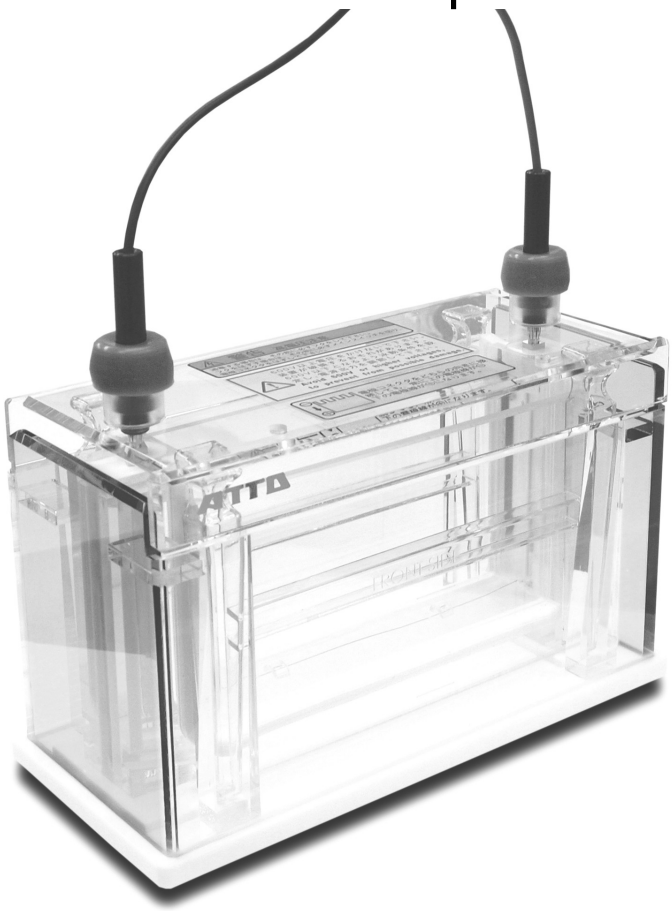
Instruction Manual

Wide-slab Gel Electrophoresis
WSE-1170
Multi‐Lane Gel Electrophoresis system



June 1st, 2017 3rd Edition

ATTO
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Introduction

Thank you for purchasing ATTO Corporation's Mini-Size electrophoresis system "WSE-1170 Multi-Lane Gel Electrophoresis system " 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, guarantee and services 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 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 tainted, 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 type. 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

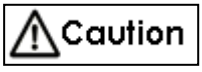
To use this system safely, it is a must to operate it properly. Do not use this product until you read this document carefully and understand the content sufficiently. Precautions on usage and safety described in this document are applied to the use of this system only for the specified purpose of use. 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 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 caused by the electrophoresis system or any damage to the system, please understand and follow the correct operation method shown in this manual.

If you have questions or concern related to the principle of electrophoresis, maintenance or inspection, feel free to contact 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
	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, caused by ignoring the symbol and mishandling the system.
	This symbol indicates possible occurrence of physical damage caused by ignoring the symbol and mis-handling the system.
	This symbol indicates prohibition.
	This symbol indicates an important matter.
	This symbol indicates a tip related to the 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.

⚠Danger

Power supply connection 	Do not use a deformed or corroded electrode terminal, power cable whose insulation coating is peeled off, or damaged lead cable. 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 contact our company (please refer to the back cover).
No wet hand 	When handling this system, keep your hands dry. Do not touch electrode terminal or power cable with wet hands. If you do, electric shock or failure may be caused. If wet, stop using it and contact 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, 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, or and see if any liquid leakage by regular visual inspection. If you find an error, failure or defect, stop using it and contact our company (please refer to the back cover).
Reagent 	For the electrophoresis, deleterious substance, dangerous substance, or carcinogenic material may be used for preparation of buffer solution, staining or decoloring operation. Do not allow its 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.

⚠Caution

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 system 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, nor move it. Electric shock may be caused by leakage of Electrode buffer solution. Also, electric cords may get entangled and the system may fall. When moving this system, be sure to turn off the power switch and disconnect power cable, and then disconnect all cables.
Maintenance 	When you conduct maintenance or cleaning, be sure to turn off the switch of the power supply and disconnect all cables. To maintain good performance and safety of this product, please ask us for periodical maintenance, inspection and calibration (please refer to the back cover).
No disassembly 	Do not disassemble or modify this system. Interior adjustment or repair of this product should be made by our engineers. If adjustment or repair needs to be done, please ask us for it (please refer to the back cover). Our company will not accept any responsibility for any accident or failure caused by disassembly or modification done by yourself.
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 (please refer to the back cover).

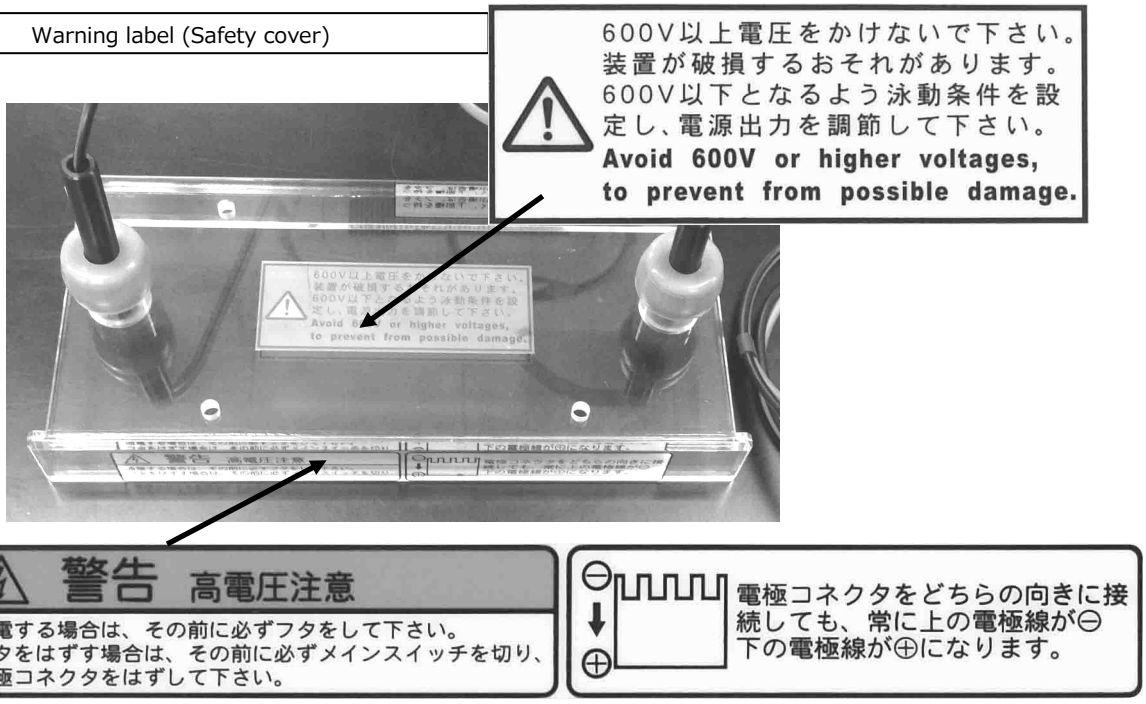
⚠Waming

Lead cable ⚠	Do not use the lead cable of this system for any other purpose than electrophoresis 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 lead cable of this system for any other system than this system.

Information

Sticker	The product seal indicates the important information for maintenance and management of the product. Do not peel it off.

Warning label (Safety cover)



Be sure to cover the lid before turning on the power. Before removing the lid, turn off the main switch and remove the electrode connector

The electrode at the top is always the cathode and the electrode at the bottom is the anode no matter which direction the electrode connector is connected.

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 judgment 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. This 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 specifications of the product and the contents of the instruction manual may be changed without prior notice.

1 Overview

1.1 Purpose

WSE-1170 Multi-Lane Gel Electrophoresis system is the system for carrying out electrophoresis of protein and nucleic acid using two wide size polyacrylamide gel (140 x 80 x 1.0mm) at the same time.

2 Inspection for unpacking

2.1 Inspection for unpacking

When you received 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 that you purchased the product or our company immediately (please refer to the back cover). Carry out the inspection within one week after you received the product. After one week elapses, damage or parts shortage may not be recovered.

Model	WSE-1170	WSE-1170W
Code No.	2322210	2322211
Main unit	Electrophoresis Chamber	
	Pressure plate	
	Safety cover（with lead cable）	

Material

Electrophoresis Chamber	acrylic , silicon
Pressure plate	polycarbonate
Safety cover	acrylic
Dummy plate	acrylic

Accessories

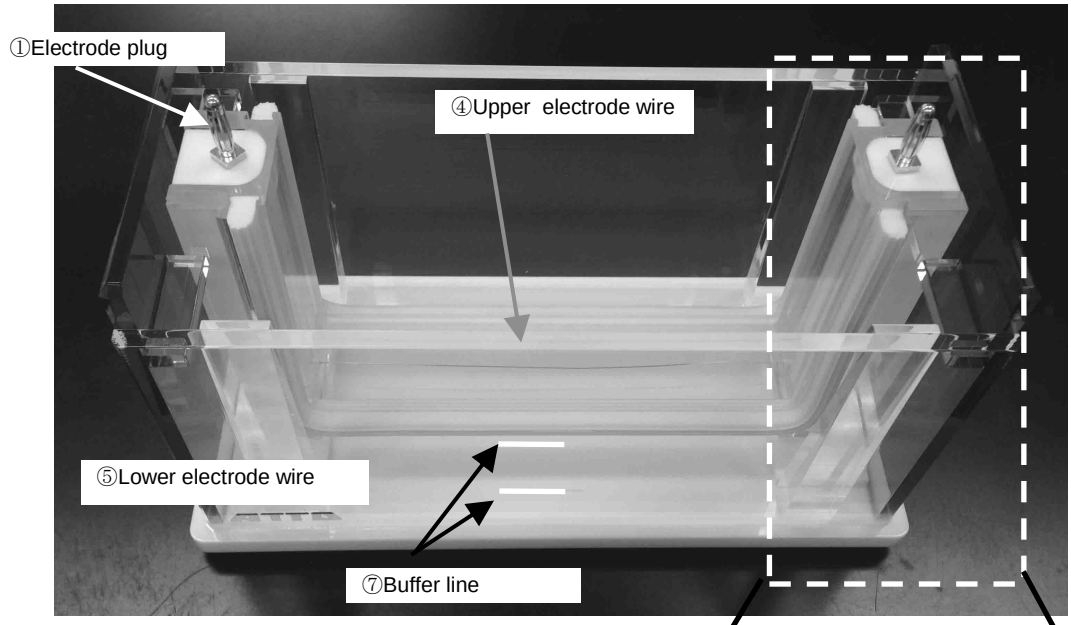
Model	WSE-1170	WSE-1170W
Dummy plate	Dummy plate (for WSE-1170)	
Instruction manual	1	
Gel cast	X	WSE-1195 Multi-Lane Gel Electrophoresis system

