NeoBlot Auto Mini,Automated western blot processor, two tray model, touchscreen

NB-12-8102

Table of Contents

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#Cat: NB-12-8102 **Size: 1unit**

Specifications and Safety.

Please read this manual thoroughly before operating the NeoBlot.We suggest that you keep this manual, as you may need to refer to it.
The NeoBlot complies with the European Community Safety requirements. Operation of the NeoBlot is subject to the conditions described in this manual. The protection provided may beimpaired, if the equipment is used in a manner not specified by Neo Biotech.

NOTE:
Notes will be used throughout the manual to inform on important points and provide useful hints.

CAUTION:
Cautions will be used to inform the reader of action that may have the potential to either harm the instrumentation or affect the quality of the data.

WARNING:
Warnings are used to provide special notice of actions that have the potential to cause harm to the operator.

For technical assistance, call, write, fax, or email.

Call: +33977400909
Fax: +33977401011
Email: info@neo-biotech.com
Write: Customer Support
Neo Biotech
74 rue des suisses
92000 Nanterre France

Specifications:

Input voltage: 100-120VAC or 220-240 VAC (check unit label)
Voltage variation: +/-10%
Phase: Single phase
Power frequency: 50Hz or 60Hz
Rated Input current: 5.0A max
Overvoltage category: Transient overvoltage category II Rated pollution applied: Pollution Degree 2

The device must be connected to a mains socket outlet with protective earthing connections.
- Japan
Use the cable that comes with the product.
- North America
Use the cable that comes with the product.
- Other countries
AC power cable is not attached to the product. Use a power cable that conforms to the regulations in the country where the product is to be used.

Blot Processing Polyurethane Trays and Tank. Mini 8–25 ml per chamber, 9.5 x 7.5 cm
Midi 20–40 ml per chamber, 9.5 x 15 cm
Delta: 3-15 ml per chamber, 9.5 x 4 cm
Mega 40-80 ml per chamber 9.5x30 cm
Dimensions (h × w × d) 44 × 30 × 26 cm
Weight 12.0 kg

Safety certifications: CE directive 2006/95/EC and 2004/108/EC, standard used EN61010-1, IEC61010-1, EN 61326- 1:2006, IEC 61326-1:2005

Installation location conditions
Operation site: Indoors
Maximum operating altitude: 2500 m or lower Operating temperature 3ºC to 42ºC
WARNING:
(1) Do not operate the instrument under voltage fluctuations exceeding 10% of the recommended line voltage. Large fluctuations may cause the instrument to fail. Use a three-pronged electrical outlet with a ground.
(2) Instrument can be used in the temperature range 3°C - 42°C, avoid freezing. Do not install the equipment at a place where the temperature changed frequently.
Instrument can be used under a humidity range of 30 - 80% (RH). Relative humidity less than 80% from 3ºC to 30ºC, decreasing linearly to 50% from 31ºC to 42 ºC
(3) Do not install the equipment near a heating element.
(4) Do not install the equipment at a place where it may be exposed to corrosive gas.
(5) Do not install the instrument in a location where it may be exposed to dust, especially in locations exposed to outside air or ventilation outlets that discharge dust particles.
(6) Do not install the equipment at a place constantly or excessively exposed to oscillations or impacts.
(7) Do not install the instrument in a location where it may be exposed to direct sunlight.
(8) Avoid strong magnetic fields and sources of high-frequency waves. The instrument may not function properly near strong magnetic fields or high frequency wave sources.

The WEEE (Waste Electrical and Electronic Equipment) symbol indicates that this product should not be disposed of in unsorted municipal waste. Follow local municipal waste ordinances for proper disposal provisions to reduce the environmental impact of WEE

Unpacking the NeoBlot.

When you receive the NeoBlot, carefully inspect the shipping box for any damages, which may have occurred during shipping. Any damage to the container may indicate damage to its contents. Open the box and take out the top portion and then the second layer with tray. Remove all solutions and accessories parts from thebox.

Using the upper portion of NeoBlot, pull NeoBlot out of the bottom holder and out of box and put it on a flat surface; it is a good idea that you have a lab colleague help with the system removal from the box. Removethe bag. Put the instrument on a solid and leveled surface, and inspect for any damage.

