

GO-Blot V2 User Manual

v3.9.1



GO-Blot V2 website: <https://mygoblot.com>

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Safety Precautions

Warning! Power Rating. GO-Blot's power cord must be connected to a power receptacle (socket) that provides voltage and current within the specified rating for the system (110-240 Volts / 30W / 250 milli-Amp). Use of an incompatible power receptacle may produce electrical shock and fire hazards. Do not use near constant moisture or standing liquids. Do not use non-grounded plug socket for power adapter connection to wall socket.

Warning! Internal Voltage. Always turn off power switch and unplug power cord before cleaning the outer surface of the equipment. The symbol === denotes Direct Current. If external grounding is required, connect an insulated wire from the aluminum base plate (underside) to the external ground. Do not use non-grounded plug socket for power adapter connection to wall socket.

Warning! Injury to body parts. The GO-Blot device contains moving cam and Reaction tray parts, avoid placing fingers, hands and other body parts into these areas.

Warning! Biohazards. Do not use biohazards with this device. Always wear safety glasses, gloves and a lab coat when working with biological reagents.

Warning! Liquids. Avoid spilling liquids on the device; fluid seepage into internal components creates the potential for short circuit and shock hazards. Wipe up all spills immediately and let the machine dry before further use. There are no hazardous gases or liquids in the device per se.

Warning! Mechanical Interruptions. In case of a mechanical interruption the device will continue running but the results might not be usual. There is a possibility of hazardous gases or liquids emanating from the device if it is intentionally or unintentionally halted during operation.

Warning! Unspecified use. Failure to use the device, parts/accessories supplied by the manufacturer or the recommended buffers and solutions in the manner described in this manual could result in hazardous conditions. Do not use the device in hazardous environments such as fume hoods. Do not use non-recommended liquids such as ethanol or acetone which could cause melting of plastics and short circuit the electrical components.

Precautions

Caution: Service. If the machine is not operating as expected, first perform the Quality Check in the troubleshooting section and if it still does not work properly, then call Cytoskeleton Inc. for technical advice. Repairs should only be performed by a Cytoskeleton technician or an authorized representative. Do not unscrew the base plate retaining screws or attempt any other maintenance or repairs, apart from the Water Wash routine or the Quality Check. If other maintenance or repairs are attempted, this will void the warranty.

Safety testing by independent laboratory

This device has been tested for electromagnetic interference and safety by an independent lab, METLAB Inc. USA. In this regard it has passed EMF US FCC CFR 47 Ch.1 Pt 15 B, and UL 61010-1, CSA C22.2 61010-1, EN 61326-1, IEC 61010-1, IEC 61010-2-051:2015 and IEC 61010-2-081:2015 safety directives. GO-Blot is RoHS compliant, indicating the use of lead free components and connections.

Physical Specifications

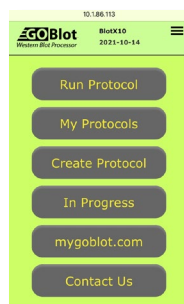
Size	16 cm wide x 27 cm deep x 25 cm high.
Weight	2.6 Kg
Electrical rating	Power adapter, 100-240VAC 0.5 Amps. 50-60Hz (converts to 12VDC 3.34Amps). GOBlot device requires 12 VDC 1 Amp with a barrel connector with negative external and positive internal connectors.
Reproducibility	+/- 10% western blot band intensity.
Accuracy	+/- 10% volume dispensing.
Operating temperature range	4 to 42°C.
Operating humidity range	10 to 100% RH.
Operating altitude	0 to 2000 meters with respect to sea level.
Operating pressure range	0.8 - 1.1 Atm. (79 to 108 kPa).
Maximum pressure inside device	10 PSI.
Normal operating pressure	<5 PSI.

Introduction

A Western blotting device has never been so flexible and easy to use as the new GO-Blot™ V2. There are three tray sizes to choose from and infinite (nearly) combinations of Protocols to create. In addition, there is a growing community of users that share their successful protocol/antibody combinations on the Global Western Portal™.

Phone interface

Choose from 10 Pre-defined Protocols or try out the new phone interface to create your own protocol.



Website

Visit mygloblot.com™ to set up your user account, and store your results to share with your lab or the global community.