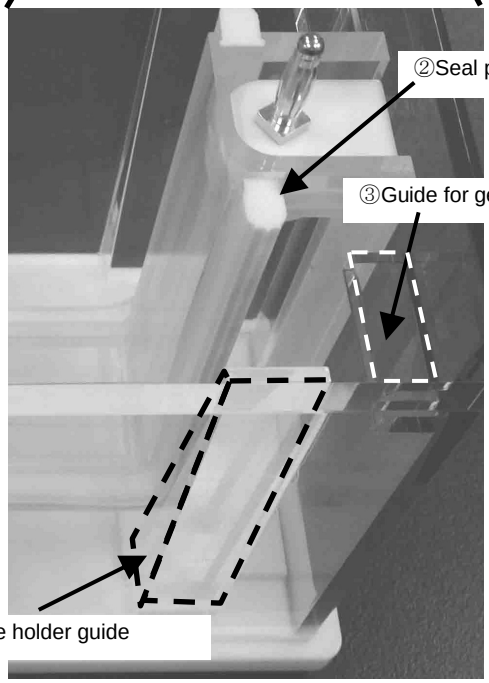
3 Name and function of each part

3.1 Name of main unit and function

（1）Electrophoresis Chamber

It is for setting upper and lower buffer and the gel plate. Electrode plug is also equipped.





① Electrode plug

A connection part with the electrode connector of the lead cable. The upper part becomes the (-) pole and the lower part becomes the (+) pole automatically by the automatic polarity switching function.

② Seal packing

Hold the upper buffer by attaching with the gel plate.

③Guide for gel set support

Guide when setting the gel plate. It is on the left and right side.

④Upper electrode wire

It becomes (-) pole.

⑤Lower electrode wire

It becomes the (+) pole.

⑥Plate holder guide

It is a guide when setting the plate holder.

⑦Buffer line

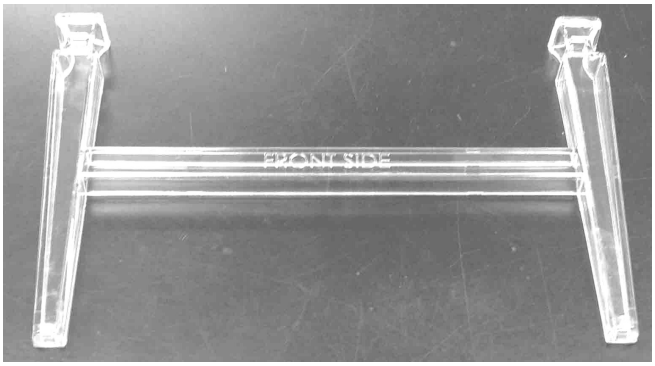
The line indicates the amount of buffer in the lower chamber.

The upper line is the maximum amount, the lower line is the minimum amount of the buffer. In order to reduce the heat generation of the gel, it is recommended to run with maximum amount.

* Gel heat generation causes smiling, worsen of the pattern.

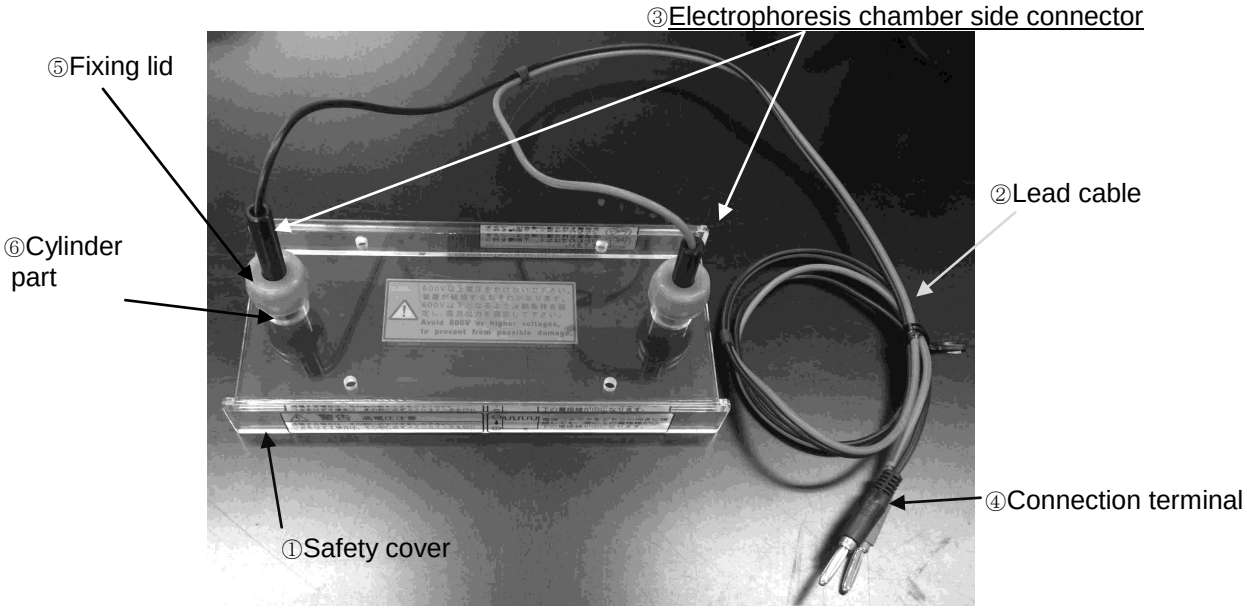
⑵ Plate holder

Hold the gel plate to the seal packing of the electrophoresis chamber.



⑶ Safety cover（with lead cable)

Set it on the top of the electrophoresis chamber to prevent contact with electrode plugs and electrode solution during electrophoresis and prevent dust entering. Lead cable is included.



① Safety cover

Prevents danger of electric shock etc. by contacting with electrophoresis buffer in electrophoresis chamber during electrophoresis.

② Lead cable

A wire for energizing the electrophoresis chamber from the power supply.

③Electrophoresis chamber side connector

Connect to the electrode plug of the electrophoresis chamber and supply current from the power supply.

④Connection terminal

Connect to the power supply and supply current from it to the electrophoresis chamber.

⑤Fixing lid

A lid for fixing the lead cable to the safety cover.

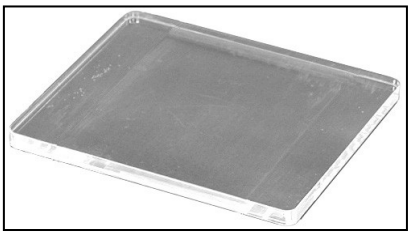
⑥Cylinder part

A cylinder part of the safety cover is for holding the fixing lid. There is a hole for connecting the lead cable and the electrode plug of the chamber.

3.2 Name and function of accessories

Dummy plate

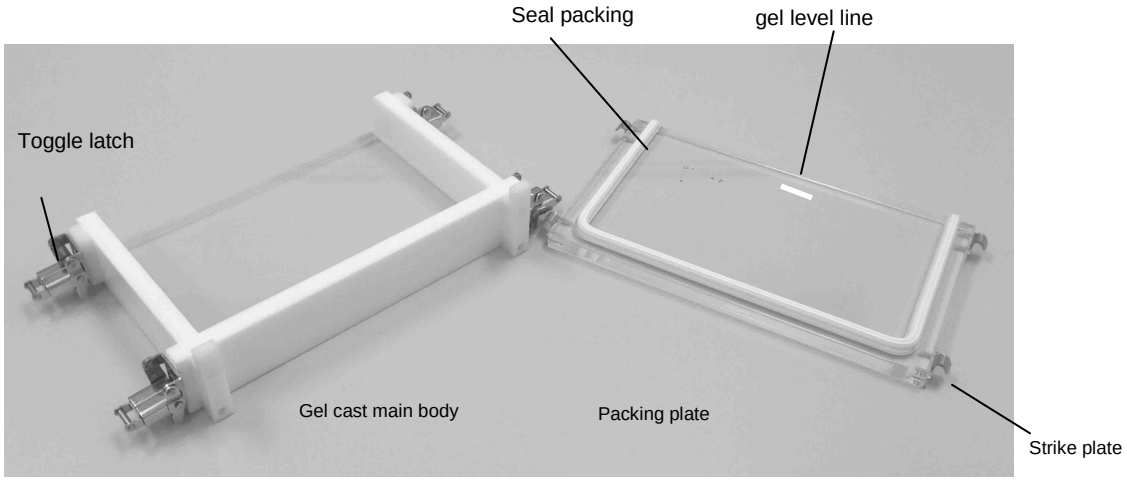
A plate used instead of a gel when running a single gel. To hold the upper buffer, can be used as a substitute for the gel.



