



FT-BI9700

GeBaRunner Handbook

Ver. 01 - January 2013

GeBaRunner is covered by USA patent number 8,241,477 assigned to Gene Bio-Application Ltd.

Introduction

General Information

GeBaRunner together with GeBaGel constitutes a ready-to-use mini-gel system for high quality separation of proteins. GeBaGel is compatible with Tris-Glycine running buffer and contains no SDS, making it ideal for protein analysis under both native and denaturing conditions. GeBaGel is compatible with standard protein detection protocols, and has an extended shelf-life for increased convenience.

Scope of this document

These instructions provide information on how to perform polyacrylamide gel electrophoresis (PAGE) using GeBaRunner and GeBaGel. Additionally, protocols for post-staining and transfer are provided.

GeBaRunner Kit Contents

- GeBaRunner:1 unit
- Adaptors: 2 units
- Hex Wrench:1 unit
- Handbook:1 unit
- EC Declaration:1 unit

Protein gel electrophoresis overview

Protein analysis including separation of proteins using PAGE (GeBaGel) followed by either Western blotting or post-staining detection methods.

Important user information




Intended use

GeBaRunner and GeBaGel are intended for PAGE.

GeBaRunner and GeBaGel are intended for research use only, and shall not be used in any clinical procedures, or for diagnostic purposes.

Safety Notices

This user documentation contains WARNINGS, CAUTIONS and NOTICES concerning the safe use of GeBaRunner. See definitions below.

Sign	Meaning
	Warning indicates a hazardous situation which, if not avoided, could result in death or serious injury. It is important not to proceed until all stated conditions are met and clearly understood.
	Caution indicates a hazardous situation which, if not avoided, could result in minor or moderate injury. It is important not to proceed until all stated conditions are met and clearly understood.
	Notice indicates instructions that must be followed to avoid damage to the product or other equipment.

Regulatory information

Introduction

This section describes the directives and standards that are fulfilled by GeBaRunner.

CE Conformity

This product complies with the European directives listed in the table below, by fulfilling the corresponding harmonized standards. A copy of the Declaration of Conformity is available on request.

Directive	Title
2006/95/EC	Low Voltage Directive (LVD)
2004/108/EC	Electromagnetic Computability (EMC) Directive

CE Marking



The CE marking and the corresponding declaration of conformity, is valid for the instrument when it is:

- used as a stand-alone unit, or
- connected to other CE marked or
- connected to other products recommended or described in the user documentation, and

International Standards

This product fulfills the requirements of the following standards:

Directive	Title	Title
EN 61010-1, IEC 61010-1, UL 61010-1, CAN/CSA-C22.2 No.61010-1	Safety requirements for electrical equipment for measurement, control, and laboratory use.	Harmonized with 2006/95/EC
EN 61010-031, IEC 61010-031	Safety requirements for handheld probe assemblies for electrical measurement and test.	Harmonized with 2006/95/EC
EN 61326-1	EMC emissions and immunity requirements for electrical equipment for measurement, control and laboratory use.	Harmonized with 2006/95/EC

Regulatory compliance of connected equipment

Any equipment connected to GeBaRunner should meet the safety requirements of EN 61010-1/IEC 61010-1, or other relevant harmonized standards. Within EU, connected equipment must be CE marked.

Safety

Safety precautions














General precautions

Sign	Meaning
	Warning Do not use GeBaRunner in any other way than described in the GeBaRunner Instructions.
	Caution Operation of GeBaRunner should be performed by properly trained personnel only.
	Caution Do not use any accessories not supplied or recommended by Gene Bio-Application Ltd.






Installation

Sign	Meaning
	Warning Access to power switch and power cord. Do not block access to the Power switch on the Power supply and the Power cord. The Power switch must always be easy to access. The Power cord must always be easy to disconnect.
	Caution During electrophoresis, very low quantities of gases are produced at the electrodes. Make sure that GeBaRunner is run in a well-ventilated area.
	Caution GeBaRunner is designed for indoor use only.