For assistance:

User or Serial number assistance:

cserve@cytoskeleton.com

Protocol assistance:

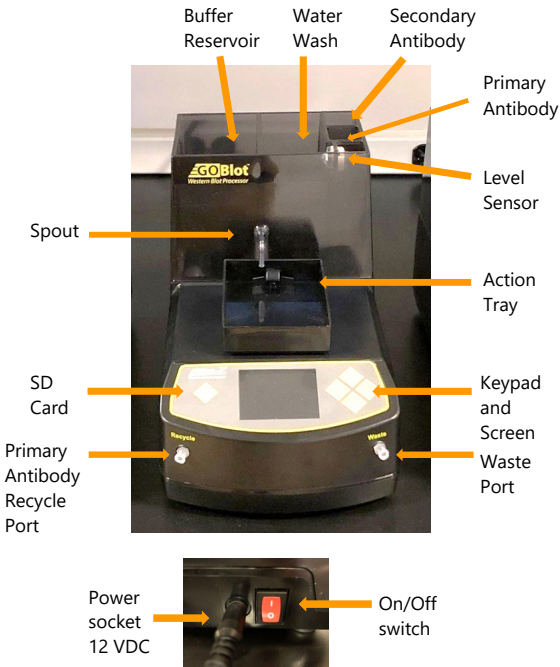
tservice@cytoskeleton.com

Telephone:

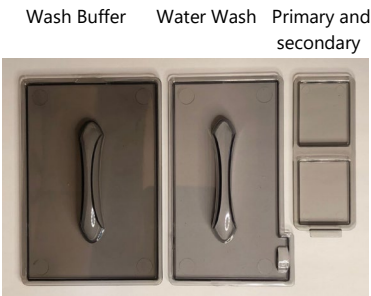
USA-303-322-2254

GO-Blot™ V2 Package Contents

Main assembly



Lids for buffer reservoirs



Small, medium and large trays



Nozzle on underside of trays



Small, medium and large
tray lids



Power adapter (110,120,220 or 240V)



Luer connectors and tubing



Syringe extractor



User Manual (not shown)

Set-up and Connectivity

Place the GO-Blot V2 device on a stable flat bench and check with a level that the lower surface of the unit is level in the x and y directions. Adjust level if necessary.

The Front Panel Layout

The Enter button is on the right side and is used to move to the next step and Run a protocol.



Attaching Action Tray

Chose a tray size and lightly smear silicone lubricant on the tray's nozzle.

Fit the tray as shown below,

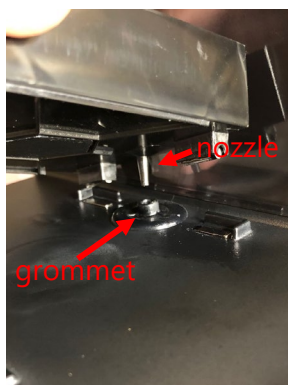
Step 1

Hook under spout



Step 2

Align nozzle with grommet's hole



Step 3

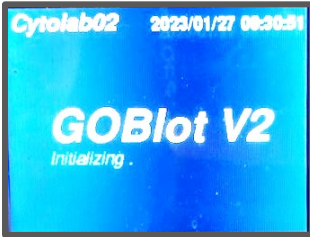
Push down very gently in the front middle.



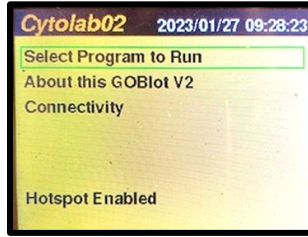
Powering up the GO-Blot V2

Turn on the power, at the back of the machine (Press “1” inwards = ON).

this screen should appear:



followed by this one:



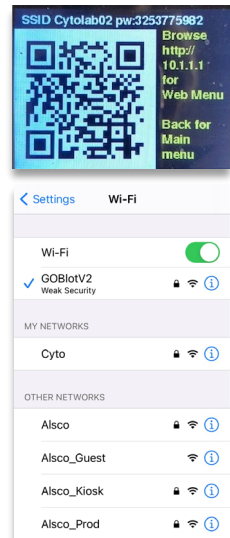
You may proceed directly to the Pre-defined Protocols at this stage by clicking selecting “Select Program to Run” with the Up and Down buttons, then press “Enter” to view the protocols. Turn to page 12 to Run a Protocol.

Note: The screen will switch between yellow/black theme to blue/white theme to preserve the screen’s quality during the lifetime of the device.

Setting up a direct connection from phone to GO-Blot V2

This connection method is the most simple and direct, it allows the user to quickly access the programming and protocol tracking functions from a mobile phone.