Dummy plate（For WSE-1170)

Multi-Lane Gel Hand-cast system（WSE-1195)

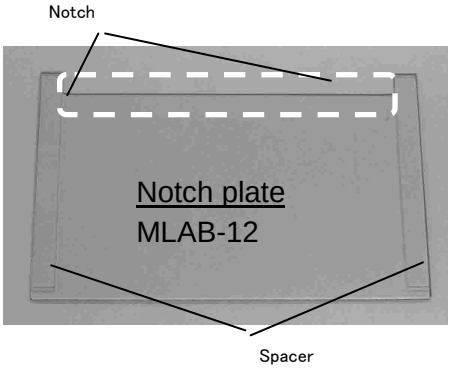
Hand –cast gel accessory only for WSE-1170 type.



① Notch plate MLAB-12 (2 plates)

Assemble Notch plate with Plane plate and make gel. When setting the gel in an electrophoresis chamber, put notch plate side to be seal packing side of the chamber.

As shown on the right, the plate has "spacer" and "notch".

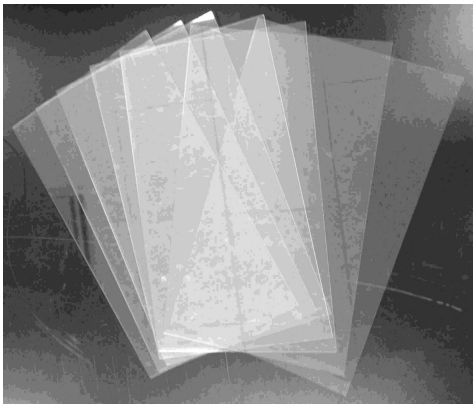


② Plane plate MLB-02 (2 plates)

Cast a gel between MLAB-12 and plane electrophoresis plate.

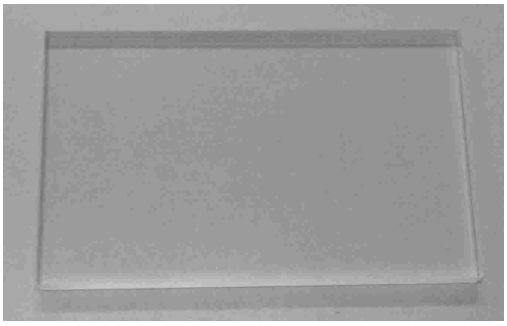
③Spacer plate（6 plates）

Put the spacer plate between a set of the gel plates and another set to prevent excess gel to be made.



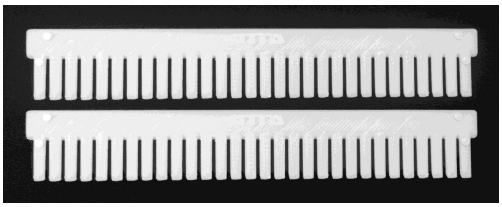
④ Dummy plate

A dummy plate is used when casting two gels. It is not used when casting 4 gels.



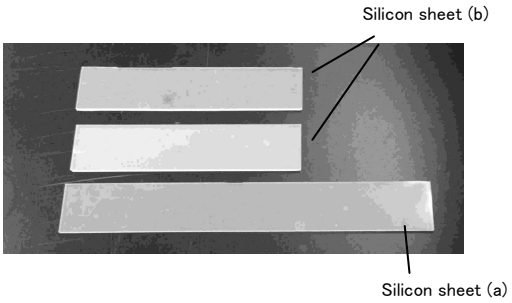
⑤ Multi-lane comb

Make wells for applying samples to the gel



⑥Silicon sheets

Silicon sheets to set on the bottom and the side of the gel cast main body. There are two types with the same width and different lengths. The longer sheets is 22 x 164 mm (a) for setting on the bottom of the gel cast main body, and another two sheets are 22 x 103 mm (b) for setting to the left and right side.



4 Preparation

4.1 Installation environment

Please use this equipment in the following environment

Location	For indoor use only
Temperature・Humidity	4- 40℃・5- 70％RH（Non condensing）

⚠Warning

This apparatus should not be placed in an environment where there is inflammable gas. There is a possibility that an explosion and/or fire could occur because it does not have an explosion -proof structure. Please install it in an environment where there is no contact with inflammable gas.
This apparatus should not be placed in an environment where there is corrosive gas that may cause a corrosion of conductors inside the apparatus and poor contact between connectors, resulting in mechanical errors and/or failure, or fire. This apparatus should not be placed in a dusty environment where dust or other dirt may collect on this apparatus, because it may cause electrical shocks, fire, or mechanical failures.

⚠Caution

This apparatus should not be used in a place where there is a strong magnetic or electric field, or excessive source-voltage fluctuation and/or electrical noise. It can be a cause of mechanical errors.
This apparatus should not be placed in a location where there is direct sunlight, or where abrupt temperature changes or high humidity occur. Do not use this apparatus when condensation has occurred.
This apparatus cannot be used outdoor. It is designed to ensure safety and performance under conditions of an ambient temperature of 4 to 40^{O}C and relative humidity of 5 to 70% (no condensation).

4.2 Preparation of peripheral system & consumable goods

Power supply

AE-8155 my PowerII 500

AE-8135 my PowerII 300

WSE-3200 Power stationIII

Gel casting system

WSE-1195 Multi Lane Casting gel kit

Heat block

Used for staining and bleaching of gel
WSC-2400 Seesaw Shaker atto

Gel dryer

Used for gel storage and drying
AE-3711 RapiDry mini

Blotting apparatus

WSE-4025 Horize BLOT 2M

WSE-4045 Horize BLOT 4M

WSE-4053 P plus membranes（260mm×3m）

CB-20A Filter paper（20×20cm）

4.3 Preparation of reagents

1. Reagents

Prepare the following reagents :

1) Protein electrophoresis and visualization

1.Polyacrylamide gel electrophoresis

Acrylamide (for electrophoresis)
N,N'-methylenebisacrylamide (for electrophoresis)
Tris (tris hydroxymethyl aminomethane) (for biochemistry)
SDS (sodium dodecyl sulfate) (for biochemistry)**
Hydrochloric acid (special grade)
Ammonium persulfate
TEMED (N,N,N',N'-tetramethylethylenediamine) (for electrophoresis)
Glycine (special grade)
Glycerin (special grade)
2-mercaptoethanol **
BPB (bromphenol blue)
** Not used for Native PAGE

2.Coomassie Brilliant Blue (CBB) staining

AE-1340 EzStain AQua

or

Acetic acid (special grade)
Methanol (special grade)
CBB (Coomassie brilliant blue) R-250 or G-250 (for electrophoresis)

2) DNA electrophoresis and visualization

1.Electrophoresis

Acrylamide (for electrophoresis)
N,N'-methylenebisacrylamide (for electrophoresis)
Tris (tris hydroxymethyl aminomethane) (for biochemistry)
Ammonium persulfate (for electrophoresis)
TEMED (N,N,N',N'-tetramethylethylenediamine) (for electrophoresis)
Boric acid (special grade)
EDTA / 2NA (ethylenediaminetetraacetic acid / disodium ethylenediaminotetraacetate)
BPB (bromphenol blue)
Sucrose (special grade)

2.Ethidium bromide staining

WSE-7130 EzFluoroStain DNA

or

Ethidium bromide

Our company sells following products.
Please use them according to application. For details of products, please contact us. (Refer to the back cover.)

	Application	Model	Product name	Code #
Gel buffer for PAGE	For (SDS-)PAGE Buffer for electrophoresis	WSE-7310	EzGelAce	2332327
	For (SDS-)PAGE Buffer for electrophoresis concentrated gel	WSE-7155	EzGel Stack	2332329
	For (SDS-)PAGE Buffer for electrophoresis separated gel	WSE-7150	EzGel Sep	2332328
For sample prepara-tion	RIPA for protein extraction buffer	WSE-7420	EzRIPA Lysis kit	2332336
	Mammalian cells (organelle) fractionation/ Extraction Kit	WSE-7421	EzSubcell Extract	2332337
	Mammalian organella (Nuclear, Mitochondria) fractionation reagent kit	WSE-7422	EzSubcell Fraction	2332338
	Solubilizing buffer for protein extracting from Yeast & Bacteria	WSE-7423	EzBactYeastCrusher	2332339
	Phosphate buffered saline for cellular experiments	WSE-7430	EzPBS(-)	2332380
	Loading dye for DNA electrophoresis	WSE-7040	EzApply DNA	2332394
	Fluorescent protein labeling kit for SDS-PAGE	WSE-7010	EzLabel FluoroNeo	2332333
	Sample preparation buffer for SDS-PAGE	AE-1430	EzApply	2332330
	Sample preparation buffer for 2D	AE-1435	EzApply 2D kit	2332335
Molecular weight marker	DNA molecular weight marker for electrophoresis	WSE-7030	EzDNA Ladder	2332342
	Prestained protein marker for SDS-PAGE/ western blotting	WSE-7020	EzProtein Ladder	2332346
	Protein molecular weight marker for SDS-PAGE	AE-1440	EzStandard	2332340
	Prestained protein marker for SDS-PAGE/ western blotting	AE-1450	EzStandardPrestainBlue	2332347
Electro-phoresis buffer	Electrophoresis buffer for SDS-PAGE	AE-1410	EzRun	2332310
	High separation type electrophoresis buffer for SDS-PAGE	AE-1412	EzRunC+	2332320
	Electrophoresis buffer for trisine PAGE	AE-1415	EzRunT	2332325
	Tris-MOPS based Electrophoresis buffer	WSE-7065	EzRunMOPS	2332326
	Electrophoresis buffer for tris glycine PAGE	WSE-7055	EzRunTG	2332323
	Tris-borate based electrophoresis buffer	WSE-7051	EzRunTBE	2332392
Gel staining reagent	Fluorescent staining reagent for DNA detection	WSE-7130	EzFluoroStainDNA	2332395
	CBB staining solution for protein detection without acetic acid and alcohol	AE-1340	EzStainAQua	2332370
	Reverse staining reagent kit for protein detection	AE-1310	EzStainReverse	2332350
	Silver staining reagent kit for protein / nucleic acid detection	AE-1360	EzStainSilver	2332360
	Small volume centrifugal filtration tube	AB-1171	ATTOPREP MF	3521370

2. Preparation of electrophoresis solutions

1) For protein electrophoresis

Prepare the following solutions beforehand.