Operation

Sign	Meaning
	Warning High voltage. Before use, check that GeBaRunner is completely dry on the outside before connecting it to a power supply. Wipe dry with a cloth.
	Warning Do not use GeBaRunner if it is not working properly, nor if it has suffered any damage, for example: <ul style="list-style-type: none">• damage caused by dropping the equipment• damage by spilling liquid onto it.
	Warning High voltage. Do not connect the high voltage cable to an external power supply if it is not working properly, nor if it has suffered any damage, for example damage to its plug or cable.
	Warning Electrical shock hazard. Always switch off the power supply and disconnect GeBaRunner from the power supply before removing the safety lid.
	Warning Do not operate GeBaRunner without a full functioning safety lid in place.
	Warning Do not exceed the maximum operating voltage of 200 V.
	Warning Do not operate the GeBaRunner Gel Box inside a metal tray.
	Warning Never operate/run the GeBaRunner without both high voltage plugs connected to the external power supply.
	Warning High voltage. Do not overfill GeBaRunner with running buffer. Maximum volume is 90mL per tank.
	Warning Do not operate GeBaRunner close to strong magnetic fields.
	Caution Hazardous substances. When using hazardous chemicals, take all suitable protective measures, such as wearing protective glasses and gloves resistant to the substances used. Follow local and/or national regulations for safe operation.
	Caution Do not move GeBaRunner during operation.
	Notice To prevent equipment damage, only operate GeBaRunner in a horizontal position.






Maintenance

Sign	Meaning
	Warning Electrical shock hazard. Always switch off the Power supply and disconnect GeBaRunner from the Power supply before maintenance and cleaning.
	Warning Cleaning. Do not autoclave, bake or microwave GeBaRunner.
	Warning Cleaning. Do not wash GeBaRunner in a mechanical washer.
	Warning Cleaning. Do not use abrasive creams or scourers.
	Notice Cleaning. Clean the instrument with distilled water and wipe dry with a soft damp tissue. Let the instrument dry completely before use..

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	Notice Cleaning. Clean the instrument with distilled water and wipe dry with a soft damp tissue. Let the instrument dry completely before use..

Labels

Labels on GeBaRunner

The illustration below shows an example of the label that is attached to the bottom of the GeBaRunner.



Safety Symbols

Sign	Meaning
	Warning! Read the user documentation before using the instrument. Do not open any covers or replace parts unless specifically stated in the user documentation.
	Warning! High Voltage. Always make sure that the instrument is disconnected from electric power before opening the lid or disconnecting any electric equipment.
	The instrument complies with the requirements for electromagnetic compliance (EMC) in Australia and New Zealand. N3732.
	The instrument complies with applicable European directives.
	The instrument is listed according to TÜV Rheinland.
	The instrument is protected throughout by double insulation.

Labels Concerning

Hazardous Substances

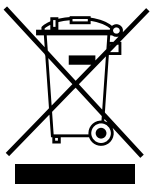
Sign	Meaning
	This symbol indicates that electrical and electronic equipment must not be disposed of as unsorted municipal waste and must be collected separately. Please contact an authorized representative of the manufacturer for information concerning the decommissioning of equipment.
	This symbol indicates that the product contains hazardous materials in excess of the limits established by the Chinese standard SJ/T11363-2006 Requirements for Concentration Limits for Certain Hazardous Substances in Electronic Information Product.

Recycling Information

GeBaRunner shall be decontaminated before decommissioning and all local regulations shall be followed with regard to scrapping of the equipment.


When taking GeBaRunner out of service, the different materials must be separated and recycled according to national and local environmental regulations.

Waste of electrical and electronic equipment must not be disposed as unsorted municipal waste and must be collected separately. Please contact an authorized representative of the manufacturer for information concerning the decommissioning of equipment.



