NeoBlot Auto, Automated western blot processor, with 4 midi trays and touchscreen

NB-12-8101-MIDI4

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#Cat: NB-12-8101-MIDI4 **Size: 1unit**

Specifications and Safety.
Please read this manual thoroughly before operating the NeoBlot Auto.
We suggest that you keep this manual, as you may need to refer to it.
The NeoBlot Auto complies with the European Community Safety requirements. Operation of the NeoBlot Auto 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 theinstrumentation 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: 110VAC or 220-240 VAC (check unit label)Voltage variation: +/-10%
Phase: Single phase
Power frequency: 50Hz or 60Hz (check unit label)Rated Input current: 5.0A max
Overvoltage category: Transient overvoltage category IIRated 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 12–40 ml per chamber, 9.5 x 15 cm
Delta: 3-15 ml per chamber, 9.5 x 4 cm
Dimensions (h × w × d) 35 × 34 × 42 cm (with Mini tray)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 lowerOperating 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 bedisposed of in unsorted municipal waste. Follow local municipal waste ordinances for proper disposal provisions to reduce the environmental impact of WEEE.

Unpacking the NeoBlot Auto.

When you receive the NeoBlot Auto, carefully inspect the shipping box for any damages, which may have occurred in shipping. Any damage to the container may indicate damage to its contents. Watch video how tounpack NeoBlot Auto

Open the box and take out the enclosed flat box with tray and tank covers and tray plugs. Remove all solutionsand accessories parts from the box.

Using the upper part of the green Styrofoam as a handle, pull NeoBlot Auto out of the box and put it on aflatsurface. Gently remove front and back of the protective Styrofoam; it is a good idea that you have a lab colleague help with the system removal from the box. Remove the 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 mayhave occurred, immediately file a claim with the carrier in accordance with their instruction before contactingNeo Biotech. The warranty does not cover in-transit damage. Notify Neo Biotech (info@neo-biotech.com) of any claim filed.

Remove the styrofoam insert between the black pumps inside tank. Put all parts back into the box and save themin case you need to send instrument back for service.

Packing List:

NeoBlot Auto with six, five or four trays
Waste tubing (may be attached to the unit or in separate
bag)Power cord
Tray lids, 2
Tank lid, 1
Tray plugs, 4
Transformer 220-240 V /110V (optional)

Installation.

Take the power cord from the box, connect it to the back of instrument and plug into power output. Check thelabel on the back panel and select a correct input voltage (110 v or 220V) and frequence (50 or 60 Hz). Attached the drain tubing to back of the instrument and place the drain tubing into a waste container (not provided) or directly into the sink.

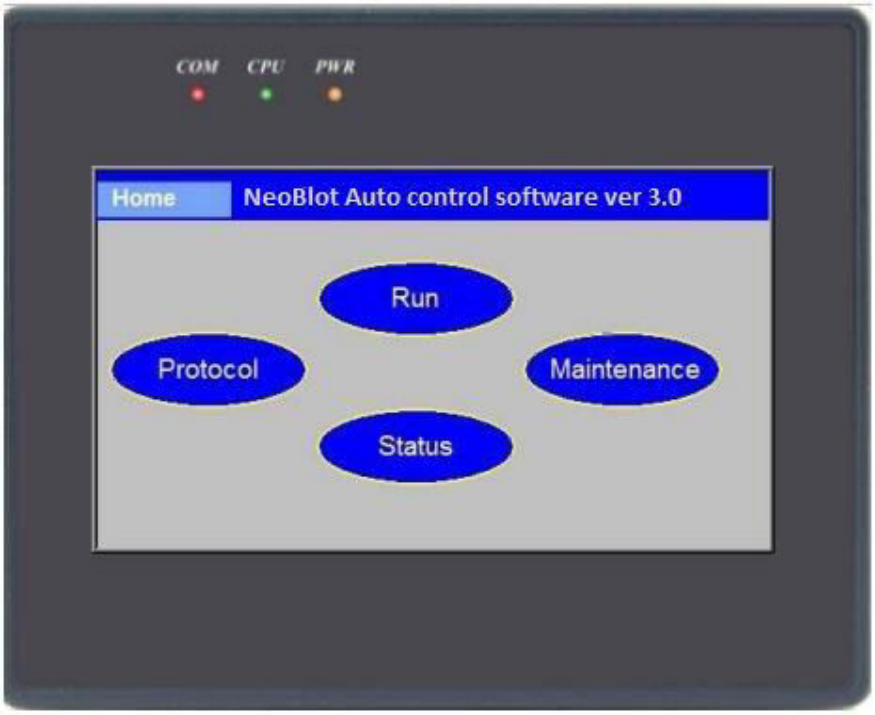
Make sure the longer tubing under the trays (see picture) is inserted into thesmall holes in the waste collectors on the left and right sides of the instrument.