Examine the unit carefully for any damage incurred during transit. If you suspect damage to the contents may have occurred, immediately file a claim with the carrier in accordance with their instruction before contacting Neo Biotech. The warranty does not cover in-transit damage. Notify Neo Biotech (info@neo-biotech.com) of any claim filed.

Put all parts back into the box and save them in case you need to send instrument back for service.

Package Contents:

NeoBlot-mini with two or more trays
Waste container (may be attached to the unit or in separate bag)
Wash Bottle
Power cord
Tray lid

Note: check packing slip for additional parts or content modifications.

Installation.

Take the power cord from the box, connect it to the back of instrument and plug into power output. Check the label on the back panel and select a correct input voltage (100-120 v or 220-240V) and frequency (50 or 60 Hz). Place waste container under instrument.

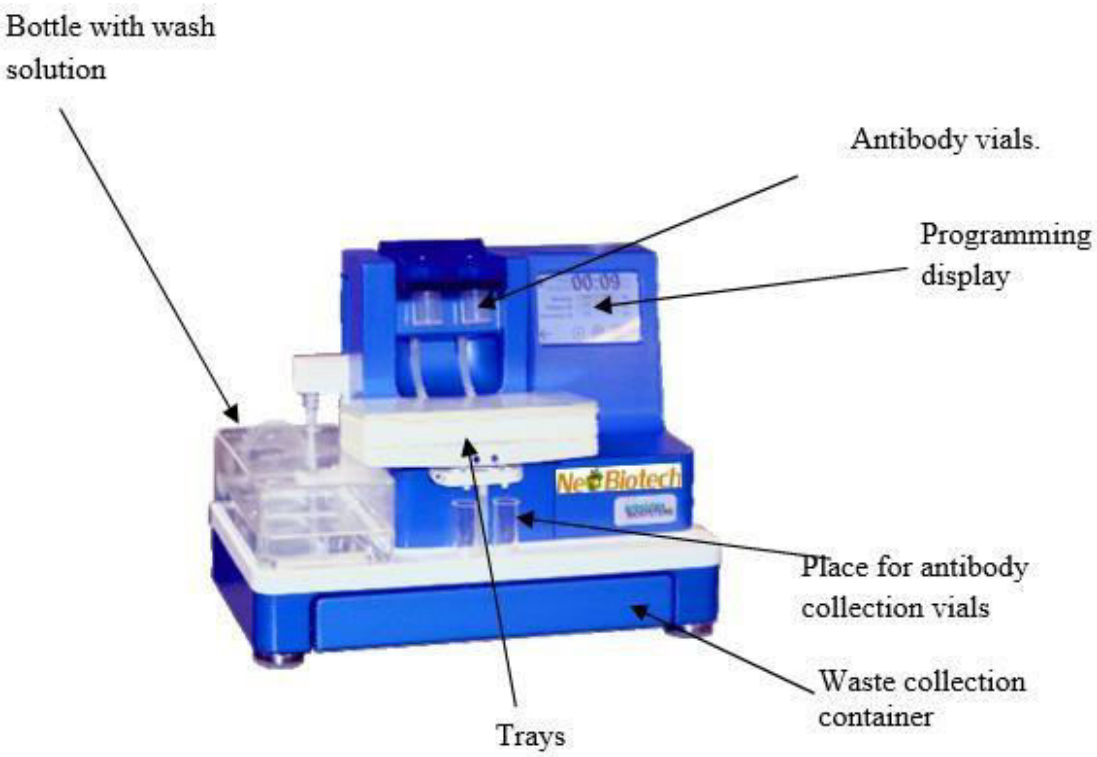
Remove protective film from the display and tray lids.

Warning: For personal safety the NeoBlot must be properly grounded. The user should have the wall receptacle and circuit checked by a qualified electrician to be assured that the receptacle is properly grounded. Where a two-prong receptacle is encountered, it is the responsibility of the user to replace it with a properly grounded three-prong wall receptacle. Do not under any circumstances, cut or remove the third ground prong from the power cord. Do not use a two-prong adapter plug.

Warning: Do not operate around flammable liquids or gases.

Overview

NeoBlot Auto Mini consists of programming display, two trays for blots, holder for vials for primary (P) and secondary (S) antibodies, waste container.



Test run

Turn on the instrument using the switch on the back of the unit. The software loading will take about 90 sec, the display should light up and you should see the first screen with information about different functions:

We recommend starting with cleaning:

Fill water or diluted cleaning solution into wash bottle (about 1L); make sure water is not leaking.

