eZwest[™] Lite

Automated Western Device

Start Your Easy Western from Here



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Version 20220615

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Components and Initial setup

eZwest[™] Lite Automated Western Device

Important! Please check that all parts listed below are included with your package.

Components	Cat.	Size	Quantity
eZwest [™] Lite Automated Western Device	L00816	1 unit	1
eZwest Membrane Cassette	L00820	1 unit	1
eZwest Cleaning Cassette	L00821	1 unit	1
Stylus Pen	L00822	1 pc	1
Power Cord	-	1 pc	1
Forceps	-	1 pc	1
Roller	-	1 pc	1
Silicone Tubing	-	3 pcs	1
Waste Bottle	-	1 pc	1
Wash Buffer Bottle	-	1 pc	1
50 mL Tube	-	3 pc	1
Membrane Spacer Sample	-	10 pcs	1
Quick Guide	-	1 pc	1
Manual	-	1 pc	1

Initial setup

For the best result, please follow these instructions to set up the device before your first experiment.

Note: Before relocating the instrument, please drain liquid in the 50 ml tubes.

Step	Description			
1	Check and make sure the power plug matches the outlet			
2	Place eZwest [™] Lite device on a steady and levelled lab bench			
3	Keep the vicinity of the instrument clean and ventilated			
4	Make sure the power switch is OFF before plug in the instrument			
5	Locate the three ports on the back of the device and connect them to corresponding bottles with supplied tubing: wash buffer port to wash buffer bottle, waste 1 and waste 2 ports to waste bottle			
6	Power on the system			
7	The device should beep and start self-check. Once the screen displays the main menu as shown on the right, the unit is ready to go.			

Warranty

GenScript warrants that eZwest[™]Lite Automated Western Device is free from defects in material and workmanship for a period of **one year** from the date of purchase or an accumulative working time of **10,000 minutes**, whichever comes first. If any defects occur during this warranty period GenScript will at its option, repair, replace, or refund the purchase price of the product at no charge to you.

Note: Damage caused by improper transportation, or any of the following actions are excluded:

- · Improper operation.
- Repair or modification done by any other party than GenScript or an authorized agent.
- · Use of fittings or other spare parts supplied by any other party than GenScript.
- · Damages caused by disasters.

For consultation and maintenance services, please contact GenScript's customer service and provide the following information. Instrument model:

Instrument serial number:

Order number:

Date of purchase:

1. Instrument Overview

1.1 Instrument Overview

eZwest[™] Lite is a fully automated blotting device that frees researchers from the manual labor of traditional western protocol. eZwest™ Lite can be programmed to run a sequence of western blot membrane incubation steps automatically including: blocking, primary antibody and secondary antibody incubations and washing. When desired, the primary antibody can be recovered for re-use. The processed membranes are ready for detection. Automation of the incubation steps minimizes errors and variability typically associated with manual operations contributing to experimental reproducibility.

The eZwest[™] Lite device is compatible with the traditional reagents and protocols used for conventional western blotting. No proprietary reagents are required. Fully customizable steps and solutions make eZwest[™] Lite a powerful tool fitting specific experiment needs for both routine and novel western blotting conditions.

Features and Benefits	Hands-free: Press "START" and walk away
	Easy-to-use: Set up in less than 10 minutes
	Reliable: Comparable results to conventional manual method
	Flexible: Fully customized procedures and solutions
	Antibody Saver: Recovery of primary antibody
	Multi-Blot Processing: Incubate up to two membranes at once

1.2 Specifications eZwest[™] Lite Automated Western Device

6.48 Kg		
230×230×230 mm, 9.06x9.06x9.06 in		
100-120 V or 220-240 V, 50/60 Hz		
15-40°C, 59-104°F		
90x83 mm, 3.5x3.2 in		
1 channel for up to 2 membranes		
Western blotting		
ABS, PP, stainless steel, silicone		
ABS		
Stainless steel, POM		

1.3 Ordering information

eZwest Membrane Cassette (included in L00816)

Accessories and other reagents

Size	Quantity	Cat.
1 unit	1	L00820
Size	Quantity	Cat.
1 unit	1	L00821
50 pcs	1	L00824
1 pc	1	L00822
	1 unit Size 1 unit 50 pcs	1 unit 1 Size Quantity 1 unit 1 50 pcs 1

1.4 Safety & Maintenance

To ensure optimal performance, we recommend regularly cleaning the eZwest instrument.