1. Click the “Down” button, to Connectivity.
2. Click “Enter” button.
3. Scan the QR code.
4. Choose GO-Blot V2 Wifi connection from your phone’s Settings>Wifi location. Make sure this is connected by seeing the check (tick) mark.
5. Go to the phone’s browser, and type in 10.1.1.1 and hit Enter or GO.
6. A green page with the GO-Blot V2 menu in green will pop up.
7. Tap Create Protocol to start programming the steps for your unique Western Protocol.



Setting up a network Wifi connection to access mygoblot.com

This method allows full functionality of mygoblot.com such as offsite tracking of protocol progress, user registration, access to archive runs, and ability to add data to the webpage and submit a protocol to the Global Western Archive.

1. Starting from the direct access mode described in the previous section, go to the home page of 10.1.1.1 IP address.
2. Click on Settings and then Server Setup.
3. Type in the following:
 - a) your network's Web name or Wep ID,
 - b) Web password,
 - c) a new IP address obtained from your IT department
 - d) the Gateway IP and Port # from your IT department.

Note1: This information will be obtained from your IT department.

Note2: Some phones have the Save button below the screen's view, in that case either shrink the screen and/or tap on AA symbol in the address bar, and chose "Hide address bar" option.

5. When the IP is confirmed, make a label and attach to the GO-Blot V2 in an obvious permanent place. Next time anyone wants to use the device, they can just type this number into their phone's web browser.
6. Click "Save" to complete the process.
7. Click on Date & Time button.
8. Enter your local zone details (e.g. GB and France are 00.00, New York is -05.00, San Francisco is -08.00, Eastern Australia is +10.00 and Beijing is +8.00 and Japan +09.00), then tap on month and year to change them, and click "Save". Note: Some phones have the Save button below the screen's view, in that case either shrink the screen and/or tap on AA symbol in the address bar, and chose "Hide address bar" option.
9. Close browser, and click "Back" on the GOBlot V2 device.
7. Open a browser on your phone, type in the IP and see the control panel in the browser.

Image of browser control panel:


Note: This control panel can also be pulled up on a local computer's browser by typing in the same IP address.



Tip: Create an icon for your phone by following these instructions:

iPhone

Go to your device's IP address in a browser

Click the  icon

Scroll down to Add to home screen

Name your Shortcut

Click Done and see this:



Android

Go to your device's IP address in a browser

Click and hold on the padlock in the address bar

Drag cursor to desktop and release and you get this:



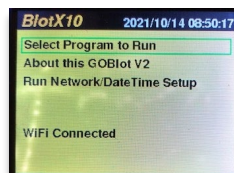
Tip: Search for a vacant IP address on your network or scan network for devices

1. Open a browser and perform a web search for "Download Advanced IP Scanner"
2. Download and install software.
3. Run the scan to see a list of IP addresses and their devices.

Pre-defined Protocols

Warning: do not load solutions until the Water Wash Protocol has run.

1. When in front of the GO-Blot V2 machine, click the "Back" button until you see the Main Menu, this screen:



2. Then scroll up or down, by clicking the "Up" or "Down" buttons to high-light the "Select Program to Run" line.

3. Click the "Enter" button.

4. Scroll down to the desired protocol and click Enter.

5. The protocol detail will show up.

Warning: do not load solutions until the Water Wash Protocol has run.

Table 1: List of Pre-defined Protocols

#	Block (min)	Block Wash (n x min)	Primary (min)	Primary Wash (n x min)	Secondary (min)	Secondary Wash (n x min)	Application
1	60	1 x 1	60	3 x 5	60	5 x 5	Short run for good antibodies
2	60	1 x 1	120	3 x 5	60	5 x 5	Medium run for good antibodies
3	60	1 x 1	240	3 x 5	60	5 x 5	Long run for good antibodies
4	60	1 x 1	720	3 x 5	60	5 x 5	Overnight run with good antibodies
5	60	1 x 1	120	3 x 5	60	6 x 10	Extensive washing
6	60	1 x 1	120	6 x 5	60	6 x 10	Extensive washing
7	60	1 x 1	240	6 x 5	60	6 x 10	Extensive washing
8	60	1 x 1	720	6 x 5	60	6 x 10	Extensive washing
9	60	1 x 1	60	5 x 5	0	0 x 0	Block and single labeled antibody
10	60	1 x 1	120	5 x 5	0	0 x 0	Block and single labeled antibody

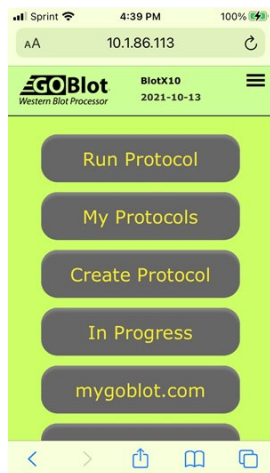
Creating a Protocol

Open a browser in a computer or phone.