Declaration of Hazardous Substances (DoHS)

The following product pollution control information is provided according to SJ/T11364-2006 Marking for Control of Pollution caused by Electronic Information Products.

Sign	Meaning
	<p>This symbol indicates the product contains hazardous materials in excess of the limits established by the Chinese standard SJ/T11363-2006 Requirements for Concentration Limits for Certain Hazardous Substances in Electronic Information Products. The number in the symbol is the Environment-friendly Use Period (EFUP), which indicates the period during which the toxic or hazardous substances or elements contained in electronic information products will not leak or mutate under normal operating conditions so that the use of such electronic information products will not result in any severe environmental pollution, any bodily injury or damage to any assets. The unit of the period is "Year".</p> <p>In order to maintain the declared EFUP, the product shall be operated normally according to the instructions and environmental conditions as defined in the product manual, and periodic maintenance schedules specified in Product Maintenance Procedures shall be followed strictly.</p> <p>Consumables or certain parts may have their own label with an EFUP value less than the product.</p> <p>Periodic replacement of those consumables or parts to maintain the declared EFUP shall be done in accordance with the Product Maintenance Procedures.</p> <p>This product must not be disposed of as unsorted municipal waste, and must be collected separately and handled properly after decommissioning.</p>

List of Hazardous substances and their concentration

Value	Meaning
0	Indicates that this toxic or hazardous substance contained in all of the homogeneous materials for this part is below the limit requirement in SJ/T11363-2006.
X	<p>Indicates that this toxic or hazardous substance contained in at least one of the homogeneous materials used for this part is above the limit requirement in SJ/T11363-2006.</p> <ul style="list-style-type: none">• Data listed in the table represents best information available at the time of publication• Applications of hazardous substances in this medical device are required to achieve its intended clinical uses, and/or to provide better protection to human beings and/or to environment, due to lack of reasonably (economically or technically) available substitutes.

List of Hazardous substances

	Components name					
	Pb	Hg	Cd	Cr6+	PBB	PBDE
GeBaRunner GR-Mini	0	0	0	0	0	0

System description

GeBaRunner

Specifications


Parameters	Parameters Value
GeBaRunner Dimensions	167 X 148 X 43.5mm (W x H x D)
GeBaGel Dimensions	116.8 X 84 X 15.6mm (W x H x D)
Maximum Voltage	200 V DC
Operating Temperature	40°C-40°C
Maximum Power	20W
Maximum Current	100mA
Degree of Protection	IPX5

General

The picture below shows the main parts of GeBaRunner (the safety lid is not shown).

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List of Hazardous substances

	Components name					
	Pb	Hg	Cd	Cr6+	PBB	PBDE
GeBaRunner	0	0	0	0	0	0
GR-Mini	0	0	0	0	0	0

System description

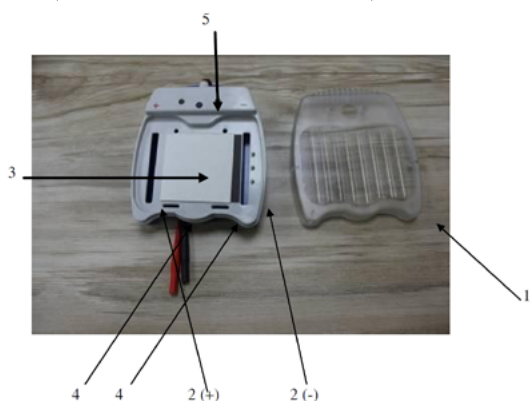
GeBaRunner

Specifications

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GeBaGel Dimensions	116.8 X 84 X 15.6mm (W x H x D)
Maximum Voltage	200 V DC
Operating Temperature	40°C-40°C
Maximum Power	20W
Maximum Current	100mA
Degree of Protection	IPX5