Component	Description
eZwest device: routine cleaning with the	After each use, please clean the device following the procedure below.
membrane cassette	 Insert the membrane cassette into the device. Go to "Method" Select "System Clean" Add 20 mL wash buffer (PBST or TBST) in the Blocking buffer tube. Keep the 1° Ab and 2° Ab tubes clean and empty. Ensure there is at least 100mL wash buffer in the wash buffer bottle and the Waste bottle is empty. Press OK to confirm the buffer volume message. Start "System Clean" method by clicking OK again. After the cleaning procedure is finished, dispose the waste solution.
eZwest device: extensive cleaning with the cleaning cassette	 After every 20 uses or every 2 weeks, perform the extensive cleaning following the procedure below. 1. Empty the Wash bottle and Waste bottle 2. Add 100 mL wash buffer in the Wash bottle 3. Insert the cleaning cassette into the device. 4. Go to "Method" 5. Select "System Clean" 6. Add 20 mL of eZwest cleaning solution (Appendix A-1) in the Blocking buffer tube. Keep the 1° Ab and 2° Ab tubes clean and empty. Click OK to confirm the buffer volume message. 7. Start "System Clean" program by clicking OK again. 8. After the cleaning procedure is finished, remove the cleaning cassette and dispose the waste solution. 9. Perform the routine cleaning protocol once.
Membrane cassette	Open the membrane cassette and rinse with distilled water or 10% ethanol/IPA and let it dry after each use.
Note	If the device will be left unused for over 2 weeks, please ensure the tubes in the device are filled with regular wash buffer (1X PBST or TBST with 20 mM EDTA).

2. Instructions

2.1 Instrument display and feature location

eZwest™ Lite automated western device

Front view





Back view



Top view



Membrane cassette







Fig. 6

Screen display (main menu)

2.2 Basic operation

Save cassette ID when using the cassette for the first time

- 1. Switch on the device.
- Insert the membrane cassette into the device as shown in Fig. 7. Make sure the pins are pushed all the way in, and the side with the "eZwest" logo is upright.

Note: The pins are designed to be movable as part of the auto-seal mechanism. They may appear to be in different position after removing the cassette, which is normal.





- 3. After the cassette is inserted, you will be asked if you want to "save ID".
- 4. Press OK to save cassette ID as show in Fig. 8.

Note: Each eZwest Lite device could save up to 5 cassette IDs.





View and modify methods

1. Enter method setting by pressing "Method" on the upper right corner of the main menu.





2. Select method on the left to show the steps of each method.



Fig. 10

3. Select a step to show and modify the associated details. **Note:** Each Method may be programmed with 6 steps.



Fig. 11

4. Each step within a Method is customizable :

a. Reagent option.

Method 1 04:25	1: Blocking	Reagent: Blocking
Method 3 00.55 Method 4 00:55 Rapid 00:03	2: Wash 3: 1° Ab 4: Wash 5: 2° Ab	Cycles: Wash Time(min) 1° Ab Speed: 2° Ab
System Clean Cassette 0	6: Wash	

Fig.12

b. Number of Cycles required to repeat the step.



Fig. 13

c. Run time for each cycle in minutes.



Fig.14

d. Pump Speed for recirculating the liquid flow through the membrane cassette.

ESC		
Method 1 04:25	1: Blocking	Reagent: Blocking
Method 2 00:55	2: Wash	
Method 3 00:55		Cycles: 1
Method 4 00:55		Time(min): 60
Rapid 00:03		Speed: Normal N
System Clean		High Normal Slow
Cassette D		



5. Once the method is set up, press "Save"

Set cassette specific methods

The eZwest[™] Lite device can store up to one dedicated method for each cassette. Upon cassette installation, select the associated method.

- 1. Method settings can be changed as described above
- 2. Change name of membrane cassette
 - a. Enter Method Setting and select the name of membrane cassette

ESC		Utr Strift
Method 1 04:28	1: Blocking	
Method 2:00:40	2: Wash	
Method 3 00:40		
Method 4: 00:40		
Rapid 00:02		
System Clean		
Cassette 0		Edi D

Fig.16

b. Press "Edit ID" and change name with the pop-up keyboard.



Fig.17

2.3 Using the Pre-programmed blotting method

- 1. Prepare blotting solutions
 - a. Blocking buffer, 10 mL
 - b. Primary antibody solution, 10 mL
 - c. Secondary antibody solution, 10 mL
 - d. Wash buffer, 10 mL per wash cycle, e.g. 90 mL if wash 3 times and each with 3 cycles
- 2. Place the prepared solutions into eZwest[™] Lite device as indicated below.

Note: Assure all fittings and caps are tightly sealed.



3. Open the membrane cassette: release the latch by rotating it up as shown in Fig. 19(i), lift the latch and pull it towards the top of the cassette as in Fig. 19(ii), the opened cassette is shown in Fig. 19(iii).