Acrylamide / bis solutions generally use 19: 1, 29: 1, 29.2: 0.8, and 37.5: 1 cross-linking solutions, depending on the molecular weight cut-off range. The following shows an example of preparation of 29.2: 0.8. If separation is performed while retaining the higher-order structure of protein (Native PAGE), remove SDS and DTT from all reagents (* reagent).

⚠Caution Acrylamide monomers are neurotoxic. Please protect your body such as gloves when handling.

Name of solution	Name of reagent	Capacity ( ) indicates final concentration
A solution (30%Acrylamide / bis solution (29.2 : 0.8) (4° C ・Can be stored for 1 month)	Acrylamide N,N'-methylenebisacrylamide Dissolve in distilled water and mess up to 100 mL.	29.2g 0.8g (30%)
B solution (4° C ・Can be stored for 1 month)	Tris SDS(*) Dissolve in distilled water, adjust to pH 8.8 with hydrochloric acid, and mess up to 100 mL., or use WSE-7150 EzGelSep.	18.2g (1.5M) 0.4g (0.4%)
C solution (4° C ・Can be stored for 1 month)	Tris SDS(*) Dissolve in distilled water, adjust to pH 6.8 with hydrochloric acid, and mess up to 100 mL., or use WSE-7155 EzGelStack	6.1g (0.5M) 0.4g (0.4%)
D solution (4° C ・Can be stored for 1 month)	Ammonium persulfate Dissolve an item above with 1.0mL of distilled water.	0.1g (10%)
SDS-PAGE electrophoresis buffer (for Laemmli) (Can be stored for 2 months at room temperature) [Not required when using AE-1410 EzRun]	Tris Glycine SDS(*) Dissolve in distilled water and mess up to 500 mL.	1.5g (25mM) 7.2g (192mM) 0.5g (0.1%)
Sample buffer (Can be stored at 4 ° C for 2 weeks) [Not required when using AE-1430 EzApply]	Solution C (0.5M tris-hydrochloric acid buffer pH6.8) SDS(*) DTT(*) Glycerin 1% BPB solution Dissolve in distilled water and mess up to 10 mL.	1.0mL (50mM) 0.1g (1%) 0.15g (100mM) 2.0mL (20%) 10µL (0.001%)
CBB staining solution (Can be stored for 1 month at room temperature) [Not required when using AE-1340 EzStainAQua]	Methanol Acetic acid CBB R-250 or G-250 Dissolve items shown above and dilute it to 1L.	300mL (30%) 100mL (10%) 1.0g (0.1%)
Decolorization solution (Can be stored at room temperature for 1 month) [Not required when using AE-1340 EzStainAQua]	Methanol Acetic acid Dissolve in distilled water and make up to 1 liter.	300mL (30%) 100mL (10%)

2) For DNA electrophoresis

Prepare the following solutions beforehand.

Solution	Reagent	Capacity ( ) indicates final concentration
Solution E (4° C ・can be stored for one month)	Acrylamide N,N'-methylenebisacrylamide Dissolve in distilled water, and mess up to 100mL.	29.2g 0.8g (30%)
Solution F (5xTBE buffer) (4° C ・can be stored for one month)	Tris Boric acid EDTA・2NA Dissolve in distilled water, and mess up to 1L. Or use WSE-7051 EzRun TBE.	53.9g (445mM) 27.5g (445mM) 3.7g (10mM)
Solution D (4° C ・can be stored for one week)	Ammonium persulfate Dissolve in 1 mL of distilled water,	0.1g (10%)
TBE Runnning buffer (1xTBE buffer) (Room temperature / can be stored for two months)	Tris Boric acid EDTA・2NA Dissolve in distilled water, and mess up to 1L. Or dilute WSE-7051 EzRun TBE to 1/10.	10.8g (89mM) 5.5g (89mM) 0.74g (2mM)
Loading dye solution (4° C ・can be stored for two months)	BPB Sucrose Dissolve in distilled water, and mess up to to 10mL.	0.04g (0.4%) 6.0g (60%)
Ethidium bromide stock solution (4°C / can be stored for two months)	Ethidium bromide Dissolve in 100mL of 1xTBE buffer.	50mg (0.05%)
Ethidium bromide staining solution (4°C / can be stored for two months)	Dilute the ethidium bromide stock solution to 1/100 with 1xTBE buffer.	(0.0005%)

3. Sample preparation for electrophoresis

The followings are shown one of the example for popular sample preparation method. Various sample preparation methods are available depending on the sample and experimental purpose. Try to find the best ways for your sample by referring to literature and other documents.

1) Protein

Solvilization of sample

Use the sample buffer described above (page 12) or use AE-1430 EzApply for protein sample preparation. For preparation from dried samples, dilute them in sample buffer at the final protein concentration of 1 to 2mg/mL . For samples with less water like tissue, add appropriate volume of the sample buffer and homogenize them. For solution-like samples add the concentrated sample buffer directly for preventing dilution.

(e.g.) human plasma sample

Sample, reagent	Capacity
Human plasma	10μL
Distilled water	490μL
10% SDS solution*	100μL
1M DTT solution*	100μL
C solution	100μL
Glycerin	200μL
Total	1mL

∗ For Native-PAGE, do not add the reagents with the * mark shown above, but add the distilled water instead.

When using AE-1430 EzApply (easy apply), please follow the following instruction.
If the sample is a solution, mix 1: 1 with the sample.
In the case of solids, homogenize by adding an appropriate amount of EzApply that has been diluted double in advance.
When using EzApply in the above example, take 490 μL distilled water in a tube, add 500 μL EzApply, and 10 μL plasma.

Heating process

Seal the lid of the tube and treat them at 95 degrees Celsius for 5-10 minutes.

Centrifugal separation

Centrifuge (15,000 rpm, 10 min). The supernatant except the surface fat layer is used as a sample.

If the sample including insoluble substances or fat, the electrophoresis pattern is affected and appeared the vertical

2) DNA

The salt concentration of samples should be identical as possible. For high salt concentration, carry out the ethanol precipitation and dissolve it in buffer including the same salt concentration as that of other samples.

If the salt concentration is different, the electrophoresis pattern is affected. High salt concentration sample (e.g. H buffer of restriction enzyme.), in particular, affects the band pattern or the electrophoresis speed of the neighboring lane.

Mix one-tenth loading dye solution (BPB) into sample solution and use it as electrophoresis sample.

4.4 Preparation of gel

Assemble the glass plate and make gel using the solution as described above.

When you use precast gel like e-PAGEL-HR , the following process is not necessary.

1. Assembly of the Glass plate

For the experimental operation, be sure to wear clean gloves for experiment on both hands.

⚠Caution Do not handle the gel plate with bare hands. There is a risk of injury.
When handling the gel plate, wear experimental gloves and protect.

If you operate the product with bare hands, experimental tools and solutions may be contaminated and you may not obtain the best experimental results. If the glass plate is dirty or adhere something, bubbles will easily appear during gel casting. Please wash well beforehand.

2. Preparation of gel

The gel concentration suitable for separation of the molecular weight range of protein and nucleic acid are shown in the following table.

Gel concentration and fractionation range

Gel concentration	Fractional molecular weight range (Protein)	Fractional molecular weight range (Nucleic acid) (1)	Fractional molecular weight range (Nucleic acid) (2)
5%	80~400 KDa		80～500bp
7.5%	40~200 KDa	200~3,000 bp	
10%	20~130 KDa	100~2,000 bp	50～300bp
12.5%	14~80 KDa	70~1,800 bp	40～200bp
15%	10~60 KDa	50~1,500 bp	25～150bp

(1)Tris-Glycine gel
(2)1 x TBE gel

1) Gel cast for protein

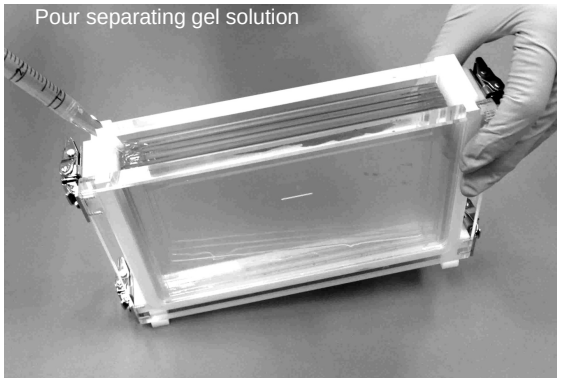
Select the gel concentration according to the molecular weight of protein. In case of native PAGE, the charge state of the sample greatly affects the mobility. Therefore, it is not possible to select the gel concentration based only on the molecular weight of the sample. Select the gel concentration by preliminary experiments.

Weigh distilled water and solutions A and B in the amounts shown in the following composition table and gently mix them. Just before the polymerization reaction, add the TEMED and D solutions and gently mix to make a separate gel solution.
* B solution goes bubbly because it contains SDS. Please do not mix vigorously.
* Start polymerization by addition of TEMED. After mixing evenly, pour the solution immediately.

	Separating gel						Stacking gel
	5%	7.5%	10%	12.5%	15%	20%	4.5%
Distilled water	17.4	15	12.6	9.9	7.5	2.4	9
Solution A	5.1	7.5	9.9	12.6	15	20.1	2.25
Solution B	7.5	7.5	7.5	7.5	7.5	7.5	-
Solution C	-	-	-	-	-	-	3.75
Solution D	0.225	0.225	0.15	0.15	0.15	0.15	0.15
TEMED	0.015	0.015	0.015	0.015	0.015	0.015	0.0075

※The capacity is required for 2 pieces of wide gel sizes. Unit : mL

While keeping the assembled the multi-lane gel hand-cast system, pour the separating gel solution from the notch part of the Glass plate gently. If poured fast, uneven polymerization may occur. Do not leave air bubbles behind.