Press maintenance button and then press cleaning button. Now you started the cleaning cycle. Make sure that solution is going into the trays and out of the trays. You may stop the cleaning cycle at any time as soon you verified that everything is working.

A.Programming the NeoBlot

Before starting actual western blot processing get familiar with programming interface

A1. User Interface overview

User interface contains 4 buttons:

protocol press this button to select existing or set up a new protocol.

run press this button to start a protocol

status press this button to check the status of the currently running protocol and time to completion

maintenance press this button to start cleaning and perform other functions (see below)

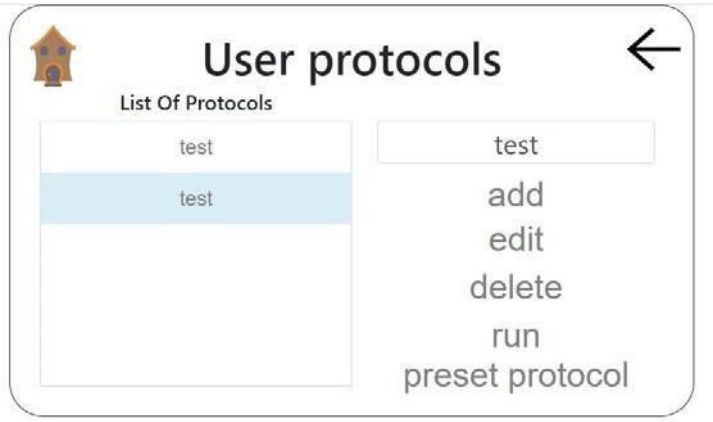
A2. Protocol overview.

NeoBlot is set up to run standard protocol for western blot starting with blocking, washing after blocking, and then primary antibody incubation, washing after primary antibodies, secondary antibodies incubation and washing after secondary antibody incubation. Each step can be programmed independently. At the end of all steps the trays are filled with washing solution and shaking is stopped.

A3. Programming NeoBlot (protocol set up).

To start protocol set up press protocol**.**

On the new screen you can select existing protocols, modify existing protocol or set up a new protocol. You can set up and save up to 50 custom protocols and there are 4 preset protocols that cannot be changed:

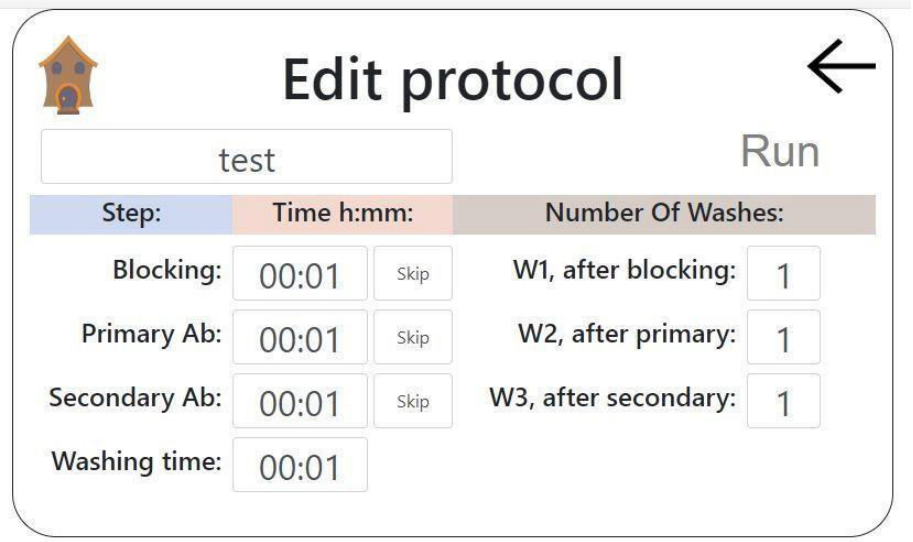


To select a preset protocol touch preset protocol you will see a choice of four preset protocols, select one to run your protocol, a new screen to start run will appear (see below)

