the Alstra module and inserting it into an empty container.



6. Disconnect the other end of the UV-inlet tube (D2) from the mixer unit and replace it with a new tube. If it is necessary to unhook the mixer unit:
 - a. Remove the vial eject tube (E) by pressing the eject lever and using a pair of tweezers.
 - b. Slightly loosen the two screws (F) holding the mixer unit to the cavity wall using the T20 Torx screwdriver.
 - c. Unhook the mixer unit from the loosened screws (F) and then remove it by carefully lowering it straight down.



Note: Do not bend the mixer motor (G) as this will damage it.

- d. Disconnect the UV-inlet tube (D2) from the mixer unit and replace it with a new tube.
 - e. Carefully lift the mixing unit straight up into its position in the microwave cavity and hang it on the two upper screws (F) on the cavity wall.
Note: Do not bend the mixer motor (G) as this will damage it.
 - f. Insert the vial eject tube (E) into the microwave cavity with the larger opening downward. Ensure that it is centered in the microwave cavity. If not, calibrate as described in step 13 on page 19.
 - g. Fasten the two screws (F) holding the mixing unit.
- 7. Connect the other end of the new UV-inlet tube to the connection on the front of the Alstra module (D1).
 - 8. Put the cavity cover (A), waste tray (B), and rack tray (C) back in their positions.

General Information

Consumables and Accessories







Only genuine Biotage consumables and accessories must be used in the system. To order consumables and accessories, see contact information on the back of this document or visit our website www.biotage.com.

Consumables


Part No.	Product
356208	Inert gas manifold (1/pk)
356288	5 mL Reactor vial with PTFE frit, (50/pk)
356289	10 mL Reactor vial with PTFE frit (50/pk)
356290	30 mL Reactor vial with PTFE frit (50/pk)
356291	5 mL Reactor vial extension (5/pk)
356221	10 mL Reactor vial extension (5/pk)
356222	30 mL Reactor vial extension (5/pk)
356239	10 mL Amino acid vessels (100/pk)
356240	30 mL Amino acid vessels (100/pk)
356241	50 mL Amino acid vessels, (100/pk)
356162	20 x 10 mL Amino acid rack
356167	24 x 30 mL Amino acid rack
356193	32 x 30 mL Amino acid rack
356198	28 x 50 mL Amino acid rack
356163	20 x 10 mL Rack cover plate
356168	24 x 30 mL Rack cover plate
356194	32 x 30 mL Rack cover plate
356199	28 x 50 mL Rack cover plate
356158	R1-4 Reagent bottle cover plate
356203	R5 Reagent bottle cover plate
356166	20 x 10 mL Foil septa, (5/pk)
356192	24 x 30 mL Foil septa, (5/pk)
356197	32 x 30 mL Foil septa, (5/pk)
356202	28 x 50 mL Foil septa, (5/pk)
356161	R1-4 Reagent bottle foil septa, (5/pk)
356206	R5 Reagent bottle foil septa, (5/pk)
356252	5 mL/10 mL Reactor vial caps, (50/pk)
356253	30 mL Reactor vial caps, (50/pk)
356292	Reactor vial plugs, (50/pk)
356330	Vacuum pump ME1C, 100-230 VAC, 50-60 Hz
356254	185 mL Reagent bottle
356041	Vial loading tool
411888	Injection maintenance kit
356385	Vial collar for 5 and 10 mL reactor vials

Accessories

Accessories that may be necessary for the “Maintenance” section are listed below.

Part No.	Description
354180SP	 Lower cavity lid seal, qty 1
354878SP	 Vent screw replacement, qty 1
354974SP	 Cavity air guide, qty 1
354973SP	 Burst wall, qty 1
355366SP	 Waste tray inserts, qty 5
355723SP	 Upper cavity lid seal, qty 1
356500SP	 UV-inlet tube, qty 1
413246SP	 Fuse T1A, 6.3x32 mm, qty 2
411458SP	 Fuse T8A, 6.3x32 mm, qty 2

Manufacturer

 Biotage Sweden AB

Contact Us

Biotage Sweden AB

Box 8
SE-751 03 Uppsala
SWEDEN

Visiting address: Vimpelgatan 5

Phone: +46 18 56 59 00

Fax: +46 18 59 19 22

E-mail: info@biotage.com

Website: www.biotage.com

Please contact your local Biotage representative. See contact information on the back of this document or visit our website www.biotage.com.

Notes

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EUROPE

Main Office: +46 18 565900
Toll Free: +800 18 565710
Fax: +46 18 591922
Order Tel: +46 18 565710
Order Fax: +46 18 565705
order@biotage.com
Support Tel: +46 18 56 59 11
Support Fax: + 46 18 56 57 11
eu-1-pointsupport@biotage.com

NORTH & LATIN AMERICA

Main Office: +1 704 654 4900
Toll Free: +1 800 446 4752
Fax: +1 704 654 4917
Order Tel: +1 704 654 4900
Order Fax: +1 434 296 8217
ordermailbox@biotage.com
Support Tel: +1 800 446 4752
Outside US: +1 704 654 4900
us-1-pointsupport@biotage.com

JAPAN

Tel: +81 3 5627 3123
Fax: +81 3 5627 3121
jp_order@biotage.com
jp-1-pointsupport@biotage.com

CHINA

Tel: +86 21 2898 6655
Fax: +86 21 2898 6153
cn_order@biotage.com
cn-1-pointsupport@biotage.com

To locate a distributor,
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