Remove protective film from the tank and tray lids.

Warning: For personal safety theNeo Blot Auto 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 powercord. Do not use a two-prong adapter plug.
Warning: Do not operate around flammable liquids or gases.

Test run

Turn on the instrument using the switch on the back of the unit. The display should light up and you should seethe first screen with information about different functions:



Check that the silicone plug is inserted in the tank between the black pumps. Make sure that the waste tubing isgoing down and is not bent. (Remember there is no pump in NeoBlot Auto the solution moves under gravity). **It is important for solution removal that the waste tubing is not bent allowing the solutions to drain properly,also the outlet of the tubing should not be immersed in the waste solution.**

Left side cleaning:

Fill water 2-3 cm above minimal level (about 2 L); make sure water is not leaking.

Press 

button and then press 

button, select left side. Now you started the cleaning

cycleon the left side. 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 on the left side is working.

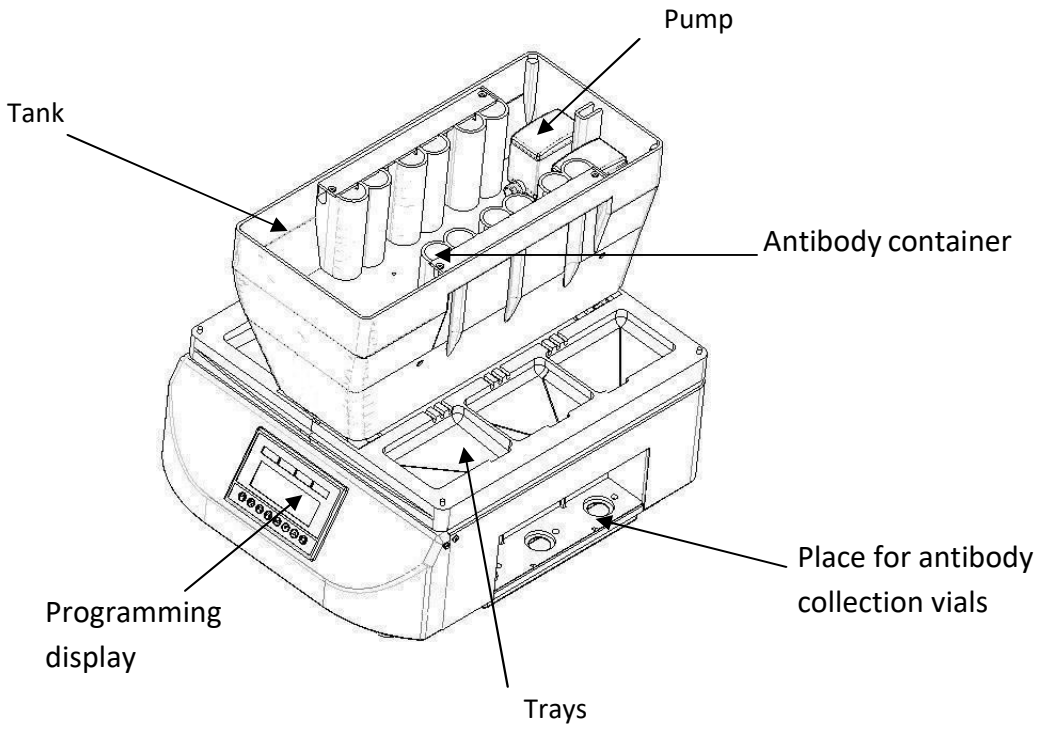
Right Side cleaning:

Repeat the same procedure for the right side, adding more solution if needed. Press 

button and select right side. The cleaning of the right side should start. The solution should be coming out from all threetubes in each tray.
Now the instrument is ready for use. Read the following instructions to program and start using the NeoBlot Auto.

Overview

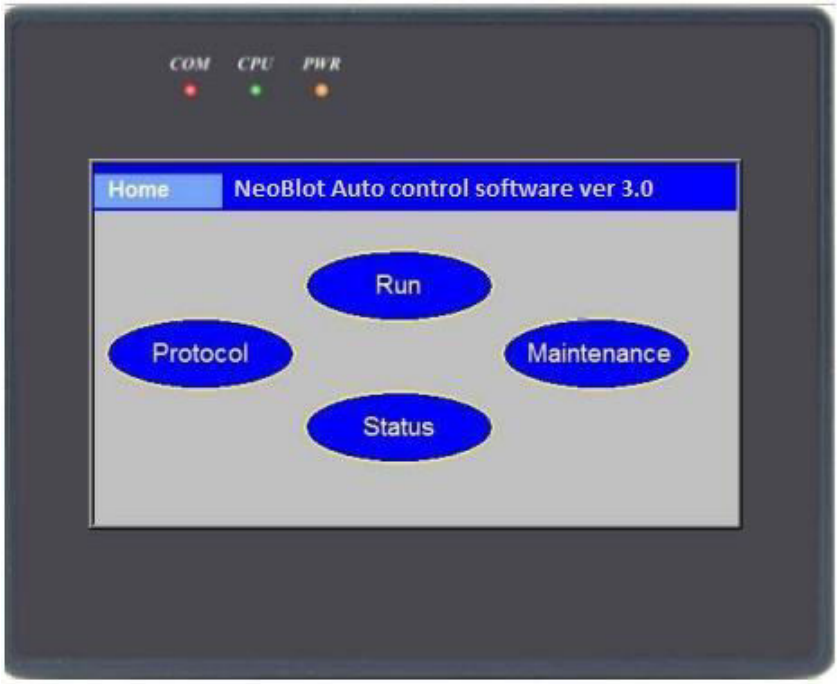
NeoBlot Auto consists of programming display, four to six trays for blots (trays #1-3 on the left side and trays#4-6 on the right side), tank with container for primary (P) and secondary (S) antibodies and pumps forcleaning.



A.Programming the NeoBlot Auto

Before starting actual western blot processing get familiar with programming interface

A1. User Interface overview



User interface contains 4 buttons:

press this button to select existing or set up a new proto-

col. 

press this button to start a protocol

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

tion 

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

A2. Protocol overview.

NeoBlot Auto 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; the antibody containers are also filled with washing solution to facilitate cleaning.
***Note*:** *In order to skip blocking and primary antibody incubation set the Primary antibody incubation time to zero or press 'skip' button. Protocol will start at second washing step.*