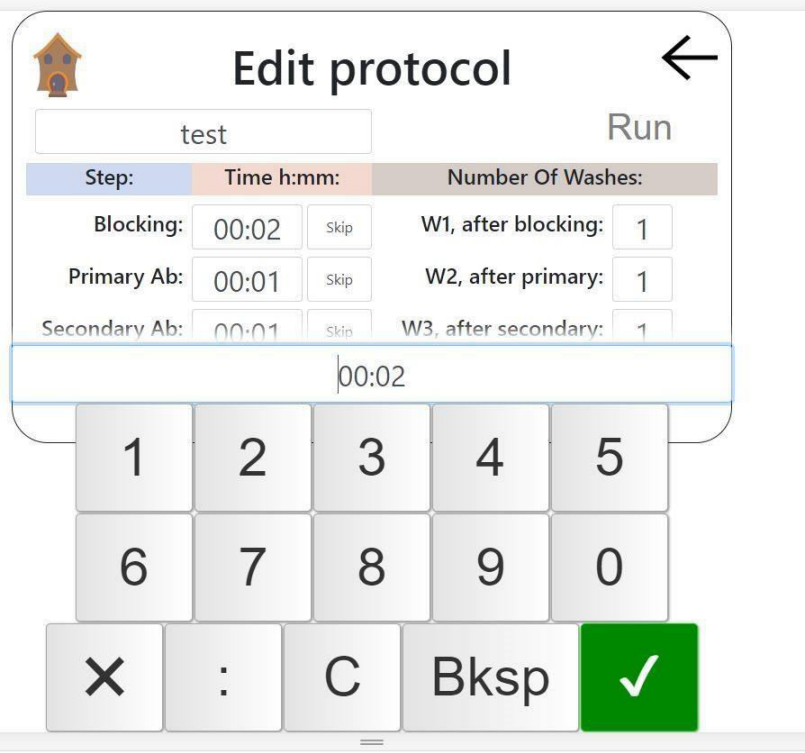
To add a new protocol touch add and then touch white area. Using keypad enter new protocol name.
To delete last protocol press delete.

To select or modify existing protocol, highlight protocol by touching it and press edit. On

the following screen you can change any protocol parameters:



Set the incubation time for blocking, primary (PA) and secondary (SA) incubation by touching adjacent white area.



Note: before entering time press C ***to clear previous setting.***
The minimum time for blocking is 5 minutes, the minimum time for PA and SA incubation is 10 minutes.
Note: press skip ***to skip the step, zero time for blocking, PA or SA will appear.***

Set the number of washing cycles:

By touching the white area on left side of the screen set the number of washing cycles after blocking, PA(primary Ab) incubation time and SA(secondary Ab) incubation time.

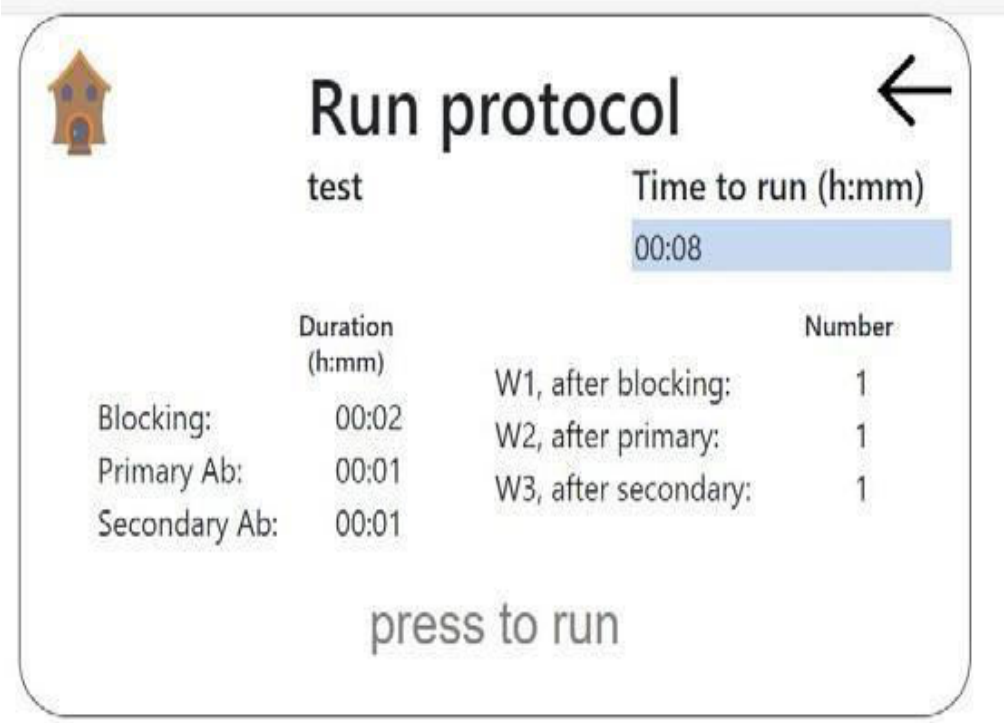
Note: you can skip washing step by selecting zero

Set the duration of washing cycle:

By touching the white area on left of **"washing time"** you select the duration of each washing cycle. It can be set between 3 and 20 min.

