Instruction Manual

Blotting System
Built-in Power supply
WSE-4115
poweredBLOT Ace

1st Edition February 19, 2016



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Introduction

Thank you for purchasing ATTO Corporation's blotting system built-in power supply "WSE-4115 powered-BLOT-Ace". This instruction manual (i.e. this document) is delivered to you together with the system so that you can make full use of the system. Not only those of you who use this system for the first time, but also those who have used it before, should read this document carefully to understand the contents. If you use this system for the first time, please read this document from the beginning in serial order. In addition to how to use it, this document contains information related to maintenance and guarantee as well. Please keep it handy all the time to make its full use. If you have any inquiries on your purchased product or the instruction manual, please feel free to contact your distributor or us (Please refer to the back cover).

About this manual

Before using the product, please read this document carefully. After reading it, please be sure to keep it for your future reference. When you relocate this system, be sure to attach this document to it. If there is any defect in this document such as misplaced or missing pages, or if this document is lost or stained, we will replace it with a new one. Please take a moment to contact the distributor you purchased the product from or our company's customer service department (Please refer to the back cover). At that time, please inform us of your product name and model. This document was created with our most careful attention; however, should you find any queries, errors or omissions, please inform our company's customer service department (please refer to the back cover).

Safety precautions

To use this system safely, it is a must to operate it properly. Do not use this product until you read this document carefully and understand the content sufficiently. Precautions on usage and safety described in this document are applied to the use of this system only for the specified purpose of use. Do not use this system for any other purpose than described here, or do not use this system by any other method than described here. If you use this system for any other purpose or by any other method than described in this manual, you will be held responsible for all necessary safety measures as operator. If you operate the system for the first time, you need to be given instructions from an experienced operator with proper knowledge, and to understand its principle and method. Not only people who operate the system for the first time but also people who have ever used it after receiving professional education should keep this instruction manual handy to make its effective use. In order to prevent any electric shock or any damage to the system, please understand and follow the correct operation method shown in this manual. If you have questions or concerns related to the principle of the system, maintenance or inspection, feel free to contact your distributor or our company (Please refer to the back cover).

Safety symbols

To use this system safely and maintain the safe status, the following symbols are indicated in the instruction manual and on the system's main unit. Please note the meaning of each symbol and observe each item.

Symbol	Description
⚠Danger	This symbol indicates emergent danger, such as death or heavy injury caused by ignoring the symbol and mishandling the system.
This symbol indicates possibility of danger, such as death or injury, cause the symbol and mishandling the system.	
This symbol indicates possible occurrence of physical damage caused by ignoring symbol and mishandling the system.	
This symbol indicates prohibition.	
This symbol indicates an important matter.	
	This symbol indicates a tip related to operation.

Operation precautions

These are precautions for preventing fire, electric shock and other accident or failure. Read and understand the information well, and be sure to observe it.



Power supply connection	Do not use a deformed or corroded electrode terminal, AC adapter, power cable whose insulation coating is peeled off, or damaged power cable. Do not connect any power supply other than the power supply attached to this system to the system. Before operating this system, check and confirm that there is no damage to it. Otherwise, it may catch fire or cause electric shock due to loose connection. If there is any damage, stop using this system and contract your distributor or our company (please refer to the back cover). After use, be sure to turn off the power switch and disconnect it from the outlet. When disconnecting AC adapter from the outlet, be sure to turn off the power switch, and then disconnect it by holding the AC adapter instead of pulling the cable.
Not wet hand	When handling this system, keep your hands dry. Do not touch AC adapter or connection terminal with wet hands. If you do, electric shock or failure may be caused. If the power supply part or AC adapter gets wet, do not use it. If you do, electric shock or failure may be caused. If wetted, stop using it and contact your distributor or our company (please refer to the back cover).
Main unit	Do not put any foreign object into this system. If you do, electric shock or failure may be caused. If the external surface of this system gets wet, do not use it. If you do, If you do, electric shock or failure may be caused. When using it, wipe moisture off the surface and dry it.
Maintenance	If an error occurs or if there seems to be an error or failure while this system is being used, stop using it immediately. If you find any defect at the time of inspection, do not use this system. If you do, electric shock or defect may be caused. While this system is in use, check if there is any error, such as abnormal sound or smoking etc. by regular visual inspection. If you find an error, failure or defect, stop using it and contact your distributor or our company (please refer to the back cover).
Reagent	For the electrophoresis and blotting, deleterious substance, dangerous substance or carcinogenic material may be used for preparation of buffer solution, staining or decolorizing operation. Do not allow them direct contact to human body. If you do, fatal accident or body injury, like burn, may be caused. When using chemicals, protect your body with gloves and a mask. Carefully read and observe precautions on handling attached to the chemicals.

\bigwedge Warning

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Installation	Do not install the system on an unstable table, tilted place or a heavily vibrating place. Install it on an experimental table with horizontal, stable and solid surface. Otherwise, electric shock due to falling or liquid leakage may be caused. Do not put any object on this system. If you do, electric shock due to falling may be caused.
Main unit	This is not of explosion-proof structure. Install it at the place where there is no exposure to fire or combustible gas. When taking this system out of a low-temperature room for use, take measures against dew condensation before moving it. If condensation is seen, dry it completely. Otherwise, electric shock or failure may be caused.
Transfer	While this system is in operation, do not touch any parts other than control panel, nor move it. Also, electric cords may get tangled and the system may fall. When moving this system, be sure to turn off the power switch and disconnect AC adapter, and then disconnect all wiring cables.
Maintenance	When you conduct maintenance or cleaning, be sure to turn off the switch of the power supply and disconnect AC adapter. To maintain good performance and safety of this product, please ask us for periodical maintenance, inspection and calibration (please refer to the back cover).
Disassembly	Do not disassembly or remodel the system. Do not remove the external cover. Adjustment or repair of this product should be made by our engineers. Our company will not accept any responsibility for any accident or failure caused by disassembly or modification done by you.
Sticker	Do not peel off the warning stickers. They indicate a dangerous section of this system. If it is peeled off or cannot be read due to stain, please contact us, referring to the back cover.

Maming

AC adapter

Do not use the power supply part and AC adapter of this system for any purpose other than operation of this system. If you do, failure or accident may be caused.



We will not accept responsibility for any accident or failure caused by using the power supply and AC adapter of this system for any system other than this one.

