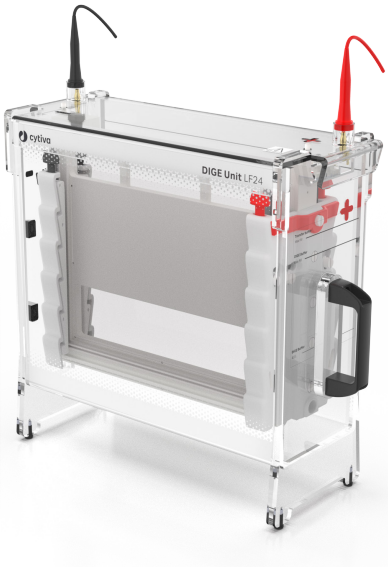
# Amersham™ DIGE Unit LF24

## Operating Instructions

Original instructions




cytiva.com

# Table of Contents

# 1 Introduction

## About this chapter

This chapter contains information about this manual and associated user documentation, important user information and intended use of the product.

## In this chapter

# 1.1 Important user information

## Read this before operating the product

**All users must read the entire *Operating Instructions* before installing, operating, or maintaining the product.**

Always keep the *Operating Instructions* at hand when operating the product.

Do not install, operate, or perform maintenance on the product in any other way than described in the user documentation. If you do, you may be exposed or expose others to hazards that can lead to personal injury and you may cause damage to the equipment.

## Intended use of the product

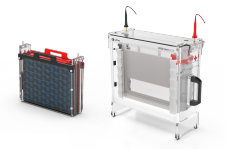
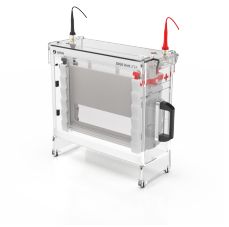
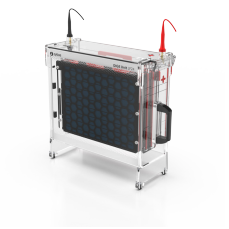
The Amersham™ DIGE Unit LF24 instrument is intended for protein separation with two-dimensional (2-D) electrophoresis. The electrophoresis module is designed exclusively for use with the precast DIGE Gels LF24.

The Amersham Transfer Unit LF24 module is intended for electrotransfer of proteins. The Transfer Unit LF24 module must be used together with the DIGE Unit LF24 tank.

The product is intended to be used by trained laboratory staff members in research laboratories within academia and industry. The product must not be used in any clinical procedures, or for diagnostic purposes.

## Product definition

The following table shows the definitions for the product, the electrophoresis module and the electrotransfer module used in this manual.

| Word | Definition | Illustration |
|---|---|---|
| The product | The combination of the DIGE Unit LF24 instrument and the Transfer Unit LF24 module. |  |
| The electrophoresis module | The DIGE Unit LF24 instrument (i.e. the tank and the gel cassette holder). |  |
| The electrotransfer module | The DIGE Unit LF24 tank and the Transfer Unit LF24 module (i.e. the tank, the transfer cassettes and the transfer cassette holder). |  |
| The gel | The precast DIGE Gels LF24. |  |

## Prerequisites

In order to operate the product in the way it is intended:

- The user must understand the concepts of electrophoresis and electrotransfer techniques.
- The user must read and understand the Safety Instructions chapter in the *Operating Instructions*.
- The product must be installed in accordance with the site requirements and instructions in the *Operating Instructions*.

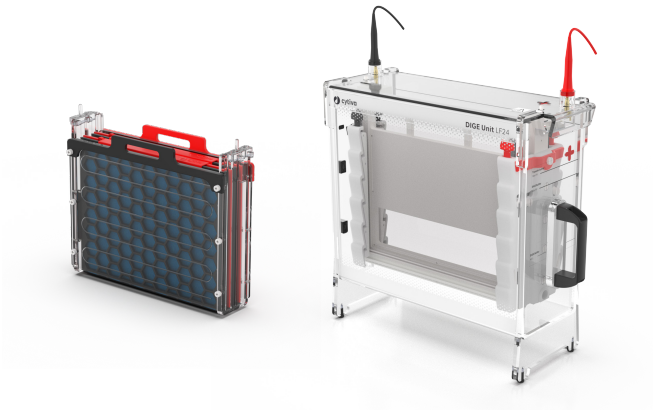
# 1.2 About this manual

## Purpose of this manual

This manual provides information needed to install, operate and maintain the product in a safe way.

## Scope of this manual

This Operating Instruction manual is valid for the DIGE Unit LF24 instrument and the Transfer Unit LF24 module. The illustration below shows the DIGE Unit LF24 instrument and the Transfer Unit LF24 module.



## Typographical conventions

Software items are identified in the text by ***bold italic*** text.

Hardware items are identified in the text by **bold** text.

***Tip:*** *The text can include clickable hyperlinks to reference information.*

## Notes and tips

***Note:*** *A note is used to indicate information that is important for trouble-free and optimal use of the product.*

***Tip:*** *A tip contains useful information that can improve or optimize your procedures.*

# 1.3 Associated documentation

## Introduction

This section describes the user documentation delivered with the product, and how to find related literature that can be downloaded or ordered from Cytiva.

## User documentation for the product

The user documentation is listed in the table below.

The *Operating Instructions* and translations are provided on the User Documentation CD.

| Documentation and product code | Main contents |
|---|---|
| *Amersham DIGE Unit LF24 Operating Instructions, 29701249*<br>(this document) | Instructions needed to prepare and operate the electrophoresis module and the electrotransfer module in a correct and safe way.<br>System overview, site requirements, and instructions for moving the product.<br>Instructions for basic maintenance and troubleshooting. |
| *Amersham DIGE Unit LF24 Cue Cards, 29701879* | High-level instructions needed to prepare buffers and operate the electrophoresis module and the electrotransfer module. |
| *DIGE Gels, DIGE Gels LF24 and DIGE Buffer Kit Cue Cards, 28946086* | High-level instructions needed to prepare buffers and run the DIGE gel. |
| *DIBE coverage analysis Instructions for Use, 29657336* | Instructions needed to perform the DIBE™ coverage analysis. This analysis is based on two-dimensional differential in-blot electrophoresis (2-D DIBE). |
| *2-D Electrophoresis Principles and Methods Handbook, CY14825* | Methodology handbook about 2-D electrophoresis. |
| *Molecular Imaging: Principles and Methods Handbook, CY14828* | Methodology handbook about molecular imaging, covering topics such as imaging with fluorescent dyes and western blotting. |