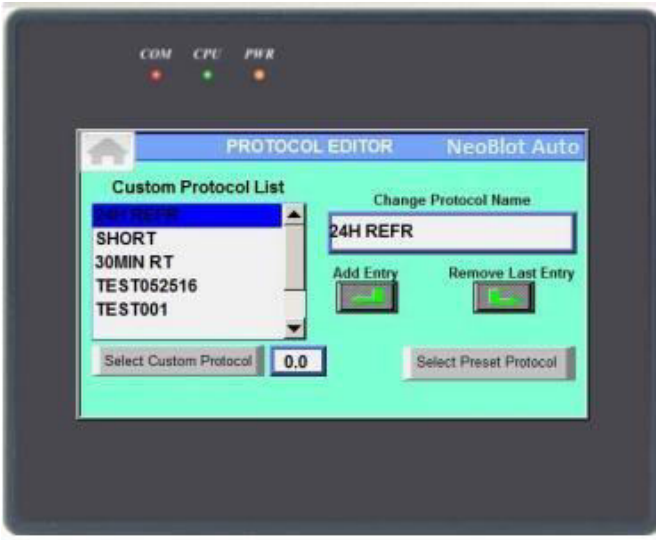
A3. Programming NeoBlot Auto (protocol set up).

To start programming the operator needs to initialize the system by turning the system OFF and ON using theON/OFF switch on the back of the unit. Please wait 3 sec before turning it on.

To start protocol set up press 

.

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 20 protocols and there are 4 preset protocols that cannot be changed:



To select a preset protocol touch 

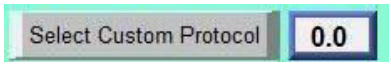
you will see a choice of four preset protocols, select one, and choose on which side(right or left) to run your protocol, a new screen to start run will appear (see below)

To add a new protocol press 

and then touch white area under "**Change Protocol Name**". Using keypadenter new protocol name.

To delete last protocol press 

.

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

.

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: The minimum time for blocking is 5 minutes, the minimum time for PA and SA incubation is 10 minutes.

Note: press  ***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 after blocking by selecting zero, but you cannot skip washing after primary andsecondary antibodies incubation.

Set the duration of washing cycle:

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

After finishing the protocol modification select side you would like to run:  .

Please note if the button is dimmed, this side is running and cannot be selected

Press  button on the top of the screen and use the following screen to verify protocol selection and tostart protocol (see below).

A.Set up and Operation

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

- Blocking solution for each blot: 12-18 ml mini tray and 18-30 ml for midi tray
- Primary Antibody (PA) for each mini trays 8-15 ml , for midi trays 18-25 ml; for delta trays 3-10 ml
- Secondary Antibody (SA) for each mini trays 12-18 ml for midi trays 18-30ml, for delta trays 4-12 ml
- Washing buffer up to 3.5 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 NeoBlot Auto

Note: If the NeoBlot Auto 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

Note: tray cover has a cut that should be on the upper side to ensure trays are covered completely.

- Remove tank cover
- Add Primary Antibodies (P1 – P6). Insert collection vial for primary antibody collection and re-use

Note: Make sure that primary antibody (P) and trays are matched.
(For midi size trays you can use only 1 and 3 on the left side and 4 and 6 on right side)

- Add Secondary Antibodies (S1 – S6).

Note: Make sure that secondary antibody (S) and trays are matched.
(For midi size trays you can use only 1 and 3 on the left side and 4 and 6 on right side)

- Fill the reservoir with washer to the appropriate level, replace the top cover.
- Place a waste container, making sure it is below the instrument level and the waste tube does not touchthe waste solution.

Note: If you do not use all trays you can plug washing tubing with yellow plugs provided.