Type in the IP address of the GO-Blot V2 into the address bar and tap Enter or Go respectively.

This screen should pop up:

Note: The direct access method uses IP 10.1.1.1, but make sure the QR code is scanned first in order to connect the device's wifi to your phone.



1. Tap on "Create Protocol" and the second screen below should pop up.

2. Fill out each field with appropriate data, e.g. the wash steps have two data points, the first one is number of washes, the second is the length of each wash in minutes.

Note: There is no setting to change wash volume because that is automatically determined by the Buffer Reservoir volume and the Tray size.

3. When it is complete, tap "Submit" to save the new Protocol.

Note: New Protocols are listed chronologically, therefore the latest protocol will be at the end of the list.

Note: The Large tray type will be limited automatically if there are too many washes, the maximum number of washes for the Large tray is 12 total.

Tip: If you cannot see the Submit button on the iPhone version, shrink the screen and/or tap on AA symbol in the address bar, and choose "Hide address bar" option.

Tip: Pause buttons are useful to pause the run while adding a labile reagent or checking a routine's timing.

Running a Protocol

Warning: do not load solutions until the Water Wash Protocol has run.

A Water Wash is necessary to start using your GO-Blot V2 and to ensure all systems are working well. A Water Wash is also recommended after greater than 1 week without a run. Sometimes the last operator might forget to clean the machine with a Water Wash, in this situation the Water Wash must be run before using the machine.

1. While standing next to the GO-Blot V2 check that no solutions are in any reservoirs or trays.
2. Connect the luer locks with tubing to the front of the machine.
3. Place the left and right-side tubing into a waste container.
4. While on the Main Menu, scroll down by clicking the machine's "Down" button until you reach the Run Protocols line.
5. Click the "Enter" button.
6. Scroll down to highlight the Water Wash, and click Enter.
7. An alert should pop-up indicating to fill the Water Wash reservoir, let's do this by adding 425 ml to the container which covers the level sensor (see p.5 for locating the parts).
8. The alert should disappear and the option to Run the protocol appears, click Enter to run the protocol.

Note: The same approach is used to run a protocol, except you need to add Blocking solution and membrane to the tray, TBST (or PBST) to the Buffer reservoir and appropriate Primary and Secondary antibody solutions to their respective receptacles. Also, place the left-side "Recycle" tubing into a clean recycle vessel (e.g. 15 or 50 ml disposable tube). Then chose a protocol from the list. Pre-defined Protocols are at the top of the list and User defined (with a "U" before the number) ones at the end of the list.

Warning: Be sure to visit the machine and view the screen before adding antibody and buffer solutions. If a previous operator did not run the Water Wash, then it will be required before continuing, an alert will be visible, any buffers and antibody solutions loaded at this point will be washed away!

Note: If there is a sticky-high affinity antibody left over from a previous run. The best way to clean the device is to add 50 ml of either 1 x SDS-running buffer from a PAGE system (e.g. a standard composition is 50 mM Tris-HCl pH 6.8, 100 mM NaCl, 0.1% SDS) to all the reservoirs. Then choose the Water Wash protocol to run the solution through the system. Then follow with two Water Washes using pure water only.

Table 2: Sizes and volumes of Wash Buffer for the different trays:

Tray / Blot name	Membrane size (H x W cm)	Buffer volume used for each wash (ml)	Total volume of Wash Buffer to add to the reservoir (ml)
Small	8 x 2.2	4	5 x total # washes
Medium	8 x 7.2	23	26 x total # washes
Large	9 x 14	44	48 x total # washes

Note: Wash buffer can be either TBST or PBST or other aqueous solution. Do not use organic solvents.

Table 3: Recommended volumes of Primary and Secondary antibodies.