## User documentation and other literature on the web

User documentation and other literature related to the product can be downloaded from the web. Follow the steps below to access the documentation.

| Documentation | Download |
|---|---|
| DIGE Unit LF24 and Transfer Unit LF24 documentation | 1. Go to *cytiva.com*, and locate the Electrophoresis and isoelectric focusing product page.<br>2. Click on DIGE Unit LF24.<br>3. Navigate to ***RELATED DOCUMENTS***.<br>4. Select the type of document and download the chosen literature. |
| Cytiva handbooks | 1. Go to *cytiva.com/handbooks*.<br>2. Download handbooks such as:<br>• *2-D Electrophoresis Principles and Methods Handbook, CY14825*<br>• *Molecular Imaging: Principles and Methods Handbook, CY14828* |
| Other user documentation | 1. Go to *cytiva.com/instructions*.<br>2. Enter the product code of the literature in the search field. See *User documentation for the product, on page 8* for product codes of relevant literature.<br>3. Click on the link of the target document, then click on the download button. |

# 1.4 Abbreviations

## Introduction

This section explains abbreviations that appear in the user documentation for DIGE Unit LF24.

## Abbreviations

| Abbreviation | Definition (English) | Translation (local language) |
|---|---|---|
| 2-D | Two-dimensional | Two-dimensional |
| 2-D DIBE | Two-dimensional differential in-blot electrophoresis | Two-dimensional differential in-blot electrophoresis |
| DIBE | Differential in-blot electrophoresis | Differential in-blot electrophoresis |
| 2-D DIGE | Two-dimensional fluorescence difference gel electrophoresis | Two-dimensional fluorescence difference gel electrophoresis |
| DTT | Dithiothreitol | Dithiothreitol |
| DIGE | Difference gel electrophoresis | Difference gel electrophoresis |
| ELISA | Enzyme-linked immunosorbent assay | Enzyme-linked immunosorbent assay |
| HCP | Host cell protein | Host cell protein |
| IEF | Isoelectric focusing | Isoelectric focusing |
| IPG | Immobilized pH gradient | Immobilized pH gradient |
| pI | Isoelectric point | Isoelectric point |
| PPE | Personal protective equipment | Personal protective equipment |
| PVDF | Polyvinylidene fluoride | Polyvinylidene fluoride |
| SDS | Sodium dodecyl sulphate | Sodium dodecyl sulphate |
| SDS-PAGE | Sodium dodecyl sulphate-polyacrylamide gel electrophoresis | Sodium dodecyl sulphate-polyacrylamide gel electrophoresis |
| Tris | Tris(hydroxymethyl)aminomethane | Tris(hydroxymethyl)aminomethane |

# 2 Safety instructions

## About this chapter

This chapter describes safety precautions, labels and symbols that are attached to the product. In addition, the chapter describes emergency and recovery procedures.

## In this chapter



## Important

**WARNING**

**All users must read and understand the entire contents of this general safety chapter, and the specific safety precautions information in each subsequent chapter of this manual to become aware of the hazards involved.**

# 2.1 Safety precautions

## Introduction

The product is powered by an electrophoresis power supply and handles materials that can be hazardous.

Before installing, operating, or maintaining the system, you must be aware of the hazards described in this manual.

## Definitions

This user documentation contains safety notices (WARNING, CAUTION, and NOTICE) concerning the safe use of the product. See definitions below.

**WARNING**

**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**

**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**

**NOTICE** indicates instructions that must be followed to avoid damage to the product or other equipment.

## General precautions

The following general precautions must be considered at all times. There are also context-related precautions, which are written in their respective chapters.

**WARNING**

**Risk assessment**. The use of the product may cause hazardous situations. Perform a risk assessment prior to use.

**WARNING**

Do not operate the product in any other way than as described in the user documentation.

**WARNING**

Only properly trained personnel are allowed to operate and maintain the product.

**WARNING**

**Accessories.** Use only accessories supplied or recommended by Cytiva.

**WARNING**

Do not use the product if it is not working properly, or if it has suffered any damage including:

- damage to the power lead or its plug,
- damage caused by dropping the product,
- damage caused by splashing liquid onto the product.

**WARNING**

Always use appropriate Personal Protective Equipment (PPE) during operation and maintenance of this product.

**WARNING**

**Power lead.** Make sure the power lead is undamaged. Damaged power lead may result in fire or electric shock.

**WARNING**

**Hazardous substances.** When using hazardous chemicals, take all suitable protective measures, such as wearing protective clothing, glasses and gloves resistant to the substances used. Follow local and/or national regulations for safe operation and maintenance of the product.

**WARNING**

Never exceed the operating limits stated in this document and on the system label. Operation of the product outside these limits can damage equipment and cause personal injury or death.

**WARNING**

**Shock hazard**. The surface of the tank must be dry before it is connected to the electrophoresis power supply. Liquid on the outside of the tank can result in fire or electric shock.

**CAUTION**

The product is designed for indoor use only.

**CAUTION**

Handle the glass components with care! Wear appropriate personal protective equipment (PPE).

**CAUTION**

**Pinch hazard**. Be careful to avoid squeezing or crushing injuries when handling the product and the accessories.

## Flammable liquids

**WARNING**

**Ventilation system.** A fume hood or similar ventilation system must be used when working with flammable or noxious substances.

**CAUTION**

**Fire hazard**. Do not exceed the maximum buffer temperature. Heating flammable liquids above their flash point can cause fire.

**CAUTION**

**Fire hazard.** Make sure that the buffer temperature does not exceed 30°C when using ethanol in the transfer buffer. Heating flammable liquids above their flash point can cause fire.

# 2.2 Labels and symbols

## Introduction

This section describes the nameplate, labels, and other safety and regulatory information attached to the product.

## Nameplate

The nameplate provides information about the model, manufacturer, and technical data.