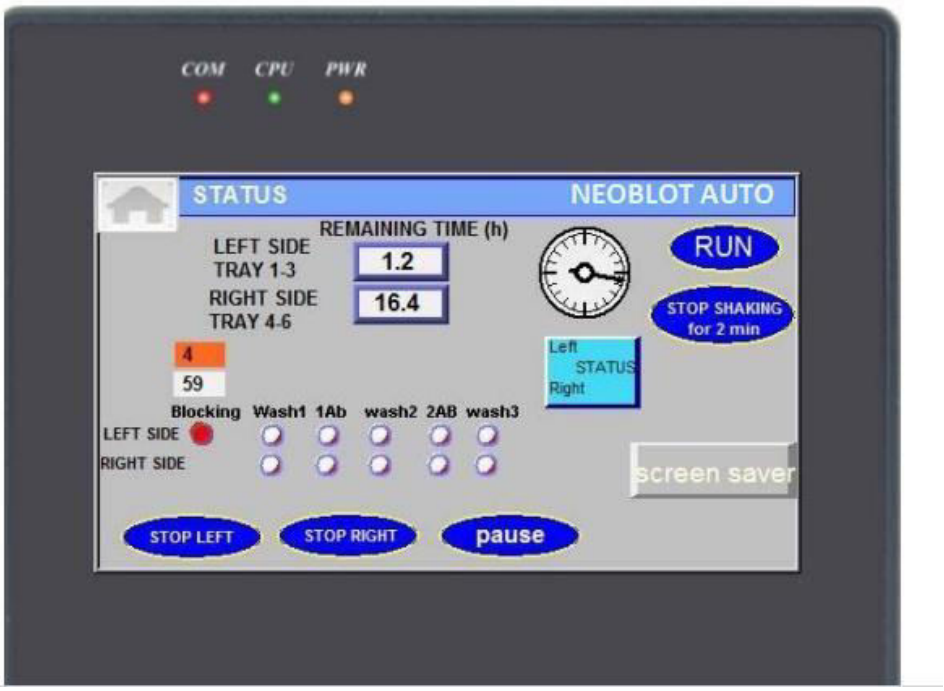
B3. Start Cycling

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

- Press 

button to open a following screen:

- Press corresponding start button to start cycling for left (Tray 1-3) or right side (Tray 4-6).
  A new screen will appear that show the status of current protocol:



Red blinking dot indicates the current step, the number above it indicate the time to completion of the current step.

In order to return to previous screen press 

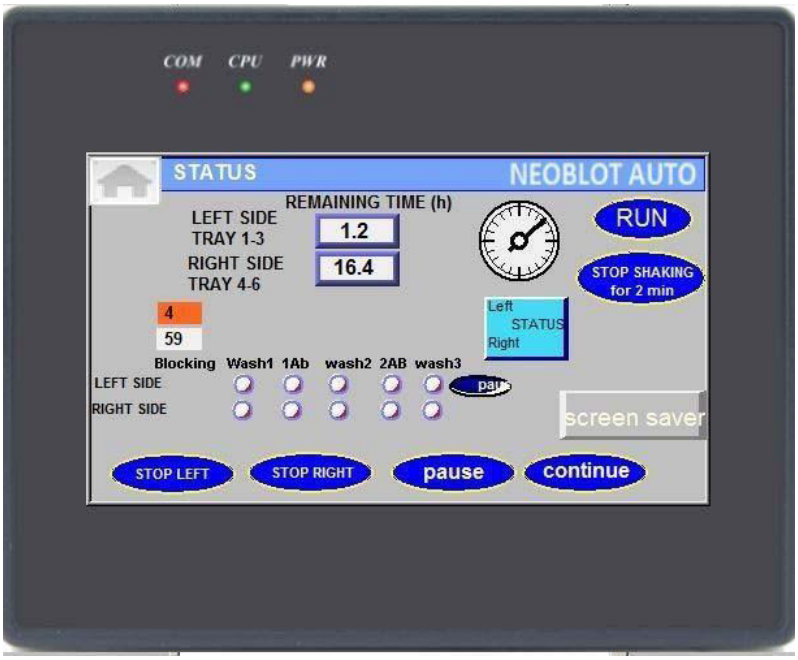
button.

B4. Pause Cycling

- you can pause protocol by pressing 

button. Shaking will not stopTo continue protocol press 

button for left or right side.



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

Use 

to stop shaking while loading blot into trays. Protocol on other side will not stop, shaking will resume in two minutes.