If this system is used outside Japan, prepare the conversion adapter complying with standards of the country where you use it, If a non-standard adapter is used, heat generation or ignition may occur. Also, AC adapter of our product in the below cannot be used for substitute.

[AE-6685、WSE-4110]

If you use, heat generation and failure of AC adapter or power supply may be caused. If you have any inquiries, please contact your distributor or us (please refer to the back cover).

If any AC adapter other than one attached to this system is used, it may cause following accident or failure; defective operation or trouble of main unit or power supply part, degradation or failure of AC adapter, or ignition.

Be sure to use AC adapter attached to this system, if you operate this system.

.↑Caution

Sticker

The name plate stickers show important information for maintenance and management of product. Do not peel it off.

The system has labels like the below on the surface for safety handling. Do not peel them off or make them dirty.



Warning label (Back of power supply part)



Notices

Application	This system is physical and chemical equipment for research. It is not medical equipment. Therefore, it cannot be used for medical practices, such as medical care-related judgement or treatment effect checking.
Export	Export of certain services or cargos is controlled by the foreign exchange law and the government decree or ministerial ordinance of foreign trade control law of Japan. The product is subject to such regulations. Even if the product is not pertinent to the government decree, it is necessary to submit the document to the customs office to that effect, if the product is pertinent, it is necessary to obtain the export license from the Ministry of Economy, Trade and Industry and submit the license to the customs office. When you export our company's product, please contact the distributor or our company's customer department in advance (please refer to the back cover).
Trademarks/ Copyright	Reprint or copy of a part or whole of the instruction manual would require the permission of the copyright. The specification of the product and the contents of the instruction manual may be changed without prior notice.

1 Overview

1.1 Purpose

WSE-4115 poweredBLOT-Ace is semi-dry blotting device for transferring sample to membrane from polyacrylamide gel of compact (60x60mm) and mini (90x80mm) gel size after electrophoresis.

2 Inspection when unpacking the product

2.1 Inspection at unpacking

When you receive the product, check if the main unit and the accessories are properly packed and there is no damage. If you find any defect or damage, please contact the distributor you purchased the product from or our company immediately (please refer to the back cover).

Carry out the inspection at the time of unpacking within one week after you receive the product. After one week elapses, damage or parts shortage may not be recovered.

2.2 Equipment configuration

The product consists of main unit and accessories,

Main unit

WSE-4115 poweredBLOT-Ace

Model	WSE-4115	WSE-4115 • C	WSE-4115 • M	
Product No.	2322490	2322491	2322492	
		Base unit		
Main unit Catho		Cathode unit		
	Dedicated	power supply for electrophoresis		

Material

Titanium plated platinum
Corrosion-resistant stainless steel
Acrylic
Acrylic
ABS
ABS

Accessories

Model	WSE-4115	WSE-4115 • C	WSE-4115 • M		
AC adapter		1			
Extension cable		1			
Roller		1			
Instruction	on 1				
Manual					
Membrane	-	WSE-4050 P plus membranes 65×65mm 20 sheets	WSE-4051 P plus membranes 85×90mm 20 sheets		
Filter	-	CB-06A Filter paper 65×65mm 400 sheets	CB-09A Filter paper 85×90mm 400 sheets		

3 Part name and function

3.1 Main unit

This system consists of (1) Power supply part, (2) Cathode unit, (3) Base unit.





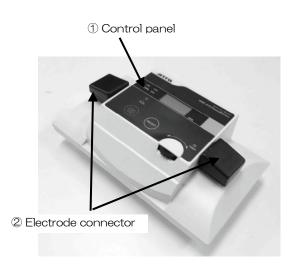


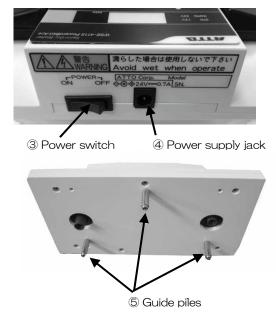
(1) Power supply part

Electrical power is supplied between cathode plate on cathode unit and anode plate on base unit under the condition and time which are set on control panel.

By timer function, supplied power stops automatically in running time set by control panel and then alarm rings.

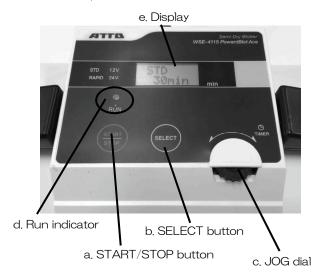
It supplies power to output is 12V, 24V by 2 switching system.





1 Control panel

This part is for selecting output mode, setting running time, operating to start/stop and displaying selection mode, output condition and time.



a. START/STOP button

Press the button for starting a run. Run indicator (RUN) blinks during a run. Press it again, and running stops

b. SELECT button

Press the button for selecting output mode, Refer to "Type of output mode" in the below for detail.

c. JOG dial

The dial is for setting running time. The measure is min. Turn the dial clockwise or counterclockwise for

setting the time shown on display. The range is 1-250 min.

d. Run indicator

The indicator blinks during running with green light, When Short/Open error occurs, blinking becomes quicker. The indicator lights off at the all time other than the above

e. Display

Selected output mode is shown on the upper of display, and running time is shown on the lower of display. Measures of time is minute. While running, remaining time is displayed on count-down basis. When running is over, "End" is shown, on the other hand, "Short Err" or "Open Err" is displayed at the time of Short/Open error.

f. Memory function

This function is to memorize running time of each mode which is set. The mode and time of the last operating are memorized, then the condition is displayed when the power is turned on. The condition is memorized when push SELECT or START/STOP button.

g. Output limit function

While running, the current value under constant voltage condition rises according to change of electrical resistance value in the gels or buffer etc. If the current value rises to the maximum value within running, the condition is transferred from constant voltage output to constant current output.

* There is no a function showing current value.

	ype	ot	<u>output</u>	mod	e
_					

There are 2 output mode like the table on right.

Press SELECT button for changing mode.

If blotting buffer for high-speed blotting is used, select "RAPID".

If standard blotting buffer is used, select "STD".

Mode	Output	Time
STD	12V	30~60
RAPID	24V	5~30 min

2 Electrode connecter

Connect it to upper electrode plug of cathode unit. Set power supply part on the predetermined position of cathode unit, push the connecter. Power supply part connects to cathode and anode plate and it can run.