## Description of symbols and text

The following symbols and text may be present on the name plate:

| Symbol / text | Description |
|---|---|
|  | **Warning!** Read the user documentation before using the product. Do not open any covers or replace parts unless specifically stated in the user documentation. |
| **Mfg. Date** | Year (YYYY) and month (MM) of manufacture |
| **Max. Voltage** | Electrical rating: Voltage (VDC ⎓) |
| **Max. Current** | Electrical rating: Max. current (mA) |
| **Max. Power** | Electrical rating: Max. power (W) |

## Safety labels

The table below describes the various safety labels that may be found on the product.

| Label | Description |
|---|---|
|  | **Warning!** Read the user documentation before using the system. Do not open any covers or replace parts unless specifically stated in the user documentation. |

# 2.3 Emergency procedures

## Introduction

This section describes how to shut down the product in an emergency situation.

## Precautions

**WARNING**

**Access to power switch and power cord with plug**. Do not block access to the power switch and power cord of the electrophoresis power supply. The power switch must always be easy to access. The power cord with plug must always be easy to disconnect.

## Emergency shutdown

In an emergency situation, follow the steps below to stop the run:

| Step | Action |
|---|---|
| 1 | Turn off the electrophoresis power supply in accordance with the instructions from the manufacturer. |
| 2 | If required, disconnect the mains power cord of the electrophoresis power supply.<br>*Result:*<br>The run is interrupted immediately. |

When the emergency situation is resolved, continue the run as described in *Chapter 5 Operation, on page 34*.

# 3 System description

## About this chapter

This chapter gives an overview of the product, and a brief description of its function.

## 2-D Electrophoresis

In two-dimensional fluorescence difference gel electrophoresis (2-D DIGE), proteins are first separated according to their isoelectric point (pI) and then by their molecular mass. For the first dimension, the sample is loaded on an immobilized pH gradient (e.g. IPG strips) and isoelectric focusing (IEF) is performed, for instance, with the IPGphor™ isoelectric focusing unit. For the second dimension, the sample, focused in an IPG strip, is placed on the cathodic (upper) surface of the gel and sealed in place with agarose. The gel cassette is then inserted into the gel cassette holder. An unused slot is filled with the dummy plate. The buffer seal is an effective current and liquid barrier. The second dimension is performed by sodium dodecyl sulphate-polyacrylamide gel electrophoresis (SDS-PAGE), for instance, with the DIGE Unit LF24 instrument. For more information on 2-D electrophoresis, see *Handbook, CY14825*.

The DIGE Unit LF24 instrument is designed to run up to two gels. The unit is designed exclusively for use with the precast DIGE Gels LF24 available from Cytiva. The precast gels are low fluorescent 12.5% polyacrylamide gels, 24 cm wide, and intended for 2-D electrophoresis. The precast gels provide a shelf-life of up to 12 months. For more information on DIGE Gels LF24, see *Cue Card, 28946086*.

## Electrotransfer

After electrophoresis, the separated proteins in the gel can be transferred to a membrane with the electrotransfer module. The transferred proteins can then be analyzed and detected with specific antibodies. For more information on protein transfer, see *Handbook, CY14828*.

The Transfer Unit LF24 module is designed for wet protein transfer. Wet electrotransfer is an efficient and robust method to create blots of the highest quality in terms of distinct, sharp spots. The voltage applied during the wet electrotransfer generates heat and increases the buffer temperature. If the analyzed sample contains heat-sensitive proteins it is recommended to maintain a low temperature during electrotransfer, with cooled buffer. See *Cooling options for electrotransfer, on page 53* for more information.

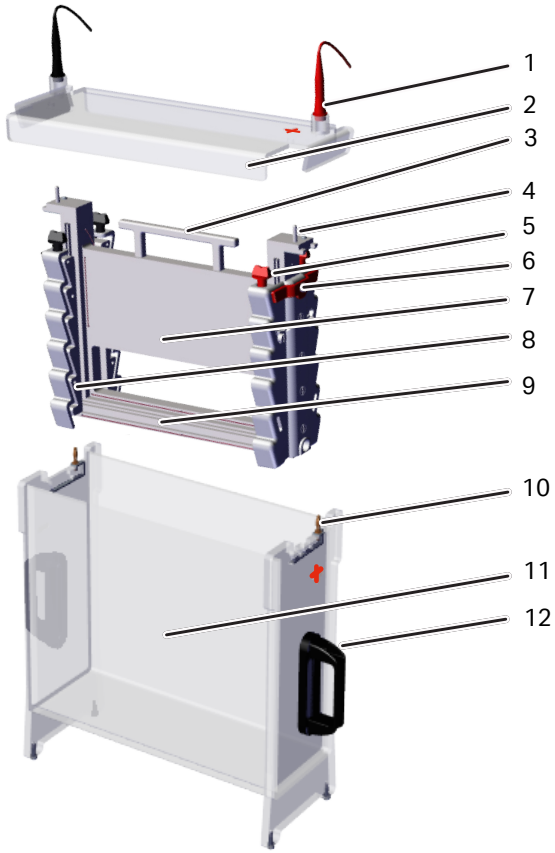
## Differential in-blot electrophoresis

The DIGE Unit LF24 instrument can be used in a DIBE (differential in-blot electrophoresis) assay workflow. The DIBE coverage analysis is a two-dimensional electrophoresis and Western blotting-based assay used to determine host cell protein (HCP) antibody coverage. HCP antibody coverage is used to determine the proportion of HCP that the HCP ELISA kit recognizes in a process specific sample.

In DIBE coverage assay, the HCP sample is first labeled with a fluorescent marker (such as, CyDye™ DIGE Fluor Cy™3 minimal dye) and then separated with 2-D electrophoresis. After electrotransfer, the proteins on the membrane are probed by HCP-specific primary antibodies and are detected with fluorescent-labeled secondary antibodies (such as, CyDye Cy5-labeled antibodies). The fluorophores on the membrane are detected in two distinct channels to create an overlay of the 2-D spot patterns into a multiplexed image. The percentage of protein spot coverage by the antibody detection is then analyzed with the Melanie software. For more information on the DIBE coverage analysis, see the *Instructions for Use, 29657336*.

## Illustration of the electrophoresis module

The illustration below shows the main parts and gives a brief description of the electrophoresis module.