Tray / Blot name	Primary antibody volume (ml)	Secondary antibody volume (ml)
Small	4 ideal	5
	3 to 5 range	
Medium	15 ideal	20
	10 to 20 range	
Large	36 ideal	40
	30 to 40 range	

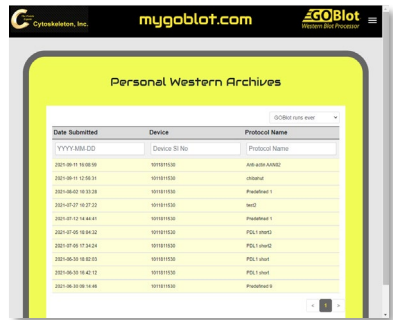
Using mygoblot.com

User and password

The original user will receive the login credentials by email. please contact tservice@cytoskeleton.com if you do not have access to it.

Retrieving data

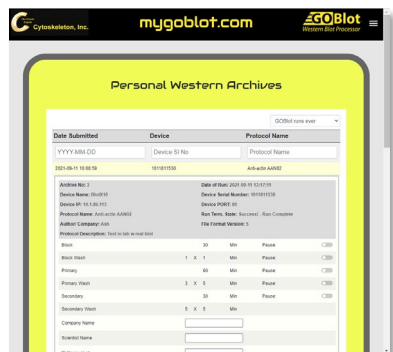
1. Logon to <https://mygoblot.com>
 2. Click on the fourth bar down called, "Personal Western Archive™"
 3. A list will pop-up as shown in the second screenshot.
 4. Click on the one you want to open
- Tip** – You can select the name or date range by the table's title drop down menus.
5. The third screenshot shows the run data.



Uploading results to complete an archive

Note: The empty boxes are ones you can fill in with experimental data and save, or save and submit for publication on the Global Western Portal.

1. Click in a box to start adding text.
2. Click on a yellow + sign to add a hyperlink
3. Click on upload files to add your experimental blot image.



4. Click Save to save data.

Note: Partially filled out fields are fine to save, but empty field files cannot be submitted to the Global Western Portal.

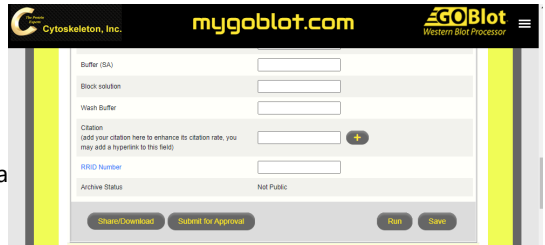
Submitting a Routine to the Global Western Portal™

1. Fill out all fields of a Personal Western Archive.

2. Click Save to save content.

3. Scroll to the end of the file and locate the “Submit for Approval” button, and click it.

4. With two days you will receive a notification of approval or additional questions before approval.

A screenshot of the mygloblot.com website. The header includes the Cytoskeleton, Inc. logo, the mygloblot.com URL, and the GOBlot Western Blot Processor logo. The main content area is a form with fields for Buffer (SA), Block solution, Wash Buffer, Clatlon (with a note to add clatlon rate and a plus icon), and RRD Number. Below these is an Archive Status field set to 'Not Public'. At the bottom of the form are four buttons: 'Share/Download', 'Submit for Approval', 'Run', and 'Save'.

Downloading a Routine from Global Western Portal™

1. Go to the homepage of <https://mygloblot.com>

2. Click on Global Western Portal.

3. Using the table's title search field, e.g. type in the Antigen of interest.

4. A short list of matches will pop-up. Click on one row.

5. Scroll to the bottom of the record, and click the “Share/Download” button.

6. Chose one of the options and click on it, a “json” file will download.

7. Receiving on the phone requires opening the email app and “sending” the json file to the Downloads folder.

Importing a file to a GO-Blot V2 device.

1. Open the GO-Blot V2 device control panel in a browser (see Section 4 above).

2. Click Create Protocol.

3. Scroll down to the “Import file” button.

4. Select the file from your download folder.

A screenshot of the GO-Blot V2 device control panel on a smartphone. The header shows the GOBlot logo, 'Western Blot Processor', and '2021-10-13'. Below is a 'CREATE PROTOCOL' section with a note to 'Enter a unique Protocol details.'. The form includes fields for Protocol (set to 'Test'), Created By (set to 'Tester'), and Action Tray Size (with options Small, Medium, Large). Below these are six steps: Step 1 Block (1 Min), Step 2 Wash (1 X 1 Min), Step 3 Primary (1 Min), Step 4 Wash (1 X 1 Min), Step 5 Secondary (1 Min), and Step 6 Wash (1 X 1 Min). At the bottom are two buttons: 'Import File' and 'Submit'.