3 Power switch

Push "ON" side for switching on or "OFF" side for switching off. When the switch is on, output mode and setting time are shown on the display.

4 Power supply jack

Connect it to AC adapter attached to this system (24V DC 1.5A).



Do not use any AC adaptor other than the one attached to this system. If you use it, it may cause following accident or failure: defective operation or trouble of main unit or power supply part, degradation or failure of AC adapter, or ignition.

⑤ Guide piles

Insert them in cathode unit when it is set.

(2) Cathode unit

This unit has cathode plate which can be used with a base unit, If assembling power supply part, it is cathode (-).

1)Guide holes

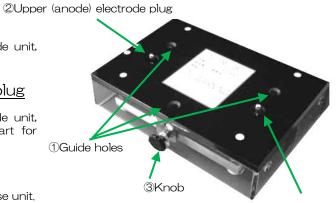
When power supply part is attached to cathode unit, insert guide piles in the holes,

2Upper (cathode/anode) electrode plug

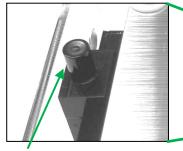
After power supply part is attached to cathode unit, press electrode connecter of power supply part for connecting.

3Knob

Press a knob for removing cathode unit from base unit.

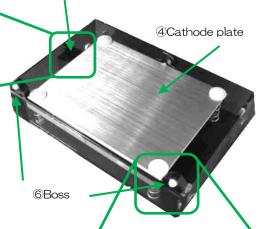


2Upper (cathode) electrode plug



©Upper (anode) electrode terminal (Enlarged view)

©Upper (anode) electrode terminal



4Cathode plate

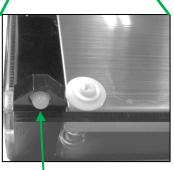
This is electrode plate which is made of chemical resistance stainless steel, and it would be cathode.

©Upper (anode) electrode terminal

This is electrode terminal of cathode unit which is connected to lower (anode) terminal.

6Boss

When cathode unit is set on base unit, insert this part in hole of base unit.



6Boss (Enlarged view)

(3) Base unit

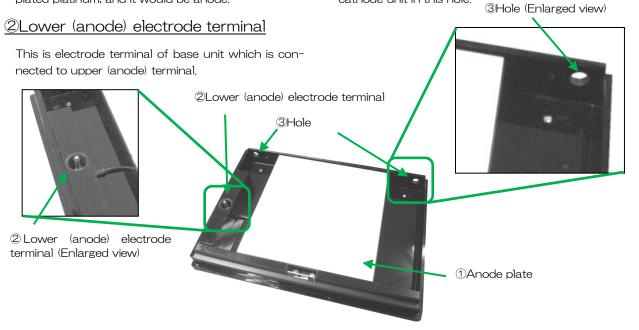
This unit has anode plate which can be used with cathode unit. If assembling power supply part, it is anode (+).

1 Anode plate

This is electrode plate which is made of titanium plated platinum, and it would be anode.

3 Hole

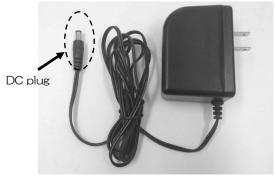
When cathode unit is set on base unit, insert boss of cathode unit in this hole.



3.2 Accessories

AC adapter (24V DC 1.5A)

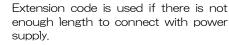
AC100V to 240V is available. Supply DC24V.



 AC adapter of AE-6685 or WSE-4110 is not available for WSE-4115 system.

Roller

Roller is used for removing excess blotting buffer and left bubbles in filter, membrane and gel.



Extension code



4 Preparation

4.1 Installation environment

Please use this system under the following condition.

Location	Inside a room only
Ambient temperature/	4~40°C • 5~70%RH (No dew condensation)
Relative humidity	

√Waming

Do not install the product in combustible gas atmosphere. It is not of explosion-proof structure, so the product may cause explosion or fire. Install it in an environment that does not contact combustible gas.

Do not install the product in corrosive gas environment. This is because it can cause corrosion of conductor inside this product or contact failure of connector, which may lead to malfunction or failure.

Do not install the product in an environment with much dust or dirt. Dust or dirt may get attached to the product, which can cause electric shock, fire or failure.

♠Caution

Do not use the product at a place where there is strong magnetic field or electric field around, or a place where there is much waveform strain of input power supply or noise. Malfunction may be caused,

function may be caused. Do not install the product where it is exposed to direct sunlight, where temperature suddenly changes, or where humidity is high. If dew condensation occurs, do not use this product. This system cannot be used outdoors. It is designed so that safety and performance can be ensured under the environmental conditions; ambient temperature 4°C~40°C, relative humidity

4.2 Preparation of peripheral system and consumables

Prepare membranes and filters applied to gel size.

 $5\%\sim70\%$ (No dew condensation).

Mini gel (85x90x1mm thickness)		Compact gel (60x60x0.75mm thickness)
Membrane	WSE-4051 P plus membranes (85× 90mm)	WSE-4050 P plus membrane (65 \times 65mm)
Filter	CB-09A Filter paper (85×90mm)	CB-06A Filter paper (65×65mm)

^{*}The number of filters varies according to blotting buffer you use.

4.3 Preparation of reagent

1. Required reagent

Prepare the below reagent in accordance with purpose.

- 1) Blotting buffer
- 2) Primary antibody and Enzyme labeled secondary antibody
- 3) Blocking reagent
- 4) Wash buffer
- 5) Detection agent
- 6) Stripping reagent

We can provide the products described in [7.4, Consumable goods] (Page 26) so please utilize them for your purpose.

For products detail, please contact our company.

If you use our blotting buffer AE-1460 or WSE-7120 (Refer to [4.3 Preparation of reagent]), prepare 3 pieces of filter for each electrodes, 6 pieces in total. If you use our blotting buffer AE-1465, prepare 2 pieces of filter for each electrodes, 4 pieces in total.

^{*}The number of filters is supposed to use our 0.9mm thickness filter. If you use another filter whose thickness is different, refer to the above amount and adjust it to the same thickness as the above.

2. Preparation

Refer to manual, article and so on of each reagent for preparation. It describes simply how to prepare with mainly our reagent in the below. Also refer to instruction manual attached to each reagent.