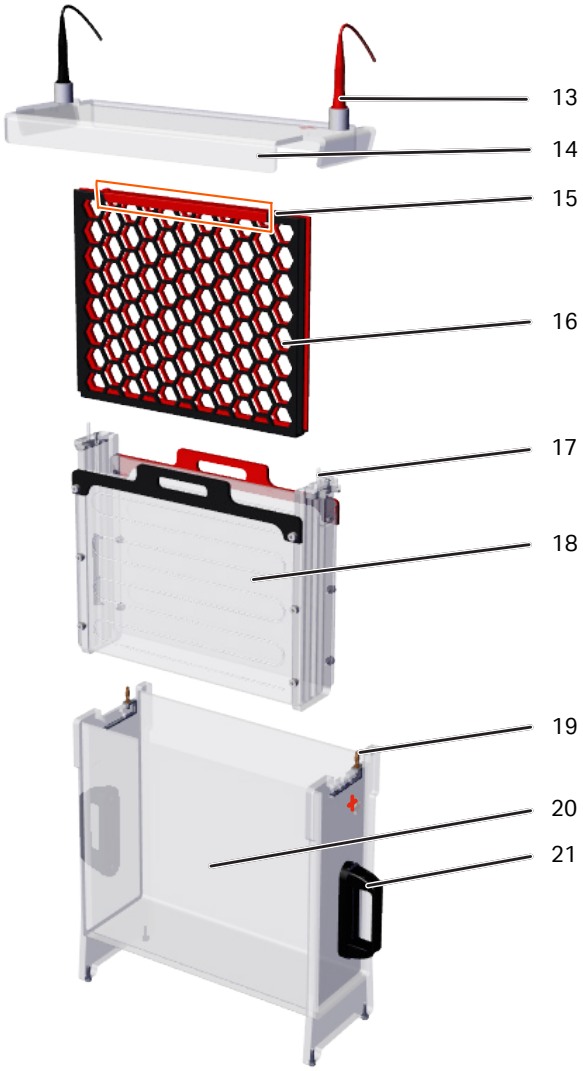



| Part | Description |
|---|---|
| 1 | Power lead (4 mm) |
| 2 | Safety lid |
| 3 | Handle on gel cassette holder |
| 4 | Placeholder pin |
| 5 | Clamp screw |
| 6 | Polarity marker |
| 7 | Gel cassette holder |

| Part | Description |
|---|---|
| 8 | Plastic clamp |
| 9 | Rubber gasket |
| 10 | Banana plug |
| 11 | Tank |
| 12 | Handle on tank |

## Illustration of the electrotransfer module

The illustration below shows the parts and gives brief descriptions of the electrotransfer module.




| Part | Description |
|---|---|
| 13 | Power lead (4 mm) |
| 14 | Safety lid |
| 15 | Hinge on the transfer cassette |

| Part | Description |
|---|---|
| 16 | Transfer cassette |
| 17 | Placeholder pin |
| 18 | Transfer cassette holder |
| 19 | Banana plug |
| 20 | Tank |
| 21 | Handle on tank |

# 4 Installation

## About this chapter

This chapter provides required information to enable users to unpack, install, move and transport the product.

## In this chapter

# 4.1 Safety precautions

## Installing and moving the product

**CAUTION**

**Move carefully.** When lifting and moving the product be careful not to drop it. This may cause injury.

**CAUTION**

**Stable bench.** Make sure that the product is placed on a stable, level bench with adequate space for ventilation.

**CAUTION**

**Turn off power.** Turn off the power switch of the electrophoresis power supply, and disconnect power leads from the electrophoresis power supply before moving the product.

# 4.2 Site preparation

## Introduction

This section describes the site planning and preparation that must be performed before the product is installed.

## In this section

# 4.2.1 Delivery, storage and unpacking

## Introduction

This section describes the requirements for receiving the delivery box and storing the product before installation.

## When you receive the delivery

- Record on the receiving documents if there is any apparent damage on the delivery box. Inform your Cytiva representative of such damage.
- Move the delivery box to a protected location indoors.

## Delivery box

The products are shipped in delivery boxes with the following dimensions and weight:

| Contents | Dimensions (mm) | Weight |
|---|---|---|
| DIGE Unit LF24 instrument | 522 × 525 × 257 (width × height × depth) | 7.1 kg (15.6 lb) |
| Transfer Unit LF24 module | 412 × 355 × 207 (width × height × depth) | 2.1 kg (4.6 lb) |

## Storage requirements

The delivery boxes must be stored at a protected place indoors. The storage place for unopened boxes must meet the following requirements:

| Parameter | Allowed range |
|---|---|
| Ambient temperature, storage | 4°C to 40°C |
| Relative humidity | 20% to 95%, non-condensing |

## Unpacking the product

Remove straps and packing material and stand the product upright before installation.

Check the content of the delivery according to the table below. Inform your Cytiva representative in case of any missing components.

| Components | Combined product | DIGE Unit LF24 | Transfer Unit LF24 |
|---|---|---|---|
| Tank | ✓ | ✓ | ✗ |
| Safety lid with power leads | ✓ | ✓ | ✗ |
| Gel cassette holder | ✓ | ✓ | ✗ |
| Dummy plate | ✓ | ✓ | ✗ |
| Transfer cassette holder | ✓ | ✗ | ✓ |
| Transfer cassette (2 ×) | ✓ | ✗ | ✓ |
| Additional hinges for the transfer cassette (2 ×) | ✓ | ✗ | ✓ |
| Sponges (4 ×) | ✓ | ✗ | ✓ |

## Transportation

Before moving the product:

- Remove the liquids and the gels or transfer cassettes from the tank
- Disconnect the power leads from the electrophoresis power supply

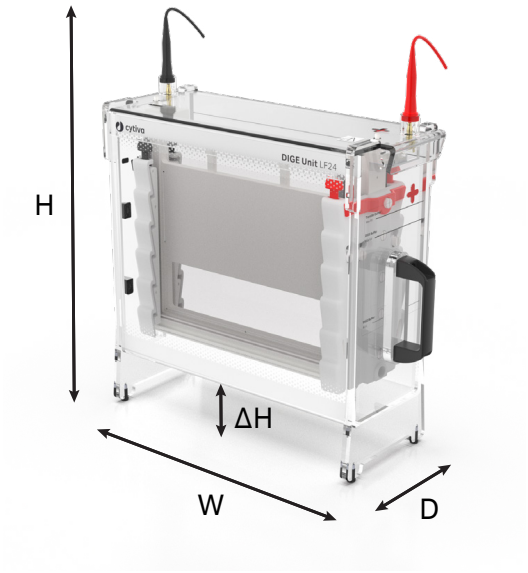
# 4.2.2 Dimensions and weight

## Introduction

This section describes the requirements for the room where the product is placed.