After finishing the protocol modification select protocol you like to run.
Please note if the button is dimmed, the filed cannot be selected

Press run button on the right side of the screen and use the following screen to verify protocol selection and to start protocol (see below).



B. Set up and Operation

B1. Before starting, you will need to run electrophoresis, transfer protein to membrane and prepare the following solutions:

- Blocking solution for each blot: 8-15 ml mini tray and 15-25 ml for midi tray
- Primary Antibody (PA) for each mini trays 6-15 ml , for midi trays 20-25 ml; for delta trays 3-8 ml
- Secondary Antibody (SA) for each mini trays 12-18 ml for midi trays 20-25ml, for delta trays 4-10 ml
- Washing buffer up to 2 L (depending on the number of blots)

Note: Use 0.1% Tween 20 in the washing, blocking and antibody buffers. This will reduce the surface tension of solutions and ensure even distribution of the antibody over the blot during incubation.

B2. Loading of the Blot Cycler

Note: If the NeoBlot has not been used for several days, run a cleaning cycle first.

- Remove tray covers, place membranes in the trays
- Add blocking buffers to each tray containing a membrane.
- Close the trays by replacing the covers
- Open vial holder lid
- Add Primary Antibodies (P1 – P2) in collection vials and place **in front** vial holders, close lids. Place collection vial for primary antibody collection under trays if you are planning to re-use antibodies otherwise place dummy vials if available.

Note: Make sure that primary antibody (P) and trays are matched.

- Add Secondary Antibodies (S1 – S2) in the vial holder **on the back** and close lids.

Note: Make sure that secondary antibody (S) and trays are matched. The maximal volume of solution in each vial is 25 ml

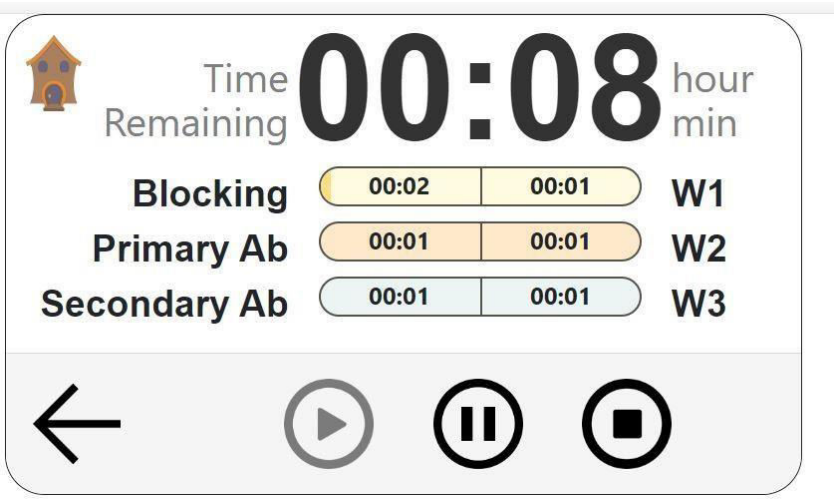
- Fill the bottle with washer to the appropriate level and tight the lid, place bottle horizontally and lower the sipper
- Place a waste container under instrument.

B3. Start Cycling

Note: you do not need to change any settings, if you are using the same protocol as before.

- Press run button:

- A new screen will appear that show the status of current protocol:



The changing color of the bar indicates the current step, the number inside indicates the time to completion of the current step.

In order to return to previous screen press arrow button.

B4. Pause Cycling

- you can pause protocol by pressing ll button.

To continue protocol press ► button.

Note: protocol will start from next step if it was close to the end of the previous step.