- 4. Placing membrane(s) in the casette
 - a. Processing a single membrane:

Place the membrane in the center of the cassette surface as shown below in Fig. 20(i), with the protein side facing up and the high MW side aligning towards the bottom edge of the cassette as Fig. 20(ii). Place a spacer across the high MW end (bottom) of the membrane along the membrane edge.

b. Processing two membranes simultaneously:

Place the first membrane as described in Fig. 20(i). Then place a second spacer across the low MW side of the first membrane along the membrane edge. Lay the second membrane on top with the protein side facing down and high MW side align towards the bottom edge of the cassette. Make sure both spacers are in between membranes.

Note: The spacers are reusable for up to 20 times. Replace spacer if deformed or contaminated.



- 5. Carefully close the cassette and lock the latch.
- Securely insert the membrane cassette into the device as shown below, make sure the side with "eZwest" logo is facing up-right.



7. Check and ensure all the tubes and bottles contain adequate amount of reagents.

Warning: insufficient buffer or incorrect solution placement may result in experiment failure.

- 8. Select the method on the main menu and click "Start" to run.
- 9. Once the procedure is finished, click "Finish", and replace the membrane cassette with the cleaning cassette. Membranes are ready for detection.
- 10.Replace all solutions in the reagent chamber with wash buffer or cleaning solution to start the system cleaning cycle when prompted according to the cleaning guidance in 1.4 Safety and Maintenance.

3. Sample Protocols

3.1 Quick Protocol:

Requires sensitive antibodies and/or higher abundant proteins. All steps performed at room temperature.

Step	Cycles	Time	Pump speed
Blocking	1	30min	Slow
Wash	3	5min	Normal
1°Antibody	1	60min	Slow
Wash	3	5min	Normal
2°Antibody	1	30min	Slow
Wash	3	5min	Normal

3.2 Normal Protocol:

All steps performed at room temperature.

Step	Cycles	Time	Pump speed
Blocking	1	60min	Slow
Wash	3	5min	Normal
1°Antibody	1	120min	Slow
Wash	3	5min	Normal
2°Antibody	1	60min	Slow
Wash	3	5min	Normal

3.3 Overnight Protocol:

Use the protocol for very low abundant proteins and/or antibodies with low affinity. **Note:** Overnight protocol can ONLY be saved and run as a cassette method.

Step	Cycles	Time	Pump speed
Blocking	1	60min	Slow
Wash	3	5min	Normal
1°Antibody	Overnight	-	-
Wash	3	5min	Normal
2°Antibody	1	60min	Slow
Wash	3	5min	Normal

1. Press "Method" button on the main interface.



Fig. 22

2. Select the cassette ID (e.g., "345") to enter the cassette program.



3. Select the steps for customization (overnight option is only available to Blocking, 1°Ab,

and 2°Ab steps).

ESC .		Line Save
Method 1 04.25 Method 2 00.55 Method 3 00.55 Method 4 00.55 Rapid 00.03 System Clean 345	1: Blocking 2: Wash 3: 1° Ab 4: Wash 5: 2° Ab 6: Wash	Reagent: Blocking V Cycles: 1 Time(min): 2 Speed: 4 5 Downwart
	E'. 04	

Fig. 24

 Open the dropdown menu for "Cycles" and select "Overnight". The same setting can be applied to 1°Ab or 2°Ab steps.

		Upp Save
Method 1 04:25 Method 2 00:55 Method 3 00:55 Method 4 00:55 Rapid 00:03	1: Blocking 2: Wash 3: 1° Ab 4: Wash 5: 2° Ab	Reagent: Blocking V Cycles: 1 V Time(min): 2 Speed: 4
System Clean 345	6: Wash	5 Overnight
Fig. 25		

5. Press "Save" button to save program.

ESC		C	Use Save
Method 1 04:25 Method 2 00:55	1: Blocking 2: Wash		Blocking 🔽 Overniat 🔽
Method 3 00:55 Method 4 00:55 Rapid 00:03	3: 1° Ab 4: Wash 5: 2° Ab		
System Clean	6: Wash		
345			



6. Press "Use" to use this method and back to main interface.

ESC			Use Same
Method 1 04:25 Method 2 00:55	1: Blocking 2: Wash	Reagent:	Blocking 🔻
Method 3 00:55 Method 4 00.55	3: 1° Ab 4: Wash 5: 2° Ab	Cycles:	Overnigt 🔽
Rapid 00:03 System Clean	5: 2° Ab 6: Wash		
345			
Fig. 27			

7. The initial required reagents for different overnight processes are shown in the table. The loading volume for each reagent is the same as the normal protocol. Press "Start" to initiate the process.