	Model	Name	Method	Amount	
Blotting	AE-1460	EzBlot	Add methanol 25mL each to unopened Bottle A-C.	50mL each in A-C.	
	AE-1465	EzFastBlot	Dilute in 1/10 with distilled water.	Approximately 200mL	
buffer	WSE-7210	EzFastBlot HMW	Dilute in 1/5 with distilled water.		
	WSE-7055	EzRun TG	Dilute in 1/10 with distilled water. Add methanol for adjusting the density to 5-20% in accordance with purpose.	Approximately 200mL	
	AE-1475	EzBlock Chemi	Dilute in 1/5 with distilled water.	650uL diluted solution	
Blocking	AE-1476	EzBlock BSA	In the case of AE-1476/77, add	per 1cm ² of membrane. In the case of Mini gel,	
buffer	AE-1477	EzBlock CAS	1/100 the amount of Tween 20 which is attached.	approximately 50mL is needed.	
	WSE-7230	EzTBS	Dilute in 1/10 with distilled water. As required, add detergent	100mL diluted solution (approximately)	
Wash buffer	WSE-7430	EzPBS(-)	such as Tween 20 or TritonX- 100 for adjusting the density to 0.02-0.5%.		
Detection agent (Coloring)	AE-1490	EzWestBlue	Use it without distilling,	100uL per 1cm ² of membrane. In the case of Mini gel, approxi- mately 10mL is needed.	
Detection agent (Coloring)	WSE-7110	EzWestLumiOne		In the case of Mini gel, 5mL is needed for each piece.	
	WSE-7120	EzWestLumi Plus	Just before detection, mix Reagent A and B Reagent in the ration of 1:1.	In the case of Mini gel, 2.5mL of each A and B per a piece.	
Stripping reagent	WSE-7240	EzReprobe	Use it without distilling. As required, add 0.6g [Enhancer] which is attached per 100mL.	30mL per a sheet of Mini gel size membrane. Also wash buffer is needed.	

5 Operation

This chapter explains how to operate the system with our high-speed blotting buffer [EzFastBlot].

5.1 Preparation of device

If cathode unit, base unit and power supply unit are assembled, separate them as per the following at first.

€ Caution

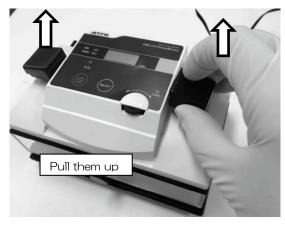
Before pulling out DC plug, disconnect AC adapter from outlet. It may cause accident such as electric shock or injury.

Turn off the power switch of power supply part. Before that, pull out DC plug of AC adapter from power supply part.

Pull up 2 electrode connecters fully.

Lift power supply part and remove it from cathode unit.

Keep pressing knob of cathode unit and lift the near part of it for opening.

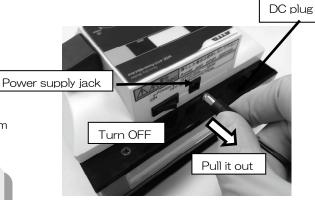




Remove cathode unit from base unit.

From this step, keep it away from base unit and operate the system until filter, membrane and gel finished to be set.





5.2 Pretreatment

Prepare containers for membrane, filter and gel each.

1 Pretreatment for membrane (Hydrophilizing)

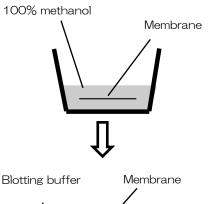
Sink P plus membrane in 100% methanol gently from the corner, and immerse it completely for about 20 seconds. After taking it out, sink membrane in blotting buffer and shake it for more than 5-10 min

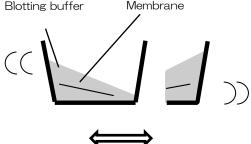
- *Do not touch membrane with bare hands, Put on clean gloves for experiments or hold it with tweezers,
- *Complete this step before the end of electrophoresis.
- Shake the container strongly and immerse membrane completely because membrane repels blotting buffer in the beginning. Insufficient shaking tends to result in uneven blotting,
- XUse same membrane size with gel.

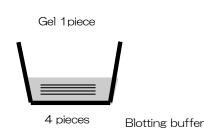


Prepare 4 pieces of filter. Immerse all filter in blotting buffer completely.

- *Complete this step before the end of gel electrophoresis.
- *Use adequate amount of blotting buffer for immersing filters completely.







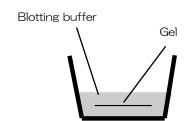


If you do not use our filter, CB-06A/09A, prepare filter whose thickness is 0.9mm, or adjust it to the same thickness and then cut with same size of gel. If the thickness and size is not even, it may result in failure or the display panel would show " $\rm Err$ " and it is not possible to operate.

3 Pretreatment for gel

After electrophoresis, immerse gel removed from glass plates with blotting buffer.

If gel is immersed for long time, gel would expand or blotting efficiency is decreased. Also display panel would show "Err" and it is not possible to operate.



5.3 Setting filter, membrane and gel

Drop about 1mL of blotting buffer on the anode plate.



Put on clean gloves for experiments to operate safely.



Take out 2 sheets of filter immersed in blotting buffer and drain the solution.

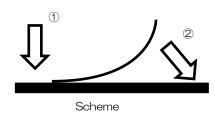


When filter is taken out of the container from this, do the same operation.

Put filter on the center of anode plate. For preventing air bubbles from entering between anode plate and filter, <u>put it from the edge of filter at first (1)</u> and then put whole of it slowly (2).









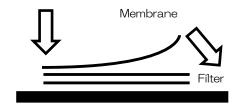
Filter must be put on the center of anode plate. If filter is out of the center, it may result in failure or the display panel would show "Err" and it is not possible to operate.

If air bubbles enter between the plate and filter, blotting may be failed. Follow the above method to preventing air bubbles from entering. Also follow the same method when you stack filter.

Stack a membrane on the second filter in order to align the edge of layers straight.

%Prevent air bubbles from entering between filter and membrane.

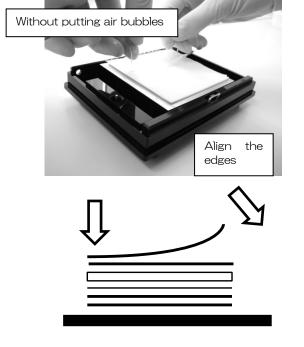
Drop about 1mL blotting buffer on the membrane.