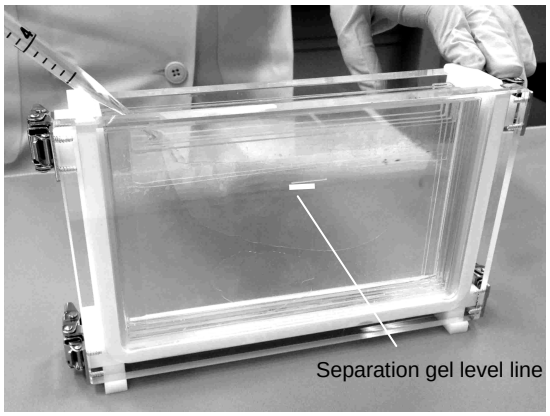
With the separating gel solution poured up to 1/3, tilt the multi-lane gel hand-cast system back and forth to make the solution spread throughout. Also, if there are air bubbles at the bottom of the glass plate etc., remove them by tilting the caster.

Tilt the body to spread evenly the gel solution

Pour the separating gel solution up to the separation gel level line marked on the multi-lane gel hand-cast system. Prepare it by tilting the caster back and forth until the gel heights of all glass plates are the same.

Overlay distilled water on the separating gel solution of each glass plate to a height of 2- 3 mm so as not to disturb the interface, and let it stand for 30 minutes or more to polymerize.

Pour separating gel solution

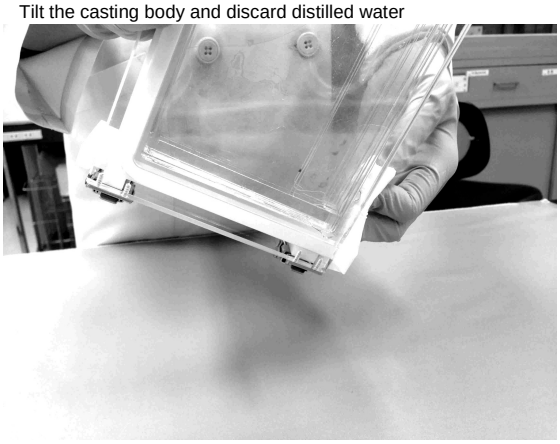


Pour the distilled water on the top of each glass plate into equal volumes. If the amount of water is different, the height of separation gel will not be evenly.

At low temperature (20°C or less), it takes longer time to polymerization. The polymerization time will vary depending on the temperature. Polymerization reaction should be done always at a constant temperature so as to maintain reproducibility of the electrophoresis pattern.

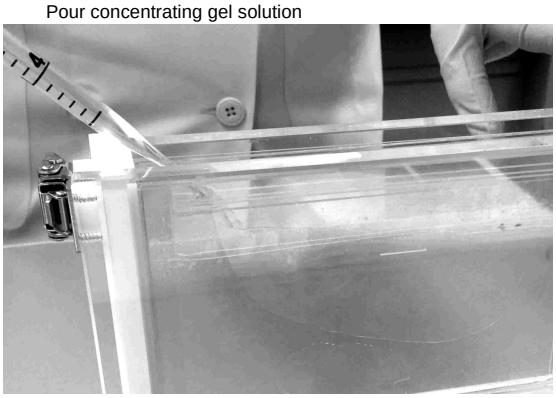
After polymerization is completed, the gel interface will be visible. Discard the distilled water. At this time, make the caster slant while holding down lightly so that the glass plate does not slide out of the multi-lane gel hand-cast system.

Tilt the casting body and discard distilled water



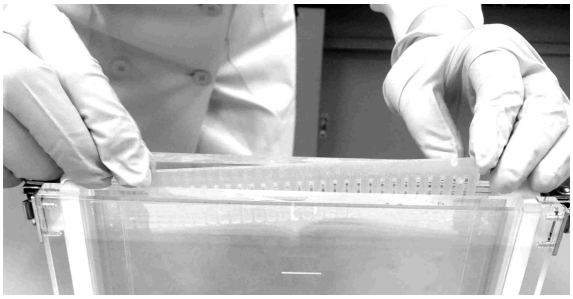
Prepare the concentrating gel solution, using the composition table on the previous page. Pour the prepared concentrating gel solution up to the top end of the glass plate.

Pour concentrating gel solution

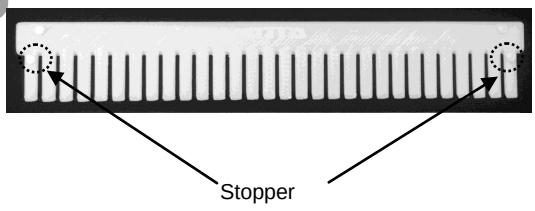


Insert the sample comb until the stopper will touch on the notch of the plate. Polymerization will complete in about 30 minutes.

Insert the sample comb



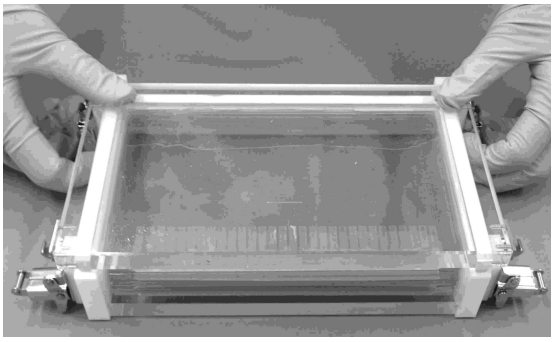
Be careful so that air bubbles will not stick to the teeth of the comb. If they do, the well will not be formed properly.



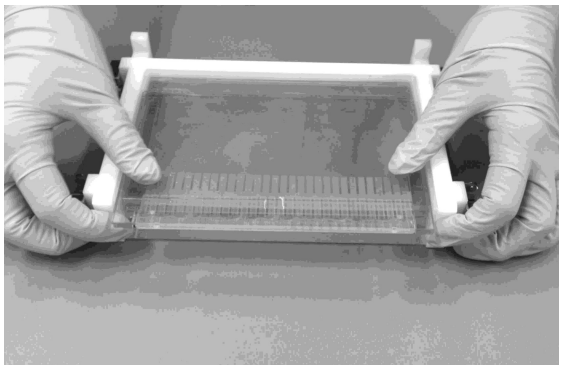
After confirming the complete gel polymerization, remove the gel from the Gel Hand-cast system. Lay the gel cast main body horizontally and unlock the left and right latches.

Remove the packing plate.

Unlock the latches



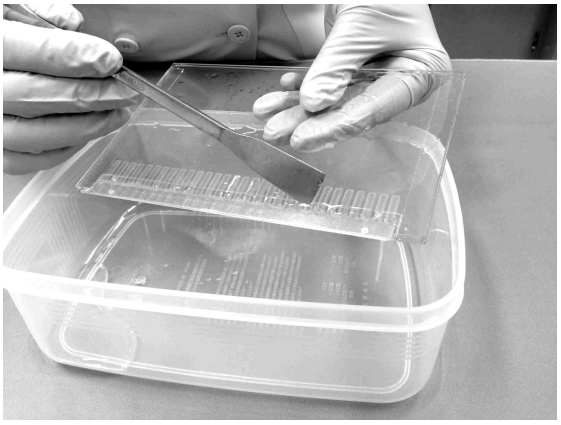
Slide the gel plate or spacer plate and remove each gel from the Gel Hand-cast system.



Removal of excess gel and unpolymerized acrylamide solution

Immediately wash off the gel solution adhering to the glass plate with distilled water. Excess gel around the comb should be removed using a spatula and so on.

If you do not use them all, wrap the rest gel to avoid for drying and keep in refrigerator.



2)DNA

Select the gel concentration from the following table according to DNA fragment size and experimental purpose（refer to the table on p15）. Stacking gel is unnecessary for separating DNA fragment in general.

Mix the solution E, F, D and distilled water according to the following table. Then add TEMED and solutions D just before polymerizing reaction of the separating gel.

If the gel is difficult to polymerize, increase the D solution and TEMED by 10% each.

	5%	6%	7.5%	8%	10%	12.5%	15%
Distilled water	18.99	18	16.5	15.99	14.01	11.49	9
Solution E	5.01	6	7.5	8.01	9.99	12.51	15
Solution F	6	6	6	6	6	6	6
Solution D	0.225	0.225	0.225	0.225	0.225	0.225	0.225
TEMED	0.01125	0.01125	0.01125	0.01125	0.01125	0.01125	0.01125

※The amount of each solution as shown in the table required for 2 pieces of wide gel sizes. Unit : mL

While the Gel Hand-cast system is kept tilted, pour gently the gel solution from the notch part of the Glass plate. If poured fast, uneven polymerization may occur. Do not leave air bubbles behind. Pour the gel solution up to the top of the gel plate, and insert the sample comb until the stopper will touch the notch part. Polymerization will complete in about 30 minutes.

Be careful not to leave air bubbles in the comb teeth. If so, the formation of the wells will be disturbed

After polymerizing the gel, remove the gels from the Gel Hand-cast system .

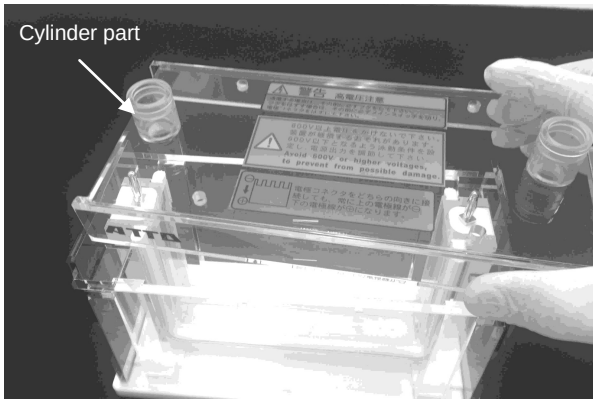
Wash off the gel solution adhering to the glass plate with distilled water. Excess gel around the comb should be removed using a spatula and so on.