## DIGE Unit LF24

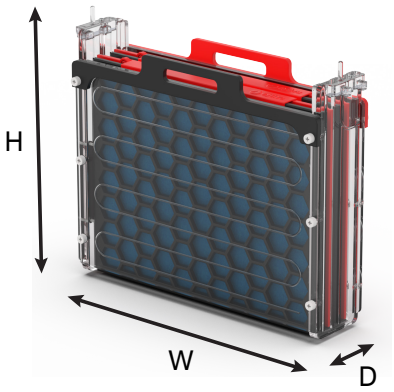
The following illustration shows the dimensions of the DIGE Unit LF24, i.e. the tank and the gel cassette holder.




| Parameter | Value |
|---|---|
| Width (W) | 43.2 cm (17 in) |
| Depth (D) | 15 cm (5.9 in) |
| Height (H) | 41.6 cm (16.4 in) |
| Height under tank (ΔH) | 6 cm (2.4 in) |
| Weight | 5.8 kg (12.8 lb) |

## Transfer Unit LF24

The following illustration shows the dimensions of the Transfer Unit LF24, i.e. the transfer cassette holder with two transfer cassettes.




| Parameter | Value |
|---|---|
| Width (W) | 33.5 cm (13.2 in) |
| Depth (D) | 9 cm (3.5 in) |
| Height (H) | 26 cm (10.2 in) |
| Weight | 1.5 kg (3.3 lb) |

## Operation weight

The following table shows the weight of the product in operation.

| Operation | Components | Weight |
|---|---|---|
| Electrophoresis | • Tank<br>• Lid<br>• Gel cassette holder<br>• Two gels<br>• Buffer (5.7 L) | 13.2 kg (29 lb) |

| Operation | Components | Weight |
|---|---|---|
| Electrotransfer | • Tank<br>• Lid<br>• Transfer cassette holder<br>• Two assembled transfer cassettes<br>• Buffer (7 L) | 11.4 kg (25.1 lb) |

## Space requirements

Prepare a clean working area on a stable laboratory bench or trolley that complies with the specifications in the following table.

| Parameter | Specification |
|---|---|
| Minimum bench area for operating the product (D x W) | 65 × 65 cm |
| Free space required around the product | At least 40 cm free space in front of the product<br>10 cm free space on all other sides |
| Load capacity | 490 N (50 kg) or higher |
| Inclination of bench surface | Horizontal ±2° |

# 4.2.3 Site environmental requirements

## Introduction

This section describes the environmental requirements and conditions for installation of the product.

**CAUTION**

Do not use the product in a dusty atmosphere or close to spraying water.

**CAUTION**

The product is designed for indoor use only.

## Environmental requirements

For details of environmental requirements for operation, storage and transport, see *Environmental requirements, on page 86*.

## Environmental conditions

The following general requirements must be fulfilled:

- The room must have exhaust ventilation.
- The product must not be exposed to sources of heat such as direct sunlight.
- The dust in the atmosphere must be kept to a minimum.

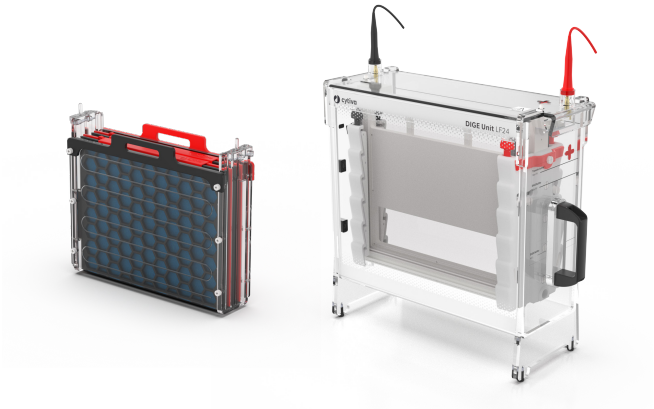
# 4.3 Installing the product

## Introduction

This section describes the procedure for installing the product.

## Place the product on the bench

Place the product on the bench. See *Space requirements, on page 31* for space requirements. Lift the tank with the handles on the side of the tank.



- For easy rinsing and draining, place the product close to the sink.
- Place the electrophoresis power supply near the product in a dry place without risk for liquid exposure.

# 5 Operation

## About this chapter

This chapter gives instructions on how to operate the product in a safe way.

## In this chapter

## 5.1 Safety precautions

**WARNING**

**Shock hazard.** The safety lid must be placed on the tank before connecting the power leads to the electrophoresis power supply. Failure to do so may result in fire or electric shock.

**WARNING**

**Shock hazard.** The electrophoresis power supply must always be disconnected and switched off when the safety lid of the tank is taken off. Failure to do so may result in fire or electric shock.

**CAUTION**

**Be careful.** Do not drop the gel cassettes when lifting and moving them. Dropping the glass plates may cause them to break and cause injury.

**NOTICE**

**Heavy.** The tank is heavy when filled with buffer. Handle the tank with care to avoid personal injury.

**NOTICE**

**Overheating.** Do not operate the product with buffer temperatures above the maximum specified technical specifications. Overheating causes irreparable damage to the product.

# 5.2 Electrophoresis module

This section gives instructions on how to operate the electrophoresis module in a safe way.

## In this section

# 5.2.1 Materials and buffers

## Required materials and equipment

The following materials and equipment are required to use the electrophoresis module. The Cytiva product codes are shown in parentheses.

- DIGE Buffer Kit (28937452)
- DIGE Gels LF24, 12.5% polyacrylamide gels, 24 cm (29706670)
- Electrophoresis power supply with a minimal rating of 800 VDC, 400 mA, such as EPS 3501 XL (18113005).