To completely stop protocol use ■stop buttons

B5. Cleaning

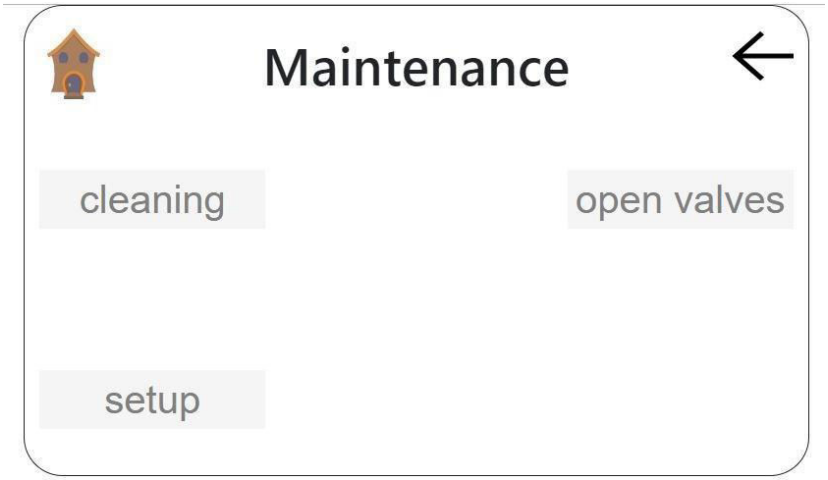
Note: you need to do a cleaning after protocol is finished
Vials for antibodies are not cleaned and should be replaced.

- Remove all membranes.
- Fill the washing bottle with diluted cleaning solution (it is a good idea n have two bottles: one for cleaning and one for washing during protocol execution)
-

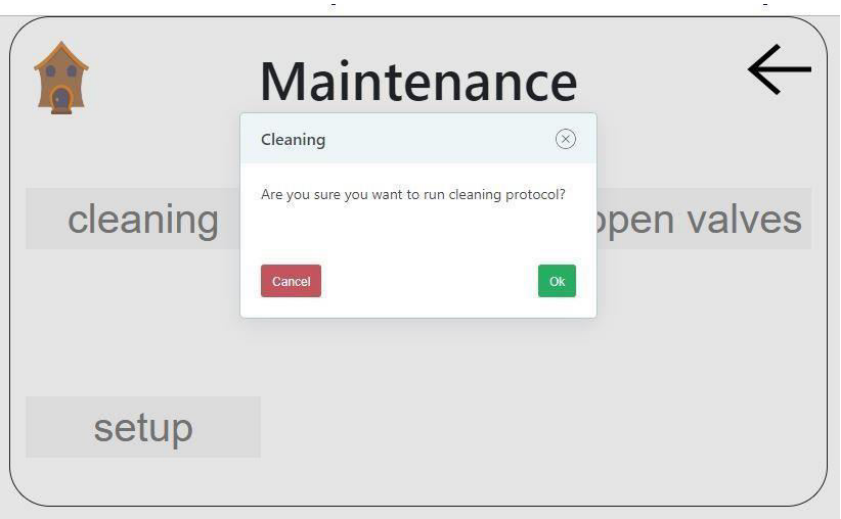
Note: Make sure there is enough cleaning solution otherwise the pump can be damaged.

- Go to the home screen and press maintenance button, a new screen will appear:

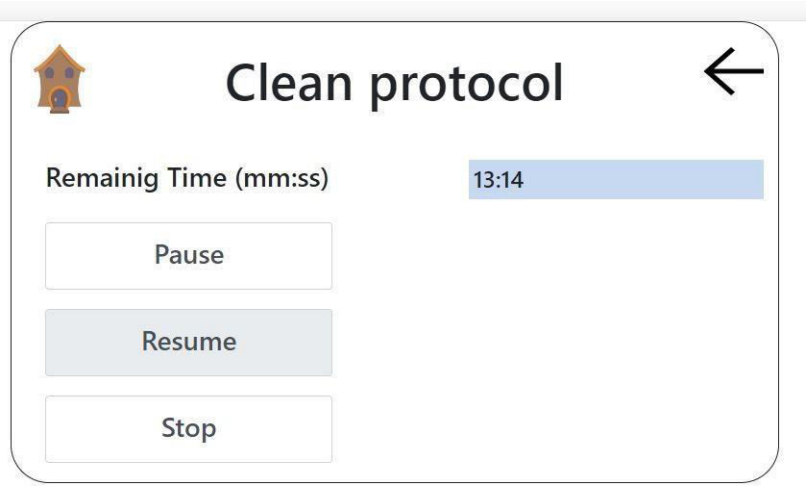
Press cleaning and click Ok on pop-up window.



click Ok on pop-up window.