To completely stop protocol use 

or 

buttons

B5. Cleaning

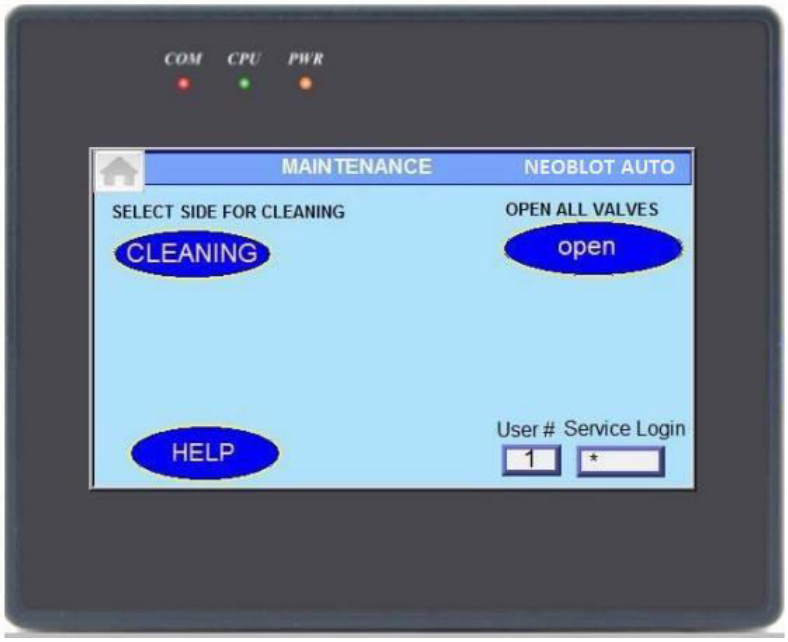
Note: you can do a cleaning only after both sides have finished cycling.

- Remove all membranes and containers for primary antibodies.
- Fill the device with cleaning solution up to Max Level (if you want a shorter cleaning cycle you can add less buffer, but at least 2 L).

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

- Go to the home screen and press 

button, a new screen will appear:



Press 

and select left or right side or both side on pop-up window.

Note: if you select both, left side starts first and the right side will continue after cleaning on the left side is completed.

Note: to remove the excess of cleaning solution from the tank you can repeat the cleaning cycler or justremove the plug located between the pumps.

Note: you can stop cleaning at any time but need to restart NeoBlot Auto before starting a new protocol.

B6. Tray size setting

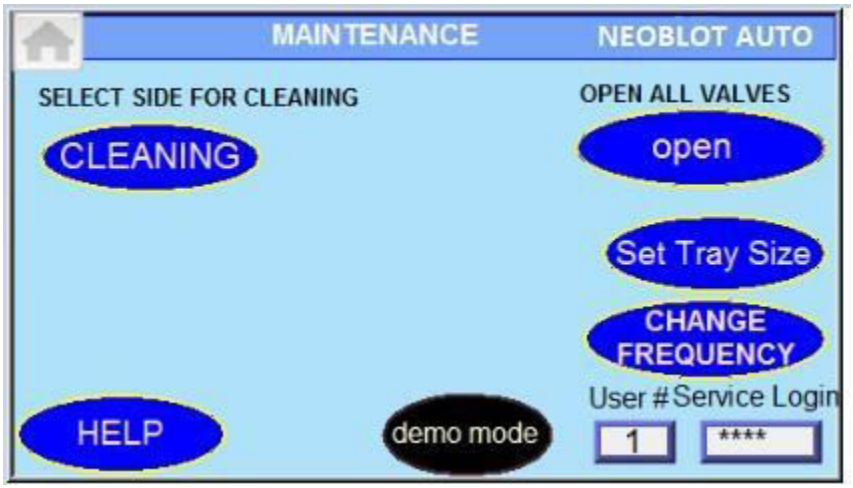
Note: Finish all protocols and remove liquid before next step. Refer to separate instruction regarding tray replacement

Go to the home screen and press 

button, a new screen will appear. Touch service login and enterpassword *4443*.