Stack gel washed with blotting buffer on the membrane.

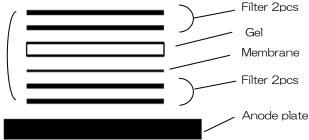
- *Whole area of gel including sample wells should be the membrane area.
- *Prevent air bubbles from entering between membrane and gel.
- **Gel should not be rearranged on membrane because sample may be transferred to membrane just with touching gel.

Stack 2 pieces of filter immersed in blotting buffer on gel in order to align the edge of layers straight without air bubbles.



Finally, filters, membrane and gel are stacked as the right picture. This construction is called "Sandwich".

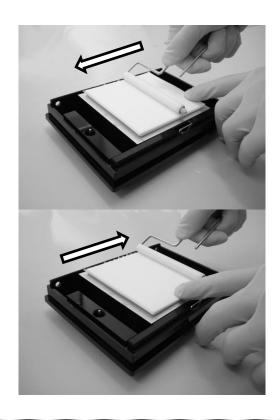
Sandwich



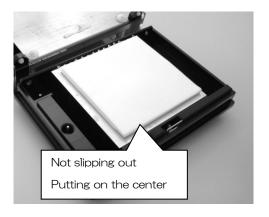
Hold a edge of sandwich for preventing from slipping out and use a roller to remove air bubbles and excess blotting buffer from the holding position.

Do the same method from the opposite direction again.

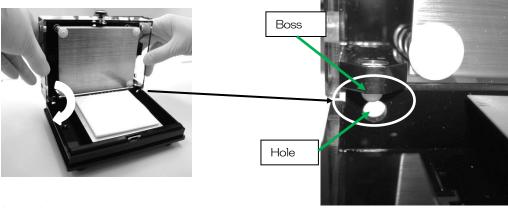
- *There is no problem even if the blotting buffer overflows.
- If there is a weak a stick between gel and membrane, it will be reason of blotting pattern flow.



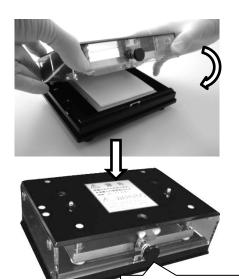
Confirm if filter, membrane or gel is not slipped out from sandwich and if the sandwich is put on the center of anode plate.



While holding cathode unit at more than right angle to base unit, match bosses of cathode respectively with holes of base unit.



Gently lower the opened front of cathode unit to shut it down, and lightly push down cathode unit until knob clicks.



Until "Tick!".



Do not remove cathode unit until operation is over. If assembled units are reopened, sandwich is collapsed. Sample on the membrane may be transferred just with touching gel.

5.4 Assembling power supply part

Confirm if main unit is powered OFF.

Insert 3 guide piles at the bottom of power supply part in the holes of the cathode unit.

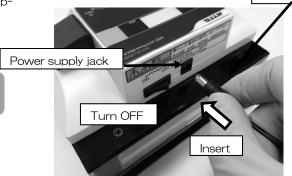
Push down 2 electrode connecter to cathode unit,



Insert DC plug of AC adapter to power supply jack at the back of power supply part.



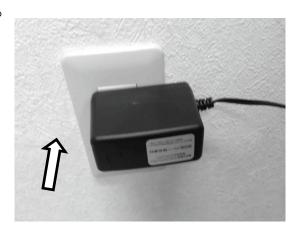
Do not use any AC adapter other than one attached to the system.



Caution

Do not use any AC adapter other than one attached to the system. If you use it, it may cause following accident or failure; defective operation or trouble of main unit or power supply part, degradation or failure of AC adapter, or ignition.

Connect AC adapter or extension code to outlet.

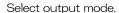


5.5 Power on

Turn on the power switch.

In this time, display shows the condition set on the last time by memory function.

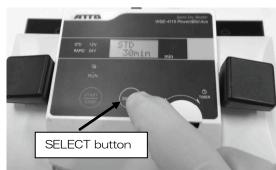
※If the condition is same as last operation, it does not need to reset.



Press select button and select "STD" or "RAPID".

Refer to the below table about blotting buffer, mode and time for selecting.





Blotting buffer	Gel	Amount	Output mode and Time		
Diotting burrer	Gei	of filter	STD(12V)	RAPID(24V)	
AE-1460 EzBlot	Hand-made gel	+,- 3 pcs	30~60 min	-	
AE-1400 EZBIOL	ATTO precast gel	each			
AE-1465 EzFastBlot	Hand-made gel	+,- 2 pcs	20~60 min	10~25 min	
AE-1465 EZFASIBIOL	ATTO precast gel	each		8∼15 min	
WSE-7210 EzFastBlot HMW	Hand-made gel	+,- 3 pcs	30~60 min	15∼30 min	
VVOL 7210 EZI ASIBIOTI IIVIVV	ATTO precast gel	each	30: -00 111111	10 00111111	

**The amount of filter on the table is supposed to use our 0.9mm thickness filter. If you use another filter whose thickness is different, refer to the above table and adjust it to the same thickness



Set suitable time with referring to preliminary experiment, In order to max blotting efficiency of target sample, it is necessary to extend a time. If it takes long time to operate, filter and membrane may get dry .Please keep this point in mind.

Set a time.

Press JOG dial and set output time.

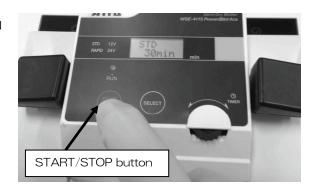
Turn JOG dial to set target time (1-250min).

(Right direction: Increase

Left direction: Decrease)



Press "START/STOP" button to start. RUN indicator blinks. Display shows remaining



While electricity is supplied, RUN indicator blinks. Display shows present mode and remaining time on countdown basis. If SELECT button is pressed while electricity is supplied, display shows set time while the button is being pressed.

- If START/STOP button is pressed while electricity is supplied, it stops. If it restarts after stop, press this button again. In this time, elapsed time till stop is reset so remaining time should be set again.
- If abnormality (Short or Open error) is detected while electricity is supplied, it stops with alarm and the message "Short Err" or "Open Err" blinks on the display. RUN indicator blinks more quickly than usual. Turn off the power switch and unplug AC adapter from outlet, Refer to the below and "Trouble shooting" (Page 22) for solving a problem.