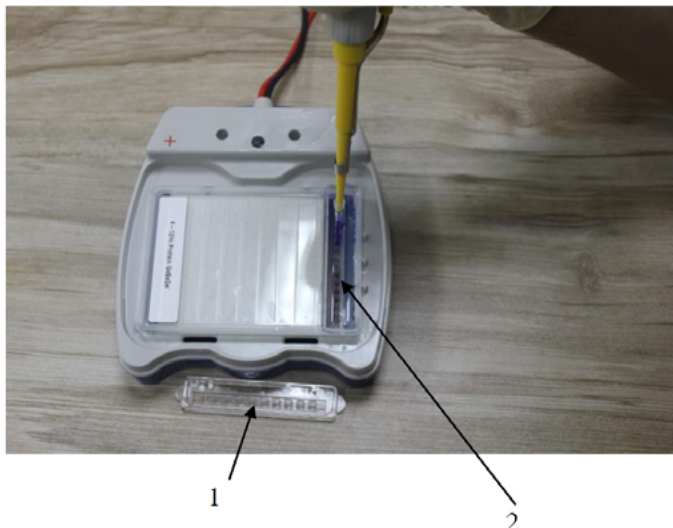
General

The picture below shows the main parts of GeBaRunner (the safety lid is not shown).



Part	Description
1	Safety lid
2 (-)	Running Buffer loading slit (Cathode side)
2 (+)	Running Buffer loading slit (Anode side)
3	Thermal equalizer plate
4	Air bubble draining slit
5	Green on-off indicator light

GeBaGel



Part	Description
1	Comb
2	Well container

Power Supply

GeBaRunner is powered by an external power supply. If other power supply with a Ø 4mm, deep socket (>6 mm internal distance between surface and socket) is used, it is recommended to use the adapter delivered with GeBaRunner.

Safety lid

GeBaRunner is equipped with a safety lid, which shall be placed on top of the base unit before a power supply is connected. The safety lid has two magnets which will activate current only when their magnetic fields are in contact with the electrodes inside the base unit of GeBaRunner. This means that GeBaRunner is only powered when the safety lid is in the correct position. Removing the safety lid will switch off the high voltage to the electrodes.

Sign	Meaning
	Warning Do not operate GeBaRunner without the safety lid.

Protocols for GeBaGel User

General

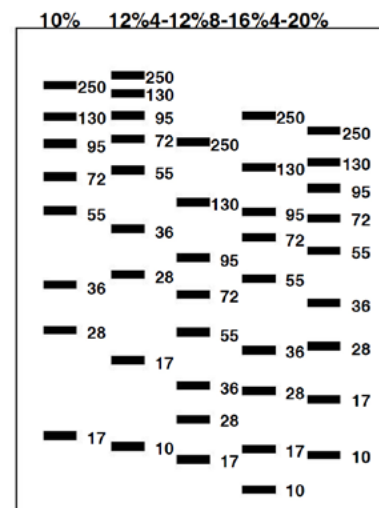
GeBaGel is designed for PAGE using GeBaRunner.

GeBaGel are available as homogenous and gradient gels in the following concentrations:

- 10%
- 12%
- 4-12%
- 8-16%
- 4-20%

For information regarding separation ranges for each GeBaGel, see Separation range, on page below.

Separation range



Gel Specifications




Parameter	Value
Gel Matrix	Acrylamide/Bis-Acrylamide
Gel Thickness	1.4mm
Gel Size	7.5 X 8cm (W x H)
Well configuration	1+1, 2+1, 10 and 15 wells
Well volumes	700, 200, 35, and 2mL/well respectively
Stacking gel	4%
Buffer system	Tris-Hcl
Shelf life	12 months

Buffer solutions

Solution	Composition
2X SDS Sample buffer	125mM Tris-HCl, pH 6.8, 4% (w/v) SDS, 20% glycerol, 0.2mg/mL of bromphenol blue, 31mg/mL of DTT.
Denatured running buffer	25mM Tris, 192mM Glycine ,0.1% SDS
Native running buffer	25mM Tris, 192mM Glycine

Installation

Precautions

Sign	Meaning
	Warning Access to power switch and power cord. Do not block access to the Power switch on the Power supply and the Power cord. The Power switch must always be easy to access. The Power cord must always be easy to disconnect.
	Caution During electrophoresis, very low quantities of gases are produced at the electrodes. Make sure that GeBaRunner is run in a well ventilated area.
	Caution GeBaRunner is designed for indoor use only.