Traget overnight step	Type of reagent needed
Blocking	Blocking, Wash
1°Ab	Blocking, Wash, 1°Ab
2°Ab	Blocking, Wash, 1°Ab, 2°Ab

8. Check buffer volumes for each required reagent.

E.g., if select Overnight Cycle under the 1°Ab step, only Wash buffer, Blocking buffer and 1°Ab solution are required to start the overnight protocol in the eZwest device.



9. After 1° Ab incubation step is finished in eZwest, the machine will automatically pause with a notice as shown in Fig. 29.





- 10. Remove the membrane cassette from eZwest and put it overnight at 4°C.
- 11. A system cleaning cycle is required after the membrane cassette is removed. Insert a cleaning cassette or any other empty cassette for the routine cleaning cycle.



12. The system will first recover the antibodies if Antibody Recovery is selected in the method setting.



13. After recovery is complete, the antibody solution can be stored away for reuse.



Fig. 32

14. Replace the blocking buffer with 20mL wash buffer and leave antibody tubes empty for the cleaning cycle. Check the buffer volumes and start system cleaning.



Fig. 34

15. Put the overnight cassette back into eZwest. The device will recognize the unfinished overnight protocol and request to continue. Click "OK" to continue or "Cancel" to abort.



16. As the blocking and primary antibody incubation steps are completed before the overnight process, prepare only wash buffer and secondary antibody solution for the remaining process. If antibody recovery is selected, put an empty tube for the recovered primary antibody. After checking the volumes by clicking "OK", the program will resume once "Start" is pressed.



4. Troubleshooting and FAQs

4.1 Error messages

Error Message	Possible causes	Solutions
RFID Error	Can't read cassette chip/no cassette	Replace cassette/ insert cassette
No Cassette	Cassette not secured in position	Check Pins and Reinsert cassette securely
Pump Error	Pump malfunction	Contact GenScript
System Error	System malfunction	Contact GenScript

4.2 FAQ

Description	Possible causes	Solutions
Strong background	Insufficient wash	 Increase number of wash cycles (set to 5 cycles for 3 minutes each)
		 Increase pump flow per wash cycles (set to normal)
Weak signal	 Antibody is too diluted Instrument needs cleaning Insufficient antibody incubation time 	 Reduce antibody dilution factor Clean eZwes[™] Lite instrument Increase antibody incubation time or reduce incubation pump flow
Fluorescent detects only strong signal around spacer	Weak target signal contrasted out by interference	 Adjust detection area to omit spacer position Cover spacer signal region with paper or tape

5. Technical Support

Visit the GenScript web site at www.genscript.com for:

- Technical resources, including manuals, vector maps and sequences, application notes, MSDSs, FAQs, formulations, citations, handbooks, etc.
- 2. Complete technical support contact information
- 3. Access to the GenScript Online Catalog
- 4. Additional product information and special offers

For more information or technical assistance, call, write, fax, or email. GenScript USA Inc. 860 Centennial Ave. Piscataway, NJ 08854 Tel: 732-885-9188, 732-885-9688

FAX: 1-732-5185-5150

Email: product@genscript.com

Appendix

A-1. Buffer recipes

eZwest cleaning solution	Recipe	
(0.1M NaOH, 10% ethanol)	NaOH	4 g
	Ethanol	100 mL
	Add water to	1 L
		Mix thoroughly
Regular wash buffer	Recipe	
-	1× PBST / TBST	1 L
	EDTA-2Na	7.5 g
		Mix thoroughly
Blocking buffer (50 mL)	Recipe	
	Dry milk (defat)	2.5 g
	1 X PBS	50 mL
		Mix thoroughly
10 X TBS buffer (1 L)	Recipe	
	Tris base	24 g
	NaCl	88 g
		Dissolve in 900 mL water, pH to 7.4 with 12N HCl
	Add water to	1 L
1 X TBST buffer (1 L)	Recipe	
	10 X TBS	100 mL
	Tween20	0.5 mL
	Add water	900 mL
		Mix thoroughly, pH to 7.4
10 X PBS buffer (1 L)	Recipe	
	Na ₂ HPO ₄ ·2H ₂ O	17.8 g
	NaCl	80 g
	KCI	2 g
	KH ₂ PO ₄	2.4 g
	Add water to	1 L
		Mix thoroughly
1 X PBST buffer (1 L)	Recipe	
	10 X PBS	100 mL
	Tween20	0.5 mL
	Add water	900 mL
		Mix thoroughly, pH should be around 7.4

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