  The electrophoresis power supply must have the following safety features:

  - Functional earth leakage
  - Start current check
  - Sudden load change detection
- Equilibration tubes or tray, such as Equilibration tubes (80646779)
- Gel cassette rack (80646798)
- Gel scanner, such as an Amersham Typhoon™ scanner
- Heating block
- IPG strips, such as Immobiline™ DryStrip pH 3 to 11 NL, 24 cm, (17600377)
- Orbital shaker or rocking platform

The following material is optional:

- Power supply adapter (18112959) to connect 4 mm power leads to 2 mm power outlet of the electrophoresis power supply EPS 3501 XL

## Electrophoresis buffers

| Buffer | Concentration | Volume |
|---|---|---|
| Agarose sealing solution | Agarose sealing solution from the DIGE buffer kit (28937452) | 1 mL/gel |
| Anode buffer (outer chamber) | Anode buffer from the DIGE buffer kit (28937452)[1] | 4.5 L |
| Cathode buffer (inner chamber) | Cathode buffer from the DIGE buffer kit (28937452)[1] | 1.2 L |

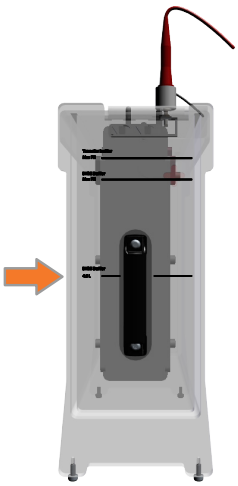
| Buffer | Concentration | Volume |
|---|---|---|
| IPG equilibration buffer[2] | 6 M Urea<br>50 mM Tris-HCl (pH 8.8)<br>30% (v/v) Glycerol<br>2% (w/v) SDS<br>0.001% (w/v) Bromophenol blue | 30 mL/strip |

[1] The DIGE buffer kit contains enough buffer for two electrophoresis runs with DIGE Unit LF24.
[2] Before use, add dithiothreitol (DTT) or iodoacetamide to the IPG equilibration buffer, see step 2 in *Preparing the gel, on page 40*. The IPG equilibration buffer can be stored at -20°C.

## 5.2.2 Preparations

### Preparing the tank

Follow the steps below to prepare the tank for an electrophoresis run.

| Step | Action |
|---|---|
| 1 | Lift out the gel cassette holder from the tank. Use the handle on the gel cassette holder to lift it. |
| 2 | Pour 125 mL concentrated anode buffer (1 bottle) into a large beaker. Rinse the bottle with distilled or deionized water, and pour it in the beaker. Fill up the beaker to 4.5 L with distilled or deionized water. Then, pour the buffer into the tank.  |
| 3 | Pour 800 mL distilled or deionized water into a graduated cylinder. Then, pour 250 mL of concentrated cathode buffer (2 bottles) into the graduated cylinder. Rinse the bottles with distilled or deionized water, and pour it into the graduated cylinder. Slowly, fill up the graduated cylinder to 1.2 L with distilled or deionized water.<br><br>***Note:***<br>*Dilute the cathode buffer slowly to avoid the creation of bubbles.* |

## Cooling options for electrophoresis

The electrophoresis run does not require cooling, except when running with the highest power setting. However, running at a lower temperature will decrease the diffusion of proteins somewhat. The buffer temperature must be kept under 35°C, see *Technical specification, on page 86*.

**NOTICE**

Do not exceed the specified temperature limits for the equipment.

Choose from the options below to cool the system during an electrophoresis run. The two cooling options can be combined.

| Cooling option | Action |
|---|---|
| Cool the buffers | Pre-cool the buffers to 4°C overnight, before starting the electrophoresis run. |
| Operate in the cold room | Perform the electrophoresis run in a cold room. |

## Preparing the gel

Prior to 2-D electrophoresis, the protein sample must be run on immobilized pH gradient (IPG) strips. For more information, see *2-D Electrophoresis, on page 18*, and *Handbook, CY14825*.

For DIGE and DIBE applications, label the protein sample with CyDye DIGE minimal dye prior to loading on the IPG strip. For more information about DIBE coverage analysis, see *Instructions for Use, 29657336*.

Follow the steps below to prepare the gel for the 2-D electrophoresis run.

| Step | Action |
|---|---|
| 1 | Equilibrate the gels to room temperature in their original packaging material. |
| | **CAUTION**<br>**Broken glass plate.** Do not use the precast gels if the glass plates are cracked. Broken glass can cause injury. |
| 2 | For each gel, heat up one aliquot of sealing solution to 95°C in a heating block. |
| 3 | Equilibrate the IPG strips in equilibration buffer: |

**Step** **Action**

**a.** Prior to use, make two IPG equilibration buffers, one containing DTT and the other containing iodoacetamide, respectively.

| Buffer | Action |
|---|---|
| DTT-containing IPG equilibration buffer | For each IPG strip, dissolve 150 mg DTT in 15 mL of IPG equilibration buffer (1.0% w/v). |
| Iodoacetamide-containing IPG equilibration buffer | For each IPG strip, dissolve 375 mg iodoacetamide in 15 mL of IPG equilibration buffer (2.5% w/v). |

**b.** Place the IPG strips in individual tubes or trays. Then, add 15 mL DTT-containing IPG equilibration buffer to each IPG strip.

**NOTICE**

Always wear gloves when working with protein samples and gels to avoid contaminations.

**c.** Incubate on an orbital shaker or rocking platform for 15 min.

***Note:***
*Be consistent with the equilibration time when equilibrating the IPG strip. The equilibration time is critical for a successful protein separation when running the second dimension of the 2-D electrophoresis run.*

**d.** Replace the DTT-containing IPG equilibration buffer with 15 mL iodoacetamide-containing IPG equilibration buffer.

**e.** Incubate on an orbital shaker or rocking platform for 15 min.

**f.** Rinse the IPG strip briefly in cathode buffer.

4 Place the gel cassette horizontally on the lab bench with the notched glass plate facing upwards.

5 Place the IPG strip on the gel:

**a.** Put the IPG strip on the protruding glass plate of the gel cassette.

***Tip:***
*It is good practice to consistently put the IPG strips with the same orientation on the gel.*

**b.** Gently push the plastic back of the IPG strip towards the upper surface of the gel with a thin ruler or a spatula.

| Step | Action |
|---|---|
| | **c.** Check that the IPG strip has contact with the gel and no air bubbles remain between the IPG strip and the gel. Repeat the previous step, if necessary. |
| 6 | Optional: Load the molecular weight marker to a sample application piece and place it adjacent to the IPG strip.<br><br>***Tip:***<br>*Running a molecular weight marker together with the sample will help with the DIBE coverage analysis.* |
| 7 | Carefully pipette 1 mL hot agarose sealing solution across the whole length of the IPG strip.<br><br>**NOTICE**<br>Do not create bubbles and do not let the agarose sealing solution cool down before use. |
| 8 | Let the agarose sealing solution set completely. |

Immediately start the 2-D electrophoresis run.