Connecting to power supplies

The power supply must have a rigid insulating sleeve output connector that fulfills the requirements for 200V (DC). If a power supply with Ø 4mm, deep socket (>6mm internal distance between surface and socket) is used, it is recommended to use the adapter delivered with GeBaRunner. For mounting instructions, see mounting the adapter, on next page.

Note: GeBaRunner is delivered with two adapters, identical apart from color, one black (-) and one red (+) (shown below).

Mounting the adapter

1. Turn off the power supply and remove the electrical contact from the power supply outlet.
2. Insert the adapter in the power supply output socket.

Sign	Meaning
	Warning Do not use the adapter in Ø 4mm socket with <6mm internal distance between surface and socket.



3. Tighten the set screw using the 1.5mm Hex wrench provided with the GeBARunner, to lock the expandable plug of the adapter into the socket.



4. Make sure that the adapter is locked into the socket.

Protocols

Electrophoresis Protocol

Sign	Meaning
	Warning Do not use GeBaRunner if it is not working properly, nor if it has suffered any damage, for example: 1. damage caused by dropping the equipment 2. damage by spilling liquid onto it.
	Warning Do not connect the high voltage cable to an external power supply if it is not working properly, nor if it has suffered any damage, for example damage to its plug or cable.

Buffer solutions

1. Prepare 1× running buffer by diluting 19mL of 10× Running Buffer (Denatured or Native running buffer) in 171mL water. 190mL buffer is sufficient for one electrophoresis gel.
2. Add 90mL of 1× running buffer to each tank of GeBaRunner.
3. Cut open the gel package and gently remove GeBaGel from the package.
4. Rinse the gel cassette with distilled water. Peel off the tapes from the two legs of the cassette.
5. Place GeBaGel in GeBaRunner so that the wells side of the cassette faces toward the cathode (-) and the other cassette leg faces toward the anode (+).
6. Wiggle the comb back and forth, and bring it straight up from the cassette to make the wells available for sample loading.
Note: Small gel pieces can be detached from the gel. This will not affect gel performance.
Note: Do not discard the comb.
7. Add 7 ml of 1X running buffer to the wells container.


Sample loading

1. Prepare samples by adding sample and 2X sample buffer in a 1:1 mixture.
Example: for 20mL sample, add 20mL sample buffer.
Note: For native conditions: use a sample buffer without SDS and DTT.
2. Incubate the samples at 95°C for 5 minutes.
3. Spin down the samples quickly in a microcentrifuge, and load the samples directly into the wells in the gel.
Note: A maximum of 0.5mg/band per well of sample can be loaded. Overloading may cause smearing and distortion.
Note: To ensure uniform mobility, load an equal volume of 1X SDS sample buffer into any unused well.
4. Place the safety lid on top of GeBaRunner.

Running conditions

Run the gel at 200V for 50-55 minutes.

Optionally: Run the gel at 100V for 10 minutes, continue run the gel at 200V for 45-50 minutes.

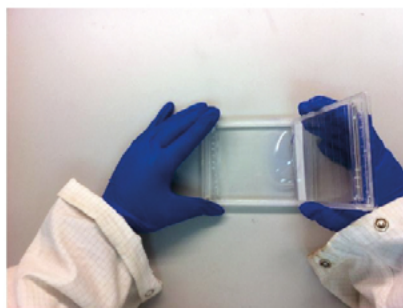
Sign	Meaning
	Warning Do not use over 200V.