While abnormality is detected, display shows "Short Err" or "Open Err".



RUN indicator blinks quickly

Open error (No load)	Both electrode connecters are not pushed down sufficiently.	Push down electrode connecters well.	
When electricity is not supplied "Open Err"	Cathode plate does not touch sandwich sufficiently.	Confirm amount of filters and thickness. If you use filters other than ours, use filters whose thickness is 0,9mm or adjust the amount to the same thickness.	
	Breaking of wire	Stop using immediately and contact your distributor or our company (Refer to the back cover).	
Short error When excessive	Power plug gets wet.	Wipe off water attached to power plug.	
current flows "Short Err"	The interior of power supply part gets wet.	Stop using immediately and contact your	
	The circuit of power supply part may be broken.	distributor or our company (Refer to the back cover).	

5.6 Finishing operation

When set time passes, electricity stops automatically and the display shows "End" with alarm.

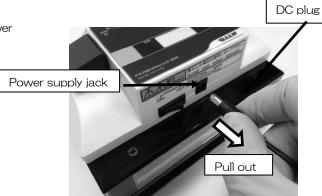
Turn off the power switch of power supply part. Confirm the display and RUN indicator put the light off.

Disconnect AC adapter or extension code from outlet. Do not keep them on connecting.





Pull out DC plug of AC adapter from power supply jack.

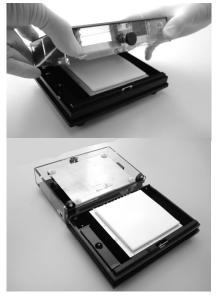


Pull up 2 electrode connecters for removing power supply part from cathode unit.



While knob of cathode unit is pressed, hold up the unit and then remove it.

**Filter or whole of sandwich may attach to cathode plate. When cathode unit is held up, pay attention to their falling down.

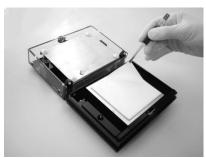


Remove filter and gel from the top of sandwich to bottom, and collect membrane. Go to next step, specific detection by antigenantibody reaction, coloring and so on.

* Do not reuse filters, It may be caused deterioration of blotting efficiency or nonuniformity.



Put clean gloves for experiments when you operate with membrane.



5.7 Detection

This chapter explains how to detect briefly. For more detail, refer to instruction manual of reagent or article.

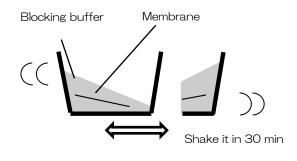
1. Blocking

Immerse membrane in blocking buffer soon after blotting and shake it in 30 min at the room temperature (50mL/gel).

※If it takes long time (more than 1 hour) for blocking, it may cause over-blocking.

2. Antibody reaction

- 1) Dilute primary antibody with EzTBS including 0.1% Tween or EzPBS (10mL/gel).
 - ※Dilution rate of antibody varies depending on antibody. Refer to the manual attached to each antibody. In general, it is several hundred times ∼ several thousand times dilution.



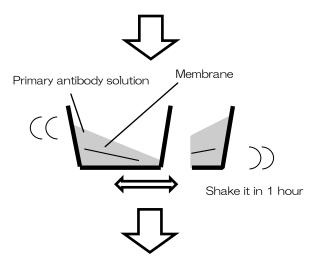


- 2) Pour antibody solution of 1) to the container which is a size larger than membrane, and shake it in 1 hour at the room temperature.
 - **Reaction time of antibody and temperature vary depending on antibody. Refer to the manual attached to each antibody. In general, shake it in 30min to 1 hour at the room temperature or 37°C, or in a night at 4°C.
- 3) Discard antibody. Add 50mL of EzTBE (TBS-T) including 0.1% Tween and shake it in 5 min (cleaning operation). Repeat the cleaning operation more than 3 times.
- 4) Dilute HRP mark secondary antibody with EzTBS (TBS-T) including 0.1% Tween or EzPBS(10mL/gel)
 - **Dilution rate of antibody varies depending on antibody. Refer to the manual attached to each antibody. In general, it is several thousand times ~ several tens of thousand times dilution.
 - *If there is high background, increase dilution rate or add blocking agent diluted in 1/10 to antibody diluted solution for reducing it.
- 5) Add antibody of 4) to membrane after cleaning, shake it in 30 min at the room temperature.
 - **Reaction time of antibody and temperature vary depending on antibody. Refer to the manual attached to each antibody. In general, shake it in 30min to 1 hour at the room temperature or 37°C, or in a night at 4°C.
- 6) Discard antibody, Add 50mL of EzTBS (TBS-T) including 0.1% Tween and shake it in 5min (cleaning operation). Repeat the cleaning operation more than 3 times,
 - %In the case of too much cleaning operation, signal may be decreased.
 - In the case of insufficient cleaning operation, background may be high.

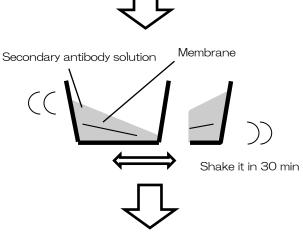
3.Detection

This describes how to detect with our WSE-7110 EzWestLumiOne for emission detection.

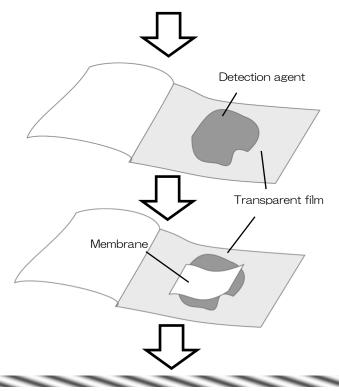
- 1)Pour detection agent to the clean container which is a size larger than membrane.
 - %Transparent film or plastic film is available to substitute for tray.
- 2) Immerse membrane after cleaning to detection agent. Confirm if the agent spreads to whole of membrane.
 - ※If membrane is not immersed evenly, it may cause physaliform background or nonunifromity of blotting.



Cleaning operation in 5 min. (More than 3 times)



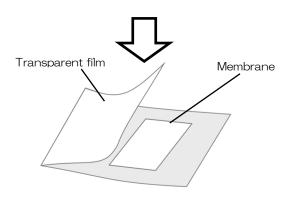
Cleaning operation in 5 min. (More than 3 times)



3) Sandwich membrane with transparent film or plastic film without air bubbles.