## 5.2.3 Running the gel

***Tip:*** *Use cooling during the 2-D electrophoresis run for heat-sensitive samples. See Cooling options for electrophoresis, on page 40.*

Follow the steps below to run the samples in the second dimension of the 2-D electrophoresis run.

| Step | Action |
|---|---|
| 1 | One by one, insert the gel cassettes into the gel cassette holder: |

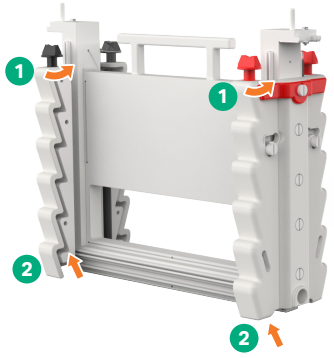
**a.** Place the gel cassette holder on a flat surface, such as a lab bench.

**b.** Loosen the black and red clamp screws (**1**).

**NOTICE**

Do not screw out the black and red clamp screws from the gel cassette holder. This increases the risk of losing the clamp screws.

**c.** Push up the plastic clamps (**2**).




**NOTICE**

Do not remove the polarity markers from the sides of the gel cassette holder.

***Tip:***

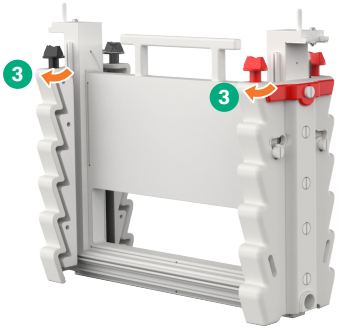
*If the plastic clamps do not stay in the open position, tighten the screws that hold the clamp. For instructions, see Section 6.5 Tighten the screws of the gel cassette holder, on page 75.*

| Step | Action |
|---|---|

**d.** Insert the gel cassette into the gel cassette holder with the notched glass plate facing inwards. Push the gel cassette down until it touches the lab bench surface.

> **NOTICE**
>
> Always wear gloves when working with protein samples and gels to avoid contaminations.

**e.** Tighten the red and black clamp screws (**3**) to close the plastic clamp.

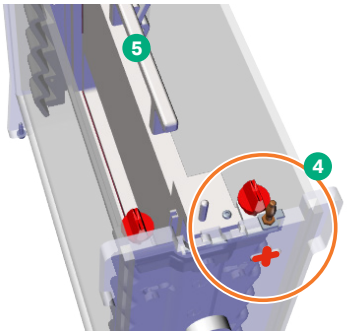



> **CAUTION**
>
> **Cutting injury.** Do not overtighten the clamps after inserting the gels into the gel holder. This could break the glass plates and cause injury.

**f.** Check that the gel cassette is flush with the bottom of the gel cassette holder.

**g.** Insert the second gel cassette or the dummy plate to the other side of the gel cassette holder, by following steps 1a through 1f.

| Step | Action |
|---|---|
| 2 | Align the polarity of the gel cassette holder (indicated by the red and black clamp screws) with the polarity symbols on the tank (**4**). Then, lift the gel cassette holder by its handle (**5**) into the tank. |




**CAUTION**

**Heavy.** Lift the gel cassette holder by its handle. Use both hands to lift the gel cassette holder. The gel cassette holder with gels is heavy. Dropping the gel cassette holder can cause injury.

**NOTICE**

**Banana plug damage.** Handle the gel cassette holder carefully to prevent damage to the banana plug on the tank.

3 Make sure that the inner buffer chamber is tight:

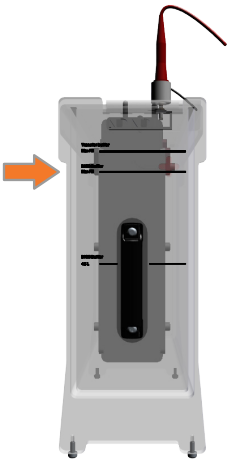
**a.** Check that no anode buffer leaks into the inner buffer chamber.

**b.** If buffer leaks into the inner buffer chamber, re-insert the gel cassettes into the gel cassette holder to make sure that the inner buffer chamber is tight, see step 1.

4 Make sure that the level of anode buffer is correct:

| Step | Action |
|---|---|

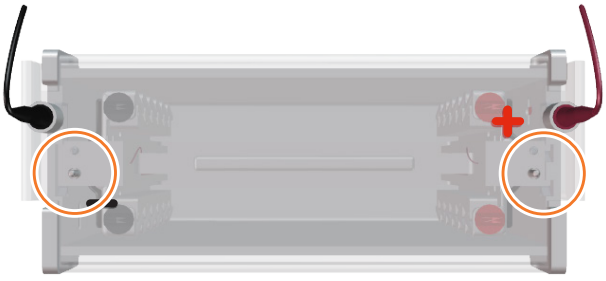
**a.** Check that the buffer level in the outer buffer chamber does not exceed the **DIGE Buffer Max Fill** line.



**b.** If required, adjust the volume of anode buffer in the outer chamber so that the buffer level is at the **DIGE Buffer Max Fill** line. If the buffer level is above the fill line, remove any excess buffer. If the buffer level is below the fill line, add distilled or deionized water up to the **DIGE Buffer Max Fill** line.

5 Fill up the inner chamber with 1.2 L cathode buffer. Make sure that the buffer level is approximately 0.5 cm above the notched gel plates. If required, add distilled water into the inner chamber to reach this buffer level.