If you do not use them all, wrap the rest gel to avoid for drying and keep in refrigerator.

5 Operation

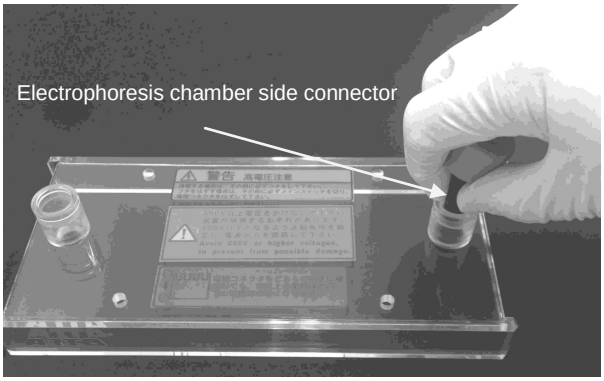
5.1 Prepare for electrophoresis

Assemble the safety cover and lead cables.

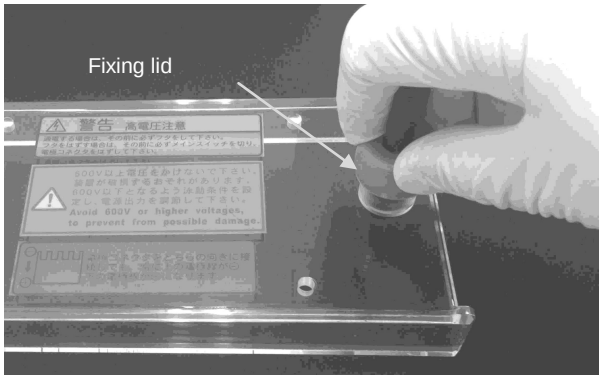


Set the lead cable on the safety cover.

Insert the electrophoresis chamber side connector of the lead cable into the cylinder part of the safety cover.

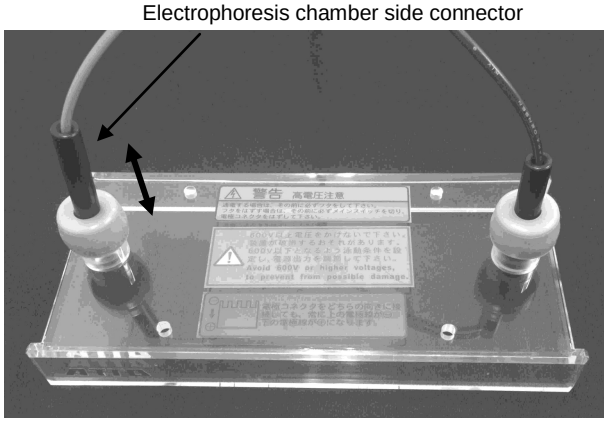


Fix the fixing lid to the cylinder part to connect the lead cable with the safety cover.



Confirm that the electrophoresis chamber side connector can move up and down smoothly in the cylinder part

Pull up the electrophoresis chamber side connector and confirm that the connector does not come off from the safety cover easily.



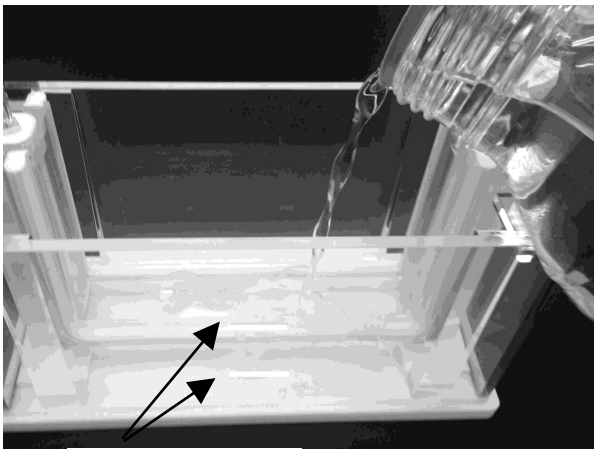
5.2 Set lower buffer

Pour the electrophoresis buffer into the electrophoresis chamber up to the buffer line. Pour gently so as not to bubbling.

The buffer lines indicate as following.

The upper buffer line indicates the maximum amount of buffer volume: 700 mL

The lower buffer line indicates minimum amount of buffer volume: 230 mL



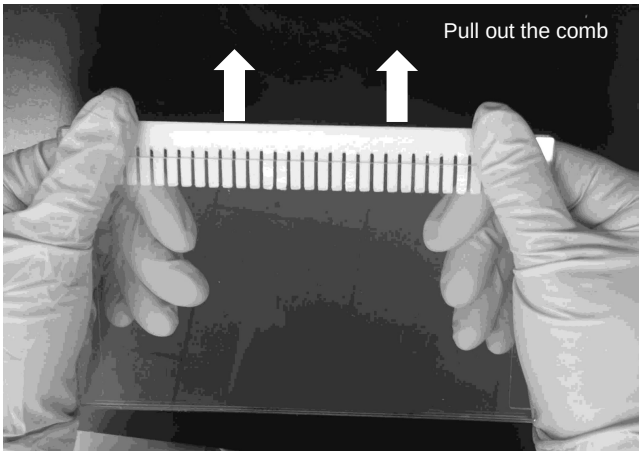
Buffer line

If the electrophoresis buffer is poured more than the upper buffer line, the buffer will spill out of the electrophoresis chamber when removing the gel after electrophoresis.

5.3 Gel setting

Gently pull up in the direction of the arrow and remove the comb. If removed quickly, it may damage the wells.

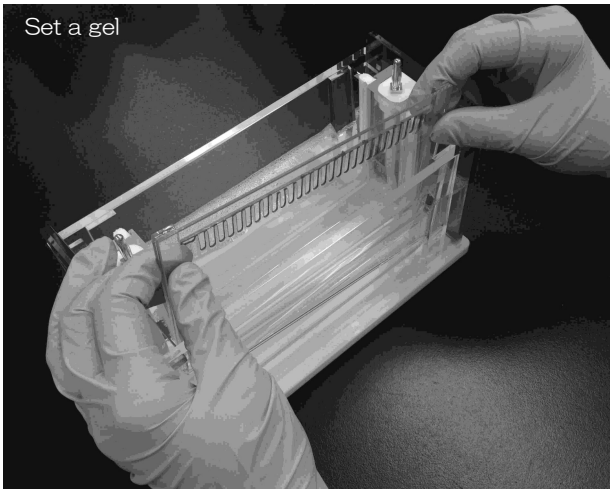
Wash the wells with a small amount of electrophoresis buffer to remove any unpolymerized acrylamide.



In case gel fragments or precipitated salt is left to the glass plate surface, wipe it off. If the glass plate dirty, it may cause the buffer to leak, because the plate will not able to fix with the seal packing tightly.

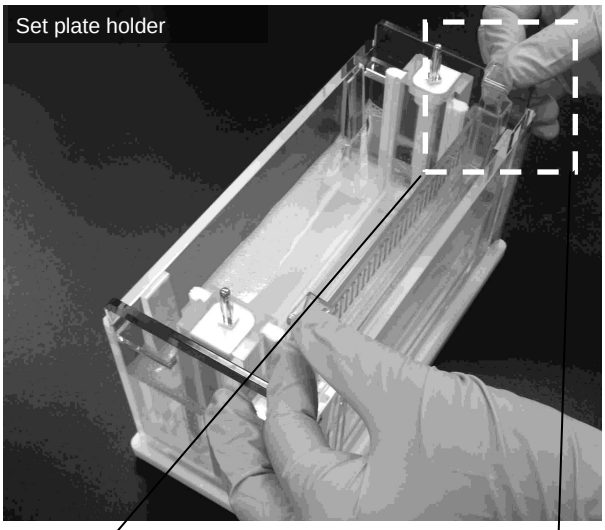
Put the gel plate in the electrophoresis chamber so as the notched side of the gel to face the seal packing of the electrophoresis chamber. Tilt the gel plate and set not to enter air bubbles the bottom side of the gel.

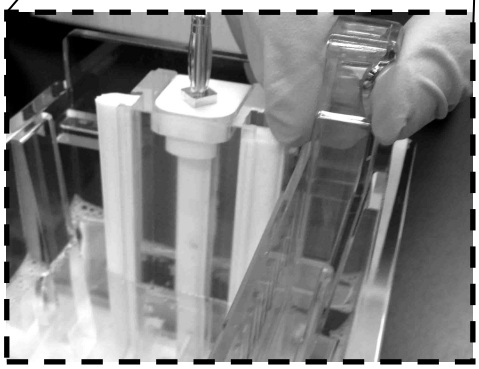
If running one gel, set the dummy plate instead of the gel. If running two gels, set the another gel on the other side in the same way.



Set the mark of "FRONT SIDE" on the plate holder comes to the front and fix the gel or dummy plate.

Press the plate holder vertically with equal force until the tip of the plate holder reaches the bottom of the electrophoresis chamber. If the plate holder is not set vertically, it may cause the buffer to leak.

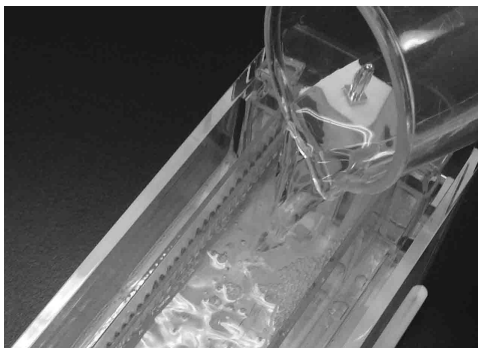




"FRONT SIDE"

Pour the electrophoresis buffer into the upper chamber.

Make the buffer amount level about 2-3 mm below from the top of the electrophoresis plate.