※If there are air bubbles between membrane and film, it may cause physaliform background or nonunifromity of blotting

*When membrane is sandwiched with plastic film, the wrinkle on the film affect the surface of membrane and it may cause wrinkle background or nonunifromity of blotting.



4) Photograph membrane reacting to detection agent by chemiluminescent imaging system.

We can provide consumable goods such as detection agent (Refer to Page 26). For more detail and catalog, please contact us (Refer to the back cover) or visit to our web page (http://www.atto.co.jp/eng).

5.8 Cleaning & Safekeeping the device

1.Cleaning the device

Clean cathode unit and base unit with running water before blotting buffer gets dry. Wash upper/lower electrode with soft sponge and neutral detergent, wash away a bubble and rinse them with pure water. After washing, dry them naturally.

White filter-shaped mark is on cathode plate but it can be removed by softly rubbing soon after the operation. Clean it with soft sponge gently.

Do not wash power supply part and AC adapter with water. Use soft cloth and neutral detergent diluted by water for cleaning it when it gets dirty. Do not use others organic solvent,

If anode/cathode plate gets dirty considerably, it may cause nonuniformity of blotting.

*Do not push the one side of cathode plate and tilt cathode plate greatly. It may cause deformation of inner screws.

2. Safekeeping the device

Do not store the device at where sunlight reaches directly, it may be exposed to corrosive gas or at high temperature.

6 Trouble shooting

Be sure to check not only the system but gel, blotting buffer, detection agent and the points relating to all operation from electrophoresis to detection. Please refer to not only this chapter but also instruction manual of the equipment or reagent relating to each operation.

Symptom	nptom Cause Solution	
Electric power is not supplied./ Voltage value does not rise./ Display shows error and electricity stops.	Composition of blotting buffer is incorrect. Electrode connecter connects imperfectly. Pretreatment of filter or membrane is insufficient.	Confirm composition of blotting buffer and method of preparation. If there is a difference or mistake, prepare it again (Refer to Page 10). Press both electrode connecters to the downward well. Immerse filters and membrane to each blotting buffer completely at the pretreatment. Shake the container strongly and immerse membrane completely because membrane immersed in methanol repels blotting buffer in the beginning.
	Breaking of wire	Stop using the device immediately and contact your distributor or our company (Refer to the back cover).
	Filter is reused.	It is impossible to reuse filter. Always use new one.

Symptom	Cause	Solution
Electric power is not supplied./ Voltage value	Cathode plate does not touch the sandwich sufficiently.	Confirm the amount of filters and the thickness, Adjust the amount as to the same thickness or confirm if it is 0.9mm thickness if you use our filter.
does not rise./ Display shows error and electricity stops.	Gel is immersed in blotting solution for a long time.	Do not immerse gel before blotting in blotting buffer for a long time (more than 5 min). Go to next step as soon as take it out from the solution (Refer to Page 12).
	Composition of blotting buffer is incorrect.	Confirm composition of blotting buffer and method of preparation. If there is a difference or mistake, prepare it again (Refer to Page 10).
	There are air bubbles in the sandwich.	Stack filter, membrane and gel not to enter even small air bubbles in the sandwich (Refer to Page 13-14).
	Pretreatment of filter or membrane is insufficient.	Immerse filters and membrane to each blotting buffer completely at the pretreatment. Shake the container strongly and immerse membrane completely because membrane immersed in methanol repels blotting buffer in the beginning.
Nonuniformity of	Sandwich is not placed on the center of anode plate. Put the sandwich on the center of anode plate.	
blotting / Dirty	Filter is reused,	It is impossible to reuse filter. Always use new one.
image	Stain or accretion is on the surface of anode plate or cathode plate.	Clean anode/cathode plate for removing stain and accretion. If the plate gets dirty considerably, contact to your distributor or our company (Refer to the back cover).
	Stacking method for filters is incorrect.	Check the number of sheets and stacking order of filter. If it is incorrect, make the sandwich again with new filter (Refer to Page 13 -14).
	Cathode plate does not touch the sandwich sufficiently.	Confirm the amount of filters and the thickness. Adjust the amount as to the same thickness or confirm if it is 0.9mm thickness if you use our filter.
	Trouble at the step of detection.	Contact the company which provides reagent used at the detection.
Band is invisible.	Blotting method is incorrect.	In the case of high molecular weight protein, this problem is improved by extending a blotting time or reducing gel concentration.
		In the case of low molecular weight protein, this problem is improved by increasing the amount of ethanol added to blotting buffer to one and a half time \sim twice.
Band spreads.	Gel and membrane do not stick to each other sufficiently.	After filter, gel and membrane are stacked, remove excess blotting solution and air bubbles by using an attached roller while holding the sandwich. If there is blotting solution between gel and membrane excessively, protein in the solution may diffuse and band may spread.

7 Maintenance

7.1 Cleaning

1. Power supply part

If the surface gets dirty, wipe by soft cloth with synthetic detergent diluted in water. If dust covers electrode connector, remove it so as not to damage the connector.



When you clean main unit, disconnect AC adaptor and turn off power switch. It may cause accident such as electric shock and injury.

2, AC adapter

When the surface gets dirty, wipe gently by soft cloth with synthetic detergent diluted in water. Do not use until it is dried sufficiently.



When you clean AC adapter, disconnect it from outlet. It may cause accident such as electric shock and injury.

3. Cathode unit

After operation, clean it with running water before blotting buffer gets dry. When the surface gets dirty, wipe gently by soft cloth with synthetic detergent diluted in water. Wash away stain on the cathode plate gently with soft sponge.

White filter-shaped mark is on cathode plate but it can be removed by softly rubbing soon after the operation. Clean it with soft sponge gently.

Do not soak it.

If electrode plug has dust, remove it without damaging the plug.

*Do not push the one side of cathode plate and tilt cathode plate greatly. It may cause deformation of inner screws



Before inspection of cathode unit, remove power supply part and base unit from it.

4. Base unit

After operation, clean it with running water before blotting buffer gets dry. When the surface gets dirty, wipe gently by soft cloth with synthetic detergent diluted in water. Wash away stain on the anode plate gently with soft sponge.

If electrode terminal has dust, remove it without damaging the connecter.