The new buttons appear: 

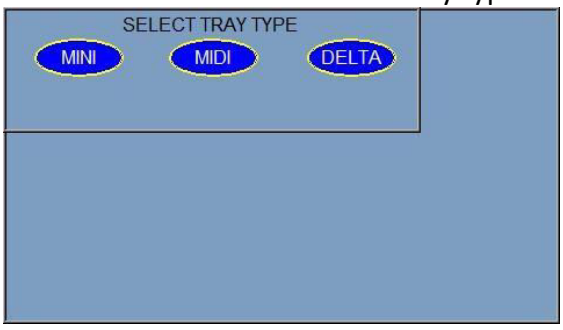
and 



Press 

button:

Select the side and then select tray type:



Go to home scree and press STATUS button and then select 

. Make the correct tray are selected.

B7. Frequency selection (Japan only)

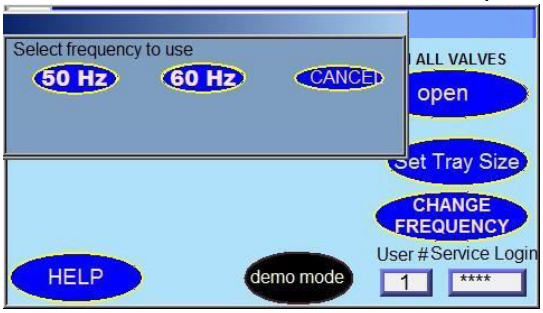
To select correct frequence for the area go to the home screen and press 

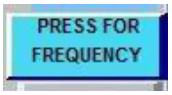
button, a new screen willappear. Touch service login and enter password *4443*.

Press 

button.

On the new screen select correct frequency for your area:



Go to home screen and press STATUS button and then select 

Verify that correct voltage frequency selected.

B8. NeoBlot Auto Maintenance:

Run cleaning cycle every time you finished western blot processing. You can use deionized or distilled water for cleaning. At least once in a week perform intensive cleaning with cleaning solution (cat #CL500).

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

Go to home screen and press 

button and then 

button.

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

Note: before opening valves check that there is no solution in the tank or the level close to minimal level.

- Now turn NeoBlot Auto off.

Note: after opening valve you need to restart NeoBlot Auto using switch on the back beforeusing itagain.

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 offthe 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 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 thatcorrect power cord is used and the fuse is replaced, contact Technical support.
Buffers leak from the trays and tank immediately	Valves remain open.	Turn instrument off, wait at least 5 sec and turned instrument on. After initialization valveswill 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 detectionreagents 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, stainmembrane 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 ofprotein being blotted. After blotting, stain membrane to measure transfer efficiency.
	Incorrect reagents added or incorrect containers are filled	Make sure that primary and secondary antibodyare 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 gelor 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.Determine antibody activity by performing a serial dilution using six trays or dot blot.
	Inactive or overly dilute primaryor secondary antibody	Increase antibody concentration as necessary.Decrease exposure time or allow signal to further decay. Prevent leakage of solutions byencasing membrane in transparency film and blotting excess substrate from edges before exposure.
High background on the blot	Film overexposed or became wet during exposure	

	Short blocking time or washingin-tensity	Increase blocking time and the number of washes
	High concentration of primary and/or secondary antibody	Determine optimal antibody concentration by performing dilution series using all six trays. De-crease antibody concentration as necessary.
	Protein is overloaded	Reduce load or dilute concentration of sample.
	Membrane, solutions, trays, or antibody containers are con-taminated	Use clean glassware and purified water to pre-pare solutions. Wear clean gloves at all times. Use forceps when handling membranes.Run cleaning protocol with cleaning buffer, increase the con-centration of cleaning buffer two times
	Protein is overloaded	Reduce load or dilute concentration of sample.
Non-specific bind-ing 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