Removing the gel from the cassette

1. Once the run is completed, switch off the power, disconnect the electrodes, remove the safety lid and remove the cassette from GeBaRunner.
2. Open the cassette by inserting the edge of the comb in the slot opposite the sample wells, and twist (see picture 1).
3. Remove the top plate from the gel cassette and allow the gel to sit on the bottom plate.
4. Cut the stacking gel, with the end of the top plate, approximately 2cm downstream of the wells. Cut the gel to remove the front (see picture 2).
5. Hold the gel cassette over a container with a suitable buffer, with the gel facing downwards.
Gently push a tweezers between the gel and the cassette until the gel is removed from the cassette.



Picture 1:
Opening the cassette with the comb



Picture 2:
Cutting the gel with the top plate

Post-Staining of the gel

The GeBaGel can be stain in any standard staining method used to stain total proteins in polyacrylamide gels such as any Coomassie Staining types, Deep Purple Staining or Silver Staining.

Gene Bio-Application Ltd. is recommending using CooBlue FX Single Step staining solution ([1B8100](#))

CooBlue FX Single Step protocol

Important: This protocol is adjusted for 8 x 10 cm mini gel 1.4 mm thick.

Before Use: Mix the CooBlue FX Single Step solution by gently inverting the bottle a few times (do not shake the bottle).

Important: 1. multiple washes prior to staining are NOT required.

2. fixing Step prior to staining is NOT required.

3. a destaining step post staining is NOT usually needed.

Caution: Use caution while using the stain in a microwave oven if using the optionally protocol. Do not overheat the staining solutions.

1. After electrophoresis, cut the stacking gel and remove the gel directly into a container containing 20-25mL of the CooBlue FX -NoPreWash solution. Gently shake the container.

2. Colored protein bands will start to develop immediately and a suitable intensity is typically achieved after 15 minutes when incubation at room temperature with gentle shaking.

Optionally: Microwave at High Power for 30 seconds remove it to a shaker and shake it gently. A suitable intensity is typically achieved after only 5 minutes.

3. Photograph your gel when the required intensity has been achieved. Gels can be kept in staining solution for overnight, see that the gel remains covered with the solution. Close container to eliminate evaporation. Alternatively: the gel can be stored in ultrapure water after staining for at least 1 hour.

4. Washing the gel with water can enhances staining sensitivity and increase staining over background.

Notes: After using CooBlue FX Single Step protein staining solution, close tightly the cap and store again at 4oC. Do not freeze.

5. Once used, the staining solution should be discarded and cannot be reused.

The CooBlue FX Single Step is provided as ready to use solution and should not be diluted.

Protocol for Destaining Protein Bands for MS analysis:

1. Excise the protein band of interest and transfer to clean Eppendorf tube.

2. Add 1 ml of 30% ethanol or 30% acetone or 30% acetic acid.

3. Incubate for 20min (heating to 60-70oC will increase the rate of destaining).

4. Decant supernatant and repeat step 2&3 at least 3 times or until gel is clear.

Transfer protocol

Transfer buffer: 25mM Tris, 192mM glycine, 20% methanol. Prepare fresh and used pre chilled buffer.

Preparing the membrane

1. Cut the desired membranes 8 cm X 9 cm

2. Pre-wet the membranes:

a. NitroCellulose (NC) membrane in distilled water for five minutes followed by 10 minutes in cold transfer buffer.

b. PVDF membrane first in methanol for 20 seconds, then in distilled water for 5 minutes and finally, in cold transfer buffer for at least 10 minutes.

Preparing the Gel

Once the electrophoresis run is completed

1. Disassembly the gel and open the gel cassette.

2. Truncate the stacking and front of the gel.

3. Place the gel in pre-chilled transfer buffer for 10-15 minutes.

Preparing the transfer tank

1. Fill the transfer unit with transfer buffer, the buffer level should be between min and max and according of the apparatus manufacturer.

2. Place the opened cassette with the anode side (+) at the bottom of the pre-filled tray with at least 3cm of transfer buffer.