Before inspection of base unit, remove power supply part and cathode unit from it.

7.2 Inspection

Before using system which is retained after operation, inspect it with reading this instruction manual. If you find any abnormality at the inspection, do not use it and contact your distributor or our company.

1. Power supply

Confirm there is no damage, deformation or electrode connector corrosion by visual observation.



When you inspect power supply part, disconnect AC adapter and turn off power switch. It may cause accident such as electric shock and injury.

2. AC adapter

Confirm there is no damage, deformation, or insulation layer is not peeled off and damaged by visual observation.



When you inspect AC adapter, disconnect it from outlet. It may cause accident such as electric shock and injury.

3. Cathode unit

Confirm there is no damage, deformation or electrode plug corrosion by visual observation. Also, check if cathode plate does not get dirty, it moves up-down smoothly when pushed the center, and there is no difference at up-down motion of right and left by visual observation.



Before inspection of cathode unit, remove power supply part and base unit from it.

4. Base unit

Confirm there is no damage, deformation or electrode terminal corrosion by visual observation. When you clean it, check if anode plate is fixed certainly without looseness.

Caution

Before inspection of base unit, remove power supply part and cathode unit from it.

7.4 Warranty

ATTO Corporation warrants all its products subject to the terms and conditions set forth below.

- 1. This warranty covers all new products that are sold by ATTO Corporation (hereinafter called ATTO).
- 2. Expendable items are not covered by this agreement.
- Claims under this warranty are limited to defects in material and workmanship of the products.
- 4. Malfunction and/or damage due to neglect, abuse, operation or repair contrary to specifications and/or instructions presented by ATTO are not warranted.
- 5. ATTO shall not be liable to consequential damage, labor, loss or expense directly or indirectly arising from use of the products.
- 6. Damage due to transit is not covered by this warranty.
- 7. The warranty period is one (1) calendar year from a date when the products are shipped from ATTO to an original purchaser.
- 8. This warranty period is not applied to any defect that is reported to ATTO later than one (1) calendar month from a date od warranty termination.
- 9. ATTO shall supply parts to replace faulty parts of defective products under this warranty, free of charge.
- 10. ATTO shall repair defective products under this warranty, which cannot be repaired at field, free of charge.
- 11. ATTO shall replace defective products under warranty, which cannot be repaired, free of charge.
- 12. Freight charges for return and replacement shipments under this warranty are shared by ATTO and a purchaser, that is one way by either party and another way

by another party.

- 13. Warranty period of repaired products and replacement products or parts is three (3) calendar months from a date when the said products or parts are shipped from ATTO, or a remaining them of an original warranty period of the defective products, whichever lasts longer.
- 14. Return of the products for credit or refund is not accepted unless otherwise agreed in writing by ATTO.

7.4 Consumable goods

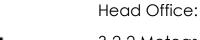
The following products are consumable goods. Please notify of the code number at the time of order.

Purpose		Model	Product name	Code no.
Blotting membrane		WSE-4050	6.5x6.5cm P plus Membranes 20/ pk	2322450
	Membrane for protein	WSE-4051	8,5x9cm P plus Membranes 20/pk	2322451
		WSE-4053	26x300cm P plus Membrane, 1 roll	2322453
	Membrane for nucleic acid	AE-6651	20x20cm N Membranes 10/pk	2392394
Dlattia e filtar	Dlatting filter	CB-06A	6.5x6.5cm Filter Paper 400/pk	2322437
Blotting filter	Blotting filter	CB-09A	8.5x9cm Filter Paper 400/pk	2392393
	Transfer buffer comprised of 3 reagent for semi-dry blotting	AE-1460	EzBlot	2332600
Blotting buffer	Transfer buffer for semi-dry and fast blotting system	AE-1465	EzFastBlot	2332590
	Semi-dry transfer buffer for high molecular weight protein	WSE-7210	EzFastBlot HMW	2332595
	Tris-Glycine buffer	WSE-7055	EzRun TG	2332323
	Non-protein blocking reagent	AE-1475	EzBlock Chemi	2332615
Blocking buffer	Bovine serum albumin block- ing reagent	AE-1476	EzBlock BSA	2332616
	Casein blocking reagent	AE-1477	EzBlock CAS	2332617
	Tris-buffered saline	WSE-7230	EzTBS	2332625
Wash buffer	Phosphate-buffered saline (Sterilized)	WSE-7430	EzPBS(-)	2332380
	10% Tween solution	WSE-7235	EzTween	2332626
	TMB substrate for HRP detection	AE-1490	EzWestBlue	2332630
Detection agent	Luminescence substrate for HRP detection	WSE-7110	EzWestLumiOne	2332632
	Luminescence substrate for	WSE- 7120S	EzWestLumi Plus	2332637
	HRP detection	WSE- 7120L	EzWestLumi Plus	2332638
Reprobe reagent	Stripping reagent	WSE-7240	EzReprobe	2332530
Molecular	Pre-stained molecular marker	AE-1450	EzStandardPrestainBlue	2332347
weight marker	3 colors pre-stained molecular marker	WSE-7020	EzProtein Ladder	2332346

8 Specification

Product name		poweredBLOT Ace		
Model		WSE-4115		
System		Horizontal semi-dry blotting		
Effective blotting a	urea	90 mm (W) x 85mm (D)		
Gel accommodatio	n	1 gel		
Effective electrode area Distance between electrodes Polarity		95mm (W) x 98 mm(D) Min 3mm , Max 10mm Cathode unit : Cathode Base unit : Anode		
Se ^r Tin	atput mode ax output tting ner arm	Constant Voltage 24W 12V, 24V 1~250 min Remaining time is displayed. Output stops/Time up/Error		
AC adapter Input Output		AC100V~240V (±10%) 50Hz/60Hz (±5%) DC24V 1.5A		
Safety measure		Safety cover, No load detection, Short circuit detection		
Location		Inside the room only $5\sim40^{\circ}\text{C}$, $5\sim70\%\text{RH}$ (No dew condensation)		
Equipment status		Transportable equipment		
Dimension, Weight	Main unit	170mm (W) × 120mm (D) x 107mm (H) 0.77Kg (When power supply unit is assembled, without protrusion and AC adapter)		
Standard component		Main unit Cathode unit, Base unit, Power supply part, AC adapter, Extension code Instruction manual		

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