Tip: If you cannot see the Submit button on the phone version, shrink the screen and/or flick the address bar off the top of the

screen e.g. on iPhone tap on AA symbol at top left, and chose "hide address bar" option.

Tip: Download folder locations vary, on the iPhone they are located in the Files App > Downloads folder, whereas for the Android they are stored in My Files or File Manager > Downloads. On the PC computer they are found in the main C:/Download folder, or for Apple computers in the Users>Downloads folder or the Finder sidebar.

Setting up users in mygoblot.com

Note: When you purchase a new GO-Blot V2 your name will be used as the main user. Also, you will be given a Serial Number, keep both of these close to the GO-Blot V2 so we can identify it if needed later.

1. Login to <https://mygoblot.com> using your user email and password.

2. Go to the My GO-Blot Devices button and you will see your machine. If you do not see it call Customer Support (303-322-2254 or email cservice@cytoskeleton.com .

3. Now go back to the home page by clicking on top of the webpage, and click the "Register User" button.

4. Type in the User's credentials and click Save.

Note: The person who bought the GO-Blot V2 unit will be the Main User, all others are called Lab Users. If this needs to change contact Customer Service at the email above.

Note: For multiple GO-Blots it is useful to have the same person buying them so that the Main User stays the same and all other lab personnel are entered via the Main User.

The screenshot shows the 'Register GOBlot [Edit]' form on the mygoblot.com website. The form is divided into two main sections: 'Device Details' and 'Authorized Users'. Under 'Device Details', there are input fields for 'Unique ID no.' (containing 'Spec0004'), 'Password', 'Device Name' (containing 'Diagon07'), 'IP' (containing '10.1.86.113'), and 'Port' (containing '80'). Under 'Authorized Users', there is a 'Search User' field and a dropdown menu showing 'kka' with the email 'kka@cytoskeleton.com'. A 'Submit' button is located at the bottom right of the form.

The screenshot shows the 'User Registration [Add]' form on the mygoblot.com website. The form has input fields for 'Name', 'E-mail (User Name)', 'Password', 'Company/Lab Name' (containing 'Cytoskeleton Inc'), and 'User Role' (containing 'Lab User'). A 'Submit' button is located at the bottom right of the form.

Maintenance and Troubleshooting

Sticky/high affinity antibodies

Occasionally you may want to flush out all residues from the GO-Blot V2 device. Especially if there is a sticky-high affinity antibody left over from a previous run. The best way to do this is to add 50 ml of either 1 x Versaclean or 1 x SDS-running buffer from a PAGE system (e.g. a standard composition is 50 mM Tris-HCl pH 6.8, 100 mM NaCl, 0.1% SDS) to all the reservoirs. Then choose the Water Wash protocol to run the solution through the system. Then perform two more Water Washes, but this time use only water to remove the buffer components.

1. While standing next to the GO-Blot V2, connect the luer locks with tubing to the front of the machine.
2. Check that no solutions are in any reservoirs or trays. If they contain solutions, then run a Water Wash protocol to remove the solutions.
3. Place both tubes into a waste container.
4. While on the Main Menu, scroll down by clicking the machine's "Down" button until you reach the My Protocols line.
5. Click the "Enter" button.
6. Scroll down to highlight the Water Wash, and click Enter.
7. An alert should pop-up indicating to fill the Water Wash reservoir, then add 425 ml to the container which covers the level sensor (see p.5 for locating the parts).
8. The alert should disappear and the option to Run the protocol appears, click Enter to run the protocol.

Sticking valves

Occasionally a sticky valve may cause a solution not to flow in the Water wash protocol, its called stiction, defined as the friction that tends to prevent stationary surfaces from being set in motion. A simple fix is to use the syringe adapter included with the kit to perform a quick suck-up action on the port in question, while the Wash routine is running.

Buffer change when using fluorescence antibodies in the GO-Blot V2

Check the manufacturer's instructions as the buffers are different from the usual TBST. The standard changes are to use TBS for blocking, TBS plus 0.2% Tween 20 for primary and TBS plus 0.2% Tween 20 and 0.02% SDS for the secondary antibody.

GO-Blot™, GO-Blot V2™, GO-Blot Western Blot Processor™, Personal Western Archive™, Global Western Portal™, and mygloblot.com are trademarks of Cytoskeleton Inc., Denver, USA. GO-Blot is patented under utility patent numbers US10,288,636, US14,541,708, EU3218112, and a US design patent D29,505,138.