3. Build the stack on the anode side, according to Figure below.

4. The sandwich can be prepared at the opposite direction.

Layer 6
Layer 5: Blotting paper
Layer 5: Blotting paper
Layer 4 Gel
Layer 3: NC or PVDF Membrane
Layer 2: Blotting paper
Layer 2: Blotting paper
Layer 1: Sponge

+

Figure 1: Schematic picture of the stack (i.e. sandwich)

1. Layer 1: Place a 3mm-thick foam sponge and press gently until the air is expelled.
2. Wet the blotting filter in transfer buffer.
3. Layer 2: Place the two pre-wetted blotting papers on the sponge and press gently until the air is expelled.
4. Layer 3: Place the desired pre-wetted membranes on layer 2 and remove all air-bubbles!
5. Equilibrate the desired gel in cold transfer buffer for 15 minutes.
6. Layer 4: Place the gel on the membrane.
7. Layer 5: Cover the gel with two pre-wetted sheets of blotting paper.
8. Layer 6: Finally place a sponge of 3mm and again press gently to expel trapped air.
9. Close the cassette and press lightly to lock the tabs.
10. Place the cassette into the tank.

Running conditions for transfer tank

Parameter	Value
Voltage	25V (maximum 400mA)
Temperature	40°C
Time	2.5 hours (with constant stirring)

Semi-dry transfer

Three Buffers System for NC or PVDF (Semi Dry Protocol)

The major advanced with this protocol is that large proteins (such as 200kDa) and small proteins (such as 11kDa) are transfer equally. That eliminates the well-known phenomena that small proteins usually transfer much faster than the large one. Also, this system allows a higher resolution of the proteins and the transfer kinetics is improved.

Anode Buffer I: 300mM Tris-HCl, pH 10.4, 20% (v/v) Methanol.

Anode Buffer II: 25mM Tris-HCl, pH 10.4, 20% (v/v) Methanol

Cathode Buffer: 25mM Tris-HCl, pH 9.4, 40mM Capronic acid, 20% (v/v) Methanol.

Alternative Cathode Buffer: 40mM D, L-norleucine (hard to dissolve, 70 oC), 25mM Tris-HCl, pH 9.4, 20% (v/v) Methanol.

Step 1:

Incubate the gel in cathode buffer for 5 min.

Step 2:

Moisten the anode with anode buffer I.

Step 3:

Cut 6 blotting papers to the size of the gel.

Step 4:

Soak 2 blotting papers in anode buffer I and place them on the anode.

Step 5:

A further layer of the adapted blotting paper is soaked with the anode buffer II added to the sandwich.

Step 6:

Pre-wet the membrane (NC) cut to the size of the gel with anode buffer II. Subsequently, the membrane is placed on the transfer sandwich. (PVDF membrane have to be successively incubate in methanol (1-2 seconds), in H₂O (5min) and in anode buffer II (5min)).

Step 7:

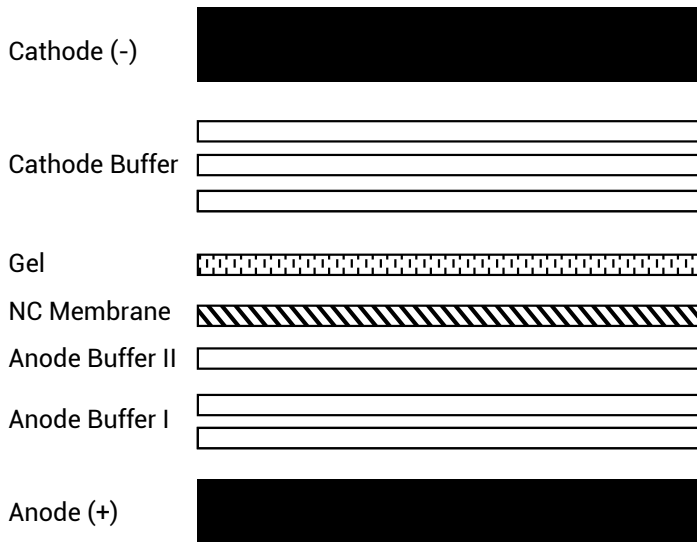
The gel is put on top of the membrane.