The new window will appear:

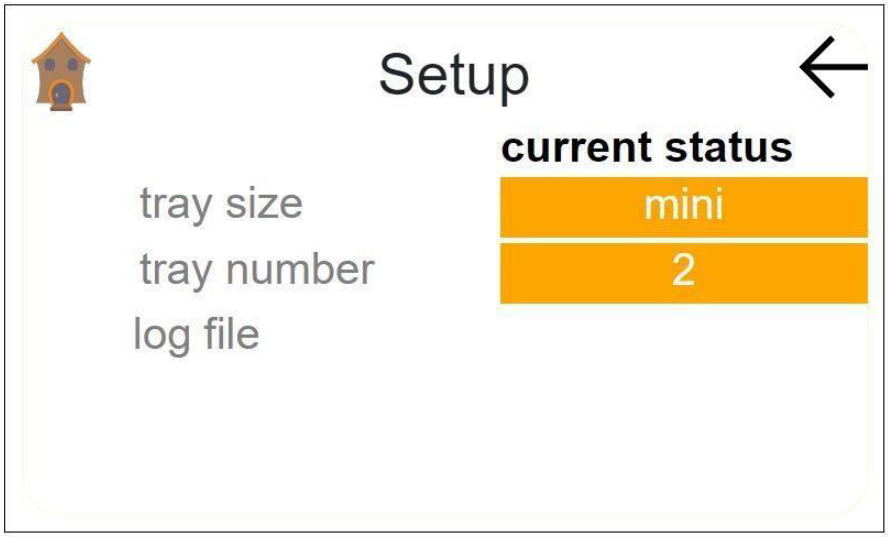


Note: you can stop or pause cleaning at any time.

B6. Tray size setting

Note: Finish all protocols and remove liquid before next step. Refer to separate instruction regarding tray replacement

Go to the home screen and press maintenance button, a new screen will appear. Touch setup button and login using password *setup*.
The new screen appears:



Press tray size button:

Select correct tray size:

Go to setup screen and make the correct tray are selected

B8. NeoBlot Maintenance:

Run cleaning cycle every time you finished western blot processing. We recommend to use the cleaning solution (cat #CL500).

If the NeoBlot is not intended to be used within next 24h, run cleaning cycle with cleaning solution and then with deionized or distilled water and open all valves. It is a good idea to keep valves open while NeoBlot is not in operation.

Go to home screen and press maintenance button and then open valves button.

- When all valves are open, the beep sound and the message will appear. Now you can turn NeoBlot off.

- You can turn NeoBlot off.

Warning: ***If there is a risk that a large volume of spilled liquid has penetrated the casing of the instruments and come into contact with the electrical components, immediately switch off the system, wipe out all spilled solution and do not operate until completely dry.***

Appendix
Troubleshooting

Problem	Possible Cause	Solution
No power (the digital display remains black longer than 2 minutes when the power is turned on)	AC power cord is not connected. Fuse has blown.	Check AC power cord connections at both ends. Use the correct cords. Replace the fuse If the problem still persists after verifying that correct power cord is used and the fuse is replaced, contact Technical support.
Buffers leak from the trays immediately	Valves remain open.	Start any protocol valve will be closed. If this does not help turn instrument off, wait at least 5 sec and turned instrument on. After initialization valves will be closed.
Weak or no signal from the blot	Detection step missed or detection reagents not working.	After the blot processing is complete, perform the detection step using your standard detection reagents and protocol manually. Make sure the detection reagents are functional.
	Insufficient incubation with detection reagent	Remove blot from detection reagent when signal-to-noise ratio is acceptable.
	Poor or incomplete transfer	Make sure transfer apparatus and membrane sandwiches are assembled correctly. Use appropriate transfer times. After blotting, stain membrane to measure transfer efficiency.
	Protein of interest ran off the gel	Use positive control and/or molecular weight marker to match gel separation range to size of protein being blotted. After blotting, stain membrane to measure transfer efficiency.
	Incorrect reagents added or incorrect containers are filled	Make sure that primary and secondary antibody are added to correct containers and number on antibody container in the tank and tray match each other.
	Sample too dilute	Load the larger amount of protein onto the gel or increase concentration of proteins.
	Poor retention of proteins or protein weakly bound to membrane	Use membranes with appropriate binding capacity. Dry PVDF membrane after protein transfer to ensure strong binding of the proteins.
	Inactive or overly dilute primary or secondary antibody	Determine antibody activity by performing a serial dilution using six trays or dot blot. Increase antibody concentration as necessary.
High background on the blot	Film overexposed or became wet during exposure	Decrease exposure time or allow signal to further decay. Prevent leakage of solutions by encasing membrane in transparency film and blotting excess substrate from edges before exposure.