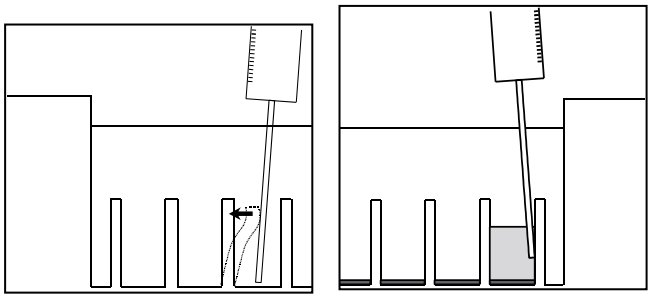


5.4 Adding samples

Wash the inside of the well and remove bubbles by pipetting. Also, if the well does not stand straight, use a syringe needle etc. to make it upright.

Apply the protein sample with a syringe or micropipette (use a tip with a thin tip that can be inserted between plates). Add 2 to 10 µL. The maximum amount of sample is about 20 µL.

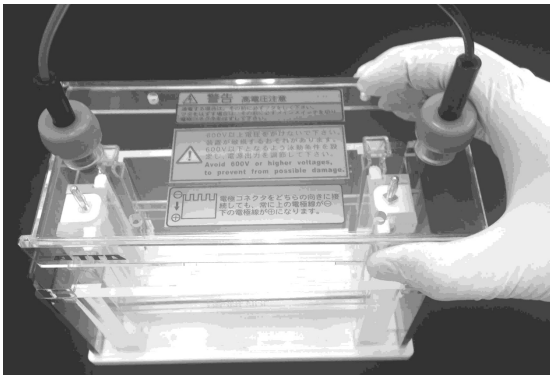
To obtain a clear electrophoresis pattern, insert the tip of the needle or pipette tip to the bottom of the well, and slowly pour the sample solution so as to the surface of sample solution rises from the bottom to the top. If dropping the sample from the top of the well, it may effect the pattern.



After applying the sample, start running immediately.

5.5 Start Electrophoresis

After applying sample, immediately set the safety cover to the electrophoresis chamber.



Make sure that the electrophoresis chamber side connector enters to the electrode plug of the upper chamber, push down the two electrode connectors.

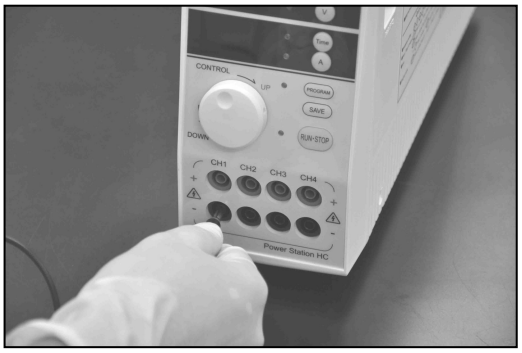
The polarity switching function switches plus and minus side automatically.

Before connecting, make sure that the power supply's main switch is off.

Connect the lead cable of the electrophoresis chamber and the power supply.

⚠Warning Do not operate with wet hands. Also, do not touch the electrode terminals or the power cables with wet hands.

⚠Warning Do not use if the power supply, electrode plug or connector is wet. It may cause an electric shock accident or a failure.
Please stop using this device and contact us.



Connect the lead cable of the electrophoresis chamber to the power supply terminal and turn on the power supply switch. Set the output conditions of the power supply, and press start button to start electrophoresis.

⚠Caution Do not apply the voltage more than 600V. Equipment may be damaged.

Set the electrophoresis conditions less than 600 V, and adjust the output from the power supply.

Supplementary explanation

A common condition is constant current of 30 mA /gel. When two gels are simultaneously electrophoresis, the constant current is 60 mA / electrophoresis chamber. The electrophoresis time at this time is 70 - 90 minutes *, and the voltage rises from about 110 V (at the beginning of electrophoresis) to 260 V (at the end of electrophoresis).

The conditions for high-speed electrophoresis of proteins using EzRun are a constant voltage of 300 V. Electrophoresis time is 30-40 minutes. The current value is about 90 mA (at the beginning of electrophoresis) to about 40 mA (at the end of electrophoresis) for one gel, and about 180 mA to 80 mA for two gels. The conditions for high-speed electrophoresis using EzMOPS are constant voltage of 250V. The electrophoresis time is about 30 minutes, the current value is about 130 mA (at the beginning of electrophoresis) to about 70 mA (at the end of electrophoresis) for one gel, and about 250 mA to about 140 mA for two gels.

* The electrophoresis time depends on the gel preparation method, the gel concentration and polymerization condition (the amount of ammonium persulfate and TEMED, and the temperature), the composition and concentration of the buffer and sample solution, and the buffer and ambient temperature.

5.6 Stop/Finish electrophoresis

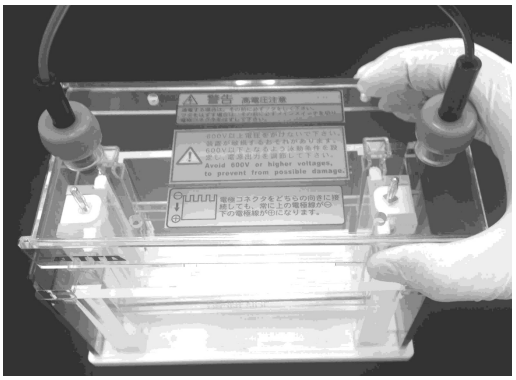
When the dye front (BPB) reaches about 5 - 10 mm above the bottom of the gel, stop electrophoresis.

Push Stop button of the power supply and turn off the main switch.

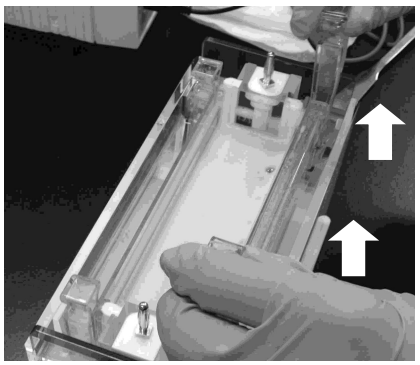
Hold the power supply and pull out the lead cables from the power supply terminal.

Hold the safety cover with hand, pull up the electrophoresis chamber side connectors, and remove them from the electrode plug of the electrophoresis chamber.

Remove the safety cover.



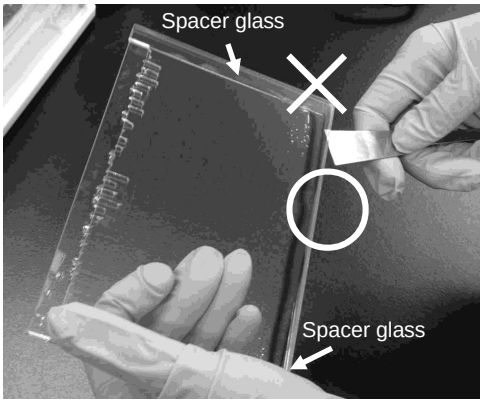
Pull up the plate holder.



Remove the gel from the chamber.

5.7 Detection

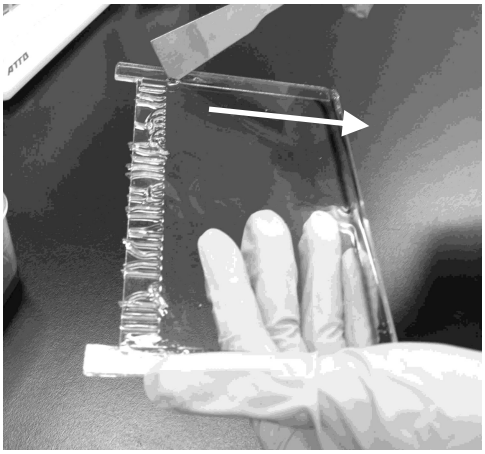
Hold the gel in hands with the plane plate (MLB-02) on top.



Insert a spatula between the two glass plates and gently move it up and down to remove the top plane plate. The spatula should not be inserted from the spacer side.

Inserting a spatula from the corner or spacer side can possibly damage the plate.

Wet the spatula or scalpel with staining solution etc. to avoid for adhering the sticky gels and then slowly cut between the gel and spacer glass. Be careful not to break the gel.



Put the gel plate in the container of staining solution so as to gel side would be down. Insert a spatula between the gel and the glass plate to peel off the gel carefully.

Keeping the spatula wet will prevent the gel from sticking and tearing.

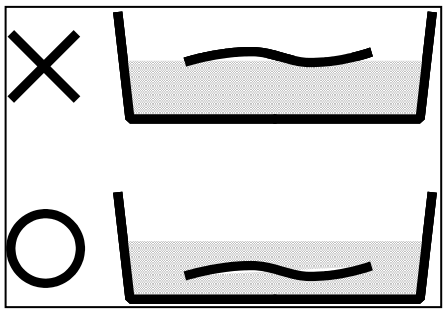
1) Protein staining

● EzStainAQua (CBB staining solution) and other staining

For EzStainAQua (CBB staining solution) and other staining such as silver stain, negative stain and fluorescent stain, follow the protocol of each instruction manual.

● CBB stain

Pour the CBB stain and shake for 60 minutes to overnight. Discard the staining solution, pour the destaining solution, and shake for 60 minutes to overnight.



2) Staining of DNA

● Ethidium bromide staining

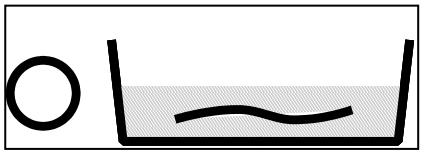
⚠Warning

Since ethidium bromide is a carcinogen, be sure to wear gloves and a white coat when handling it, so as not to touch it directly. Please take appropriate action according to your facility regarding waste disposal method. If you use a UV transilluminator, please do not use it until you read and understand the instruction manual. It may cause damage to eyes and skin. Please protect your body with protective glasses, face shield and gloves when irradiating with ultraviolet light.

Soak the electrophoresed gel in ethidium bromide staining solution and shake for 20 to 30 minutes.