Step 8:

Three layers of blotting papers are soaked in cathode buffer and placed on top of the gel.






Important: Avoid the inclusion of air bubbles in the blotting sandwich Max. Amperage for use is 2.5mA/cm² gel, usually for 30-45min.

See Figure for sandwich assembly.



Maintenance

Precautions

Sign	Meaning
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	Warning Cleaning. Do not autoclave, bake or microwave GeBaRunner.
	Warning Cleaning. Do not wash GeBaRunner in a mechanical washer.
	Warning Cleaning. Do not use abrasive creams or scourers.
	Notice Cleaning. Clean the instrument with distilled water and wipe dry with a soft damp tissue. Let the instrument dry completely before use.

Cleaning

1. Discard the running buffer.
2. Rinse thoroughly with distilled water.
3. Let the instrument air dry or wipe with a soft damp tissue.
4. Keep power cables and connectors dry.

Storage

Store the GeBaRunner in a dry location at 0°C to 65°C.

Troubleshooting

Problems	Possible causes	Remedies
Low or no current during the run	Interrupted circuit	<ul style="list-style-type: none">• Add some running buffer to each tank of GeBaRunner• Ensure that the tape is removed from the gel cassette• Ensure that the lid is in place
Streaking of proteins	<ul style="list-style-type: none">• Sample overload• Poorly soluble or weakly charged particles (such as carbohydrates in sample)• High salt concentration in the sample• Contaminants such as membranes or DNA complexes in the sample	<ul style="list-style-type: none">• Load the appropriate amount of protein• Centrifuge the samples• Change the pH of the sample buffer• Heat the sample together with SDS• Decrease the salt concentration of the sample solution using dialysis or gel filtration
Bands difficult to distinguish	Incorrect gel selection, sample overloading	<ul style="list-style-type: none">• Select a gel that separates in the desired molecular weight range• Reduce loaded protein amount• For proteins of similar molecular weight, a 2-D separation may be required
Sample spreading across the gel	<ul style="list-style-type: none">• Excess salt in the sample• Too much protein applied to the gel	<ul style="list-style-type: none">• Reduce salt by ultrafiltration• Optimize the amount of protein applied to the gel
Sample contains appreciable carbohydrate		Remove the carbohydrate by enzymatic or chemical means
Sample contains lipoproteins		<ul style="list-style-type: none">• Use a gradient gel• Try addition of a non-ionic detergent
Protein denaturation and band inversion	Excessive heating	<ul style="list-style-type: none">• Start with cold buffer (<15°C)
Diffuse protein zones in the gel after staining	<ul style="list-style-type: none">• SDS still present in the gel• Insufficient reduction of samples	<ul style="list-style-type: none">• Wash the gels extensively (3 x 10 minutes) with ultrapure water and use 30% methanol to destain the gel• Use 10% TCA to fix the proteins in the gel. Add extra reducing agent or change the reducing agent.
No green light, low current (<20mA) at start.	Incomplete circuit	<ul style="list-style-type: none">• Check all power connections• Check buffer level in tanks• Ensure that the gel cassette is in place• Ensure that there is no tape left on the gel cassette

Problems	Possible causes	Remedies
Green light, but only approx. 35mA at start.	Leak between tanks, with or without gel cassette	<ul style="list-style-type: none"> • Check that the tanks are not overfilled • Check if the unit is damaged and needs to be replaced
Green light, but approx. 65mA at start	Leak between tanks with gel cassette	<ul style="list-style-type: none"> • Check that tanks are not overfilled • Check if the unit is damaged and needs to be replaced

Ordering information

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