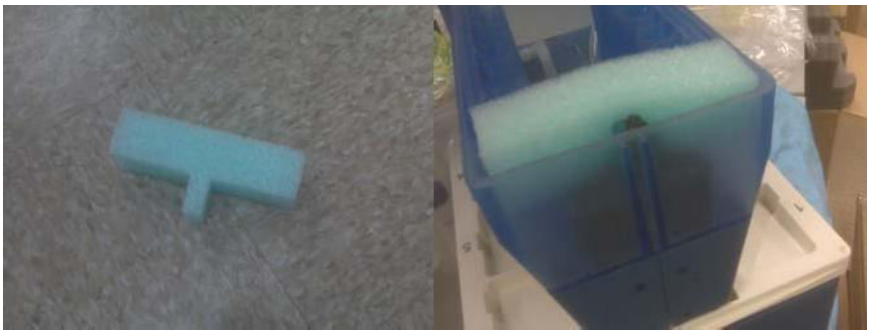
Follow the instructions below to replace the 250V, 5A rated fuse for the power socket.

1. Turn off NeoBlot Auto using switch on the back of the instrument and detach the power cord from therear ofthe instrument.
2. Open the fuse compartment located on the power entry block using a small flat blade screwdriver orfingernail 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 thefuse element, replace the 250 V, 5 A 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.

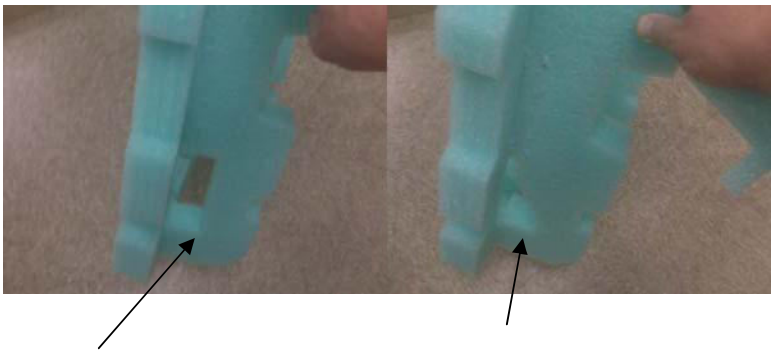
Repackaging the Instrument

Find an original box; take out an insert a flat box (originally contained tray and tank covers and dummy vials). Find a small T-bar and place between pumps into the tank.



(If you cannot find T-bar, use any appropriate material to secure pump inside tank)

There are two green Styrofoam covers: front and back (they are slightly different):



Back Front

Remove tray and cover and place them in a flat box (see below). Put on front and back sterofoam covers on theinstrument. Make someone help you to make sure that instrument is not flip over.

Using upper part of green Styrofoam as handles put NeoBlot Auto in the box:

Place insert flat box over instrument (if you cannot find flat box use any soft material to fill the gap be-tweeninstrument and box top surface):

Close and tape the box.

Ship to:
Neo Biotech
74 rue des suisses
92000 Nanterre
France.

Accessory Products

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

Description
Antibody collection vial per 100
Antibody collection vial per 50
Cleaning solution 50x, 500 ml
Cleaning solution 50x, 500 ml, 5 bottles
Cleaning solution 50x, 500 ml, 10 bottles
Hybridization buffer, 500 ml
Washing buffer 10x,1L
Washing buffer 10x 4L
Pair of Block of mini trays right & left
Replacement Block of midi trays left
Replacement Block of midi trays right
Pair of Block of midi trays right&left
Replacement tray for NeoBlot Auto-Flex, midisize
Tray cover, clear
Tank cover clear
Instrument support stand
Tray plug, bag of 6
Drain (dummy) vial (pack of 6)
Tray tubing replacement kit
Tank tubing replacement kit
Reagent Collector Insert
Electrophoresis unit, minigel size with power supply

Warranty

The NeoBlot Auto is warranted for one (1) year against defects in materials and workmanship. If any defectsshould occur during this warranty period, Neo Biotech will repair and replace the defective parts without 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
Fax: +33977401011
Email: info@neo-biotech.comWrite :
Customer Support
Neo Biotech
274 rue des suisses92000
Nanterre
France