Discard the staining solution and shake for 5 to 20 minutes with 1xTBE buffer.

Detect the bands by irradiation with ultraviolet light.



● Other staining

For staining such as silver staining and fluorescent staining, please follow the protocol of each instruction manual

5.8 System cleaning and storage

1. Cleaning of system

After use of electrophoresis chamber, glass plate and comb (only for handmade), before drying, wash them with a neutral detergent and the soft sponge and air dry before using it.

If contacted with an organic solution such as acetone or alcohol, or drying at high temperature may cause cracking, deformation or discoloration.

When cleaning the electrophoresis chamber, be careful not to cut the platinum wire on the chamber. Avoid cleaning with a metal weave sponge or test tube brush. The electrophoresis chambers are scratched and damaged by them and visibility is reduced during electrophoresis.

Discard precast gel glass plates. Performance can not be guaranteed for reuse.

Do not remove the upper chamber from the electrophoresis chamber.
It may cause breakage of platinum wire and breakage of upper chamber.

2. Storage of system

Do not store under direct sunlight, high temperatures or where there is possibility of being exposed to corrosive gases.

Do not store the electrophoresis chamber with glass plate to be set.

The multilane gel hand-cast system should not close the latch except when casting the gel. The elasticity of the seal packing is lost, which may cause liquid leakage during gel casting.

Do not store the electrophoresis chamber with the glass plate to be set.
The seal packing may deteriorate and cause buffer leakage during electrophoresis.

6. Troubleshooting

Symptom	Cause	Coping technique
Gel does not polymerize	Incorrect preparation of gel solution.	Remake each solution. If gel is not polymerized, remake the solution for each gel preparation
	Ammonium persulfate solution (solution D) is not fresh.	Prepare the ammonium persulfate solution just before use. It can be stored at 4°C for one week only.
	Room temperature or liquid temperature of gel solution is too low.	When the room temperature is 20°C or less, polymerization reaction is difficult to be caused. Maintain the temperature for polymerization between 20°C and 40°C approx.
	Other	Increase the ammonium persulfate solution and TEMED by about 10% each.
Air bubbles remain in the gel	Glass plate or comb is dirty.	Wash the glass plate and comb, dried completely and store them so that no dust will be attached to them. Do not touch gel contact surface of the plate and comb with a bare hand.
Uneven polymerization. Stripes are seen in the gel	Uneven polymerization	Uneven polymerization affects the migration pattern. If the plate or comb is dirty, or if the temperature difference between the plate or comb and the gel solution is large, uneven polymerization will occur easily. Remove each solution used for gel preparation from the refrigerator and bring it back to room temperature before use
Shape of the well looks strange	After pulling out the comb, monomer acrylamide is polymerized.	Pull out the comb just before use. After pulling it out, wash the well with electrode buffer or distilled water.
Buffer in the upper chamber leaks	Incomplete crimping of plate holder	Reinsert the plate holder vertically.
	Glass plate is dirty.	Wash the used glass plate before drying. Do not leave a gel fragment, etc. behind. After preparing the gel, wipe the gel debris, spilled gel solution, and any dirt on the outside of the gel plate with a damp paper towel.

Symptom	Cause	Coping technique
Buffer in the upper chamber leaks	Too much electrode buffer was poured in the upper chamber.	The electrode buffer sometimes leaks from between the glass plate upper edge and seal packing to lower chamber because of capillary action. Set the upper buffer so that the fluid level will be 2 to 3mm below the upper edge of plate.
	Thin film gel is formed between spacer or glass plate and other plate.	This occurs when there is a gap between the spacer and the glass plate due to the consumption of the seal packing of the multi-lane gel hand-cast system. Discontinue use of the multi-lane gel hand-cast system and contact us (please check the back cover).
	Buffer sometimes leaks slightly.	About 1 to 2mL of buffer that contains SDS may sometimes leak over night or 24 hours. It does not affect electrophoresis.
	Seal packing is damaged.	If the seal packing is broken or peeled off the electrophoresis chamber, it is necessary to repair or replace it with a new one. Stop using and contact us (please refer to the back cover).
Sample solution will not sink into the well	The well is dirty or the gel is attached to it.	Wash the well with electrode buffer using a micro pipette or syringe. If dirt or gel fragment can still not be removed, remove it using a needle of the syringe, etc.
	Specific weight of sample solution is too small.	Concentration of glycerin or sucrose in the sample solution may be insufficient. Add the proper amount.
Electrophoresis time / position of band is different from usual	Output setting is incorrect.	When electrophoresis under constant current conditions, check the set current value. To electrophoresis 2 gels under the same conditions as when using 1 gel, double the set current value. If you want to electrophoresis 1 gel under the same conditions as when you electrophoresis 2 gels, set the current value to 1/2.
	Composition or concentration of buffer or gel is wrong.	If the composition can possibly be wrong, remake it.
	pH of gel preparation solution, solution B or solution C is incorrect (in the case of protein electrophoresis).	Keep pH of these solutions. Solution B: pH8.8 Solution C: pH6.8
	Electrode buffer is reused.	Do not reuse the electrode buffer.
	Gel is too old.	For precast gel, please use by the expiration date. In case of handmade gel, please use it as soon as possible on the same day. If you do not use it on the day, store it at 4 ° C and use it until next day. Otherwise the deterioration gradually progresses and the repeatability decreases.
	The speed of gel polymerization is different.	Changing the concentration of TEMED or Ammonium persulfate in gel will affect the polymerization rate and performance of gel. Please make gel according to the gel preparation method described in this manual as much as possible. The difference between room temperature and liquid temperature during polymerization also affects the polymerization rate. When polymerization temperature is around 25 ° C, good gel can be made with good reproducibility.
	Other factor	The electrophoresis speed is affected by salt concentration of sample solution, electrode buffer, and ambient temperature in addition to above items. In order to maximize reproducibility, it is necessary to keep each condition as constant as possible.
When staining, vertical stripes appear in a lane	Dirt or insoluble component is contaminated in the sample solution or well.	Remove the insoluble component in the sample solution by centrifugal separation, etc. Wash the well with electrode buffer.
The pattern is disturbed between lanes (smiling etc.).	The amount of buffer is small.	Immersion of the electrophoresis plate in the plenty volume of lower chamber buffer improves the gel temperature stability. Pour the appropriate volume of electrode buffer described in this manual into the lower chamber.
Pattern gets distorted between lanes (smiling etc.)	Buffer capacity of electrode buffer descends.	Please make the electrode buffer again.
	Salt concentration of sample solution is different between lanes.	Salt concentration of sample solution affects the electrophoresis speed. Keep the salt concentration as constant as possible by demineralization, condensation, dilution (for nucleic acid, ethanol precipitation), etc. Be careful in the case that the restriction enzyme buffer (salt concentration is different depending on the type) is contained or in the case of dilution line.
	There are air bubbles on the lower edge of gel.	If the air bubbles are very small and few, they rarely affect the gel, but large air bubbles may affect it.

Symptom	Cause	Coping technique
The electrophoresis pattern spreads out.	The electrode buffer is not sufficient. The current flowing on the side of the gel is occured	It is likely to occur when running at high voltage under conditions where the electrode buffer volume is low (below the buffer line). If you want to run with less electrode buffer, output power should set in about half.
		It is likely to occur when using the gel partially peeled off from the glass plate. Be careful to handle the gel plate so that it does not slip or peel off. Do not use old gels or excess the expiry date precast gels

7 Maintenance

7.1 Cleaning

Clean the electrophoresis chamber, plate holder, glass plate and comb referring to “Cleaning and Storage of Equipment” (page 26).

7.2 Inspection

After long-term storage, please read this instruction manual carefully and check at the same time when using for experiment again. If there is an abnormality at the time of inspection, please contact us without using the device

Electrophoresis Chamber

Set the plane plate MLB-02 on the seal packing side, hold it to the electrophoresis chamber with plate holder, add pure water to the upper chamber, and check for water leaks.

Check the electrode plug of the electrophoresis chamber for deformation, rattling or corrosion by visually observing the connection with the lead cable.

⚠Warning When inspecting the electrophoresis chamber by connecting the lead cable, do not connect the electrophoresis power supply.

Multi-lane Hand-cast gel system

Assemble the gel plate according to “Assembling the glass plate” (page 15), add pure water to 0.5 to 1 cm above the top of the notch, and leave it for about 1 hour to check for water leaks.

7.3 Consumable goods

The followings are consumables. Please replace it with a new ones when necessary.
For anything not listed, please contact your distributor or our company.

Model	Product name	Code No.
	Plate holder for WSE-1170 2pcs 1組	2322214
MLAB-12	Notch plate (for 1mm) (2plates)	2398301
MLB-02	Plane plate (2plates)	2398302
ML10-30	Multi-lane comb (1mm thickness / 30wells / PP mold) (2plates)	2398303

7.4 Warranty

Please refer to the warranty attached to the product for warranty prescription and terms.

8. Specifications

Product name	Multi-Lane Gel Electrophoresis system
Cat. number	WSE-1170
Plate size (W x H x D, mm)	160 x 100 x 5
Gel size（W x H x D,mm)	140 x 80 x 1.0
Total thickness of glass plate (mm)	5mm
Amount of Electrode buffer liquid	650-900mL
Available number of gels for simultaneous electrophoresis	Max. 2 gels
Electrophoresis gel isothermal method	Two-sided constant temperature by using upper and lower buffer solution
Electrode	Upper chamber is negative electrode and lower chamber is positive electrode
Safety measures	Safety cover
Location of use	Only use in a room, operation ambient temperature 5 to 40°C Operation ambient humidity: 5 to 70%RH, no dew condensation
Equipment and status of equipment	Portable equipment
Dimensions / weight Main unit	204mm（W）× 98.6mm（D）× 130mm（H） 0.95Kg
Standard components	Main unit Electrophoresis chamber, Lead cable with safety cover Instruction manual

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