	Short blocking time or washing intensity High concentration of primary and/or secondary antibody	Increase blocking time and the number of washes Determine optimal antibody concentration by performing dilution series using all six trays. Decrease antibody concentration as necessary.
	Protein is overloaded	Reduce load or dilute concentration of sample.
	Membrane, solutions, trays, or antibody containers are contaminated	Use clean glassware and purified water to prepare solutions. Wear clean gloves at all times. Use forceps when handling membranes. Run cleaning protocol with cleaning buffer, increase the concentration of cleaning buffer two times
	Protein is overloaded	Reduce load or dilute concentration of sample.
Non-specific binding too high	Insufficient removal of SDS or weakly bound proteins from membrane after blotting	Follow proper protocol for membrane preparation before immunodetection.
	Short blocking time	Increase blocking time.
	Affinity of the primary antibody for the protein standards	Check with protein standard manufacturer for homologies with primary antibody.

Replacing the Fuse

Follow the instructions below to replace the 250V, 5A rated fuse for the power socket.

1. Turn off NeoBlot using switch on the back of the instrument and detach the power cord from the rear of the instrument.
2. Open the fuse compartment located on the power entry block using a small flat blade screwdriver or fingernail to gently open the fuse compartment.
3. Pull the fuse holder out of the compartment and inspect the fuse. If the fuse is burned or there is a break in the fuse element, replace with the identical type fuse.
4. Place the fuse holder back into the compartment and snap the cover closed.

For additional fuses, contact Customer Support.

Accessory Products

The following products are for use with the NeoBlot and are available separately from Neo Biotech.For more information visit www.neo-biotech.com or contact Customer Support.

vi100	Antibody collection vial per 100 (25 ml max volume)
vi1000	Antibody collection vial per 1000 (25 ml max volume)
CL500	Cleaning solution 50x, 500 ml
CL5005	Cleaning solution 50x, 500 ml, 5 bottles
CL5010	Cleaning solution 50x, 500 ml, 10 bottles
BH500	Hybridization buffer, 500 ml
BW1000	Washing buffer 10x,1L
BW2001	Washing buffer 20x,1L
BW2002	Washing buffer with tween, 20x,1L
BL500	Blocking buffer, 500 ml
SB200	Stripping buffer, 500 ml
TRMN02	block of two mini trays for model W20
TRMD02	block of two midi trays for model W20
TRDL02	block of two delta trays for model W20
TRMG01	mega tray for model W20
WHBTL02	wash bottle for model W20
DRCNT02	drain container for model W20
TRCVMD02	Tray cover for mini and delta trays, model W20
TRCVDG02	Tray cover for midi and mega trays, model W20
DRVI02	Drain (dummy) vial (pack of 2)
TRTUBW20	Set of tubing for model W20
TBDRN3	Drain tubing 3 ft
TBDRN5	Drain tubing 5 ft
TBDRN10	Drain tubing 10 ft
BL004	Base leveller (levelling mount) (each)

Warranty

The NeoBlot is warranted for one (1) year against defects in materials and workmanship. If any defects should occur during this warranty period, Neo Biotech will repair and replace the defective partswithout charge.

However, the following defects are specifically excluded:

1. Defects caused by improper operation and maintenance.
2. Repair or modification done by anyone other than Neo Biotech or their authorized agent.
3. Use with other spare parts not specified by Neo Biotech.
4. Damage caused by deliberate or accidental misuse.
5. Damage due to use of improper solvent or sample.
6. Replacement of tray tubing.

For inquiry or request for repair service, contact Neo Biotech.

Technical Help

For more information or technical assistance, call, write, fax, or email.

Call: +33977400909
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