6 Place the safety lid on the tank. Make sure that the place holder pins keep the safety lid in place, as indicated in the image below.



| Step | Action |
|---|---|

> **WARNING**
>
> **Electric shock.** Do not hold the lid by the power connectors or power leads. Damaged power connectors or power leads can cause electric shock.

7 If necessary, plug in the power supply adapter (see image below) to the power lead.

> **WARNING**
>
> **Shock hazard:** When using power supply adapters, make sure that they have the appropriate rating. Electric shock can occur when using a power supply adapter with the incorrect rating.

8 Plug in the power leads to the electrophoresis power supply. Make sure that the polarity of the power leads matches the polarity of the electrophoresis power supply.

| Step | Action |
|---|---|
| 9 | Set the electrophoresis power supply to the run conditions in the table below. |

| Program | Run phase | Voltage (V) | Current (mA) | Power (W/2 gels) | Time (h) | Cooling |
|---|---|---|---|---|---|---|
| **Day run** | 1[1] | 800 | 400 | 2 | 1 | N/A |
| | 2 | 800 | 400 | 34 | 4 to 5 | N/A |
| **Fast day run** | 1[1] | 800 | 400 | 2 | 1 | Yes[2] |
| | 2 | 800 | 400 | 100 | 1.5 to 2 | Yes[2] |
| **Overnight run** | 1[1] | 800 | 400 | 2 | 8 | N/A |
| | 2 | 80 | 400 | 2 | 9 to 11 | N/A |

[1] Sample entry phase

[2] See *Cooling options for electrophoresis, on page 40*.

**WARNING**

**Shock hazard.** Do not set the electrophoresis power supply to an electrical input above 800 VDC, 400 mA and 100 W during any part of the run. Exceeding the electrical input limits can result in fire or electric shock.

***Note:***

*Depending on the size of the proteins in the sample, the running conditions may need to be adjusted.*

10 Press the **Run** button on the electrophoresis power supply.

***Note:***

*Continue the electrophoresis run until the bromophenol blue-front reaches the end of the gel. The bromophenol blue front can be run off the gel if desired.*

11 Press the **Stop** button on the electrophoresis power supply, and disconnect the power leads from the electrophoresis power supply. Then, turn off the electrophoresis power supply.

## 5.2.4 After the electrophoresis run

Follow the steps below after an electrophoresis run.

| Step | Action |
|---|---|
| 1 | Remove the gels from the tank: |

**a.** Lift off the safety lid.

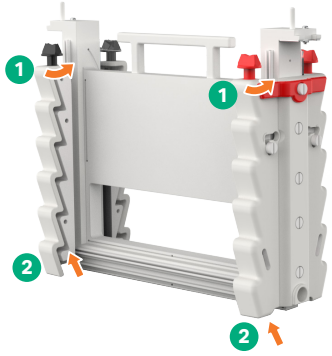
> **WARNING**
>
> **Electric shock.** Do not hold the lid by the power connectors or power leads. Damaged power connectors or power leads can cause electric shock.

**b.** Lift out the gel cassette holder by its handle from the tank.

> **CAUTION**
>
> **Heavy.** Lift the gel cassette holder by its handle. Use both hands to lift the gel cassette holder. The gel cassette holder with gels and buffer is heavy. Dropping the gel cassette holder can cause injury.

**c.** Tilt the gel cassette holder over the sink, and pour out the cathode buffer.

**d.** Loosen the black and red clamp screws (**1**). Then, push up the plastic clamps (**2**).




***Tip:***

*If the plastic clamps do not stay in the open position, tighten the screws that hold the clamp. For instructions, see* *Section 6.5 Tighten the screws of the gel cassette holder, on page 75**.*

**e.** Lift out the gel cassettes from the gel cassette holder.

| Step | Action |
|---|---|
| 2 | Optional: Store the gel cassettes at 4°C, protected from light, in a closed container (e.g. the original packaging material). Add some distilled water and seal the container to prevent the gels from drying out.<br><br>***Note:***<br><br>*To minimize spot diffusion, scan the gels as soon as possible after the run. Storage longer than 2 days will cause diffusion of proteins in the gel.* |
| 3 | If the gel cassettes were stored at 4°C after the electrophoresis run, equilibrate the gel cassettes to room temperature before scanning. |
| 4 | Scan the gel cassettes. |

After running the gel, the proteins can be transferred to a membrane with the electrotransfer module, as described in *Section 5.3 Electrotransfer module, on page 51*.

Clean the electrophoresis module as described in *Section 6.4 Cleaning after a run, on page 74*.

# 5.3 Electrotransfer module

This section gives instructions on how to operate the electrotransfer module in a safe way.

## In this section

# 5.3.1 Materials and buffers

## Required materials and equipment

The following materials and equipment are required to use the electrotransfer module. The Cytiva product codes are shown in parentheses.

- Blotting paper, such as TE76 (80621129)
- Electrophoresis power supply with a minimal rating of 800 VDC, 400 mA, such as EPS 3501 XL (18113005).

  The electrophoresis power supply must have the following safety features:

  - Functional earth leakage
  - Start current check
  - Sudden load change detection
- Plastic wedge tool, such as Wonder wedge (SE1514)
- PVDF membrane

  ***Note:*** *Nitrocellulose membranes can be used with the electrotransfer module. Changing the membrane type can require optimization of the transfer buffer and running conditions.*
- Roller
- Transfer buffer

The following material can be used with the electrotransfer module:

- Power supply adapter (18112959) to connect 4 mm power leads to 2 mm power outlet of the electrophoresis power supply EPS 3501 XL

## Electrotransfer buffer

| Buffer | Composition | Volume |
|---|---|---|
| Transfer buffer | 25 mM Tris<br>192 mM Glycine<br>20% (v/v) Methanol[1] | 7.0 L [2] |

[1] Ethanol can be used in the transfer buffer instead of methanol. Note that ethanol has a lower flashpoint than methanol.
[2] For a electrotransfer run with one transfer cassette, more buffer can be required.

## 5.3.2 Preparations

### Cooling options for electrotransfer

**CAUTION**

**Fire hazard**. Do not exceed the maximum buffer temperature. Heating flammable liquids above their flash point can cause fire.

The electrotransfer run does not require cooling, except when using ethanol in the transfer buffer.

Choose from the cooling options below to prevent overheating during an electrotransfer run. The two cooling options can be combined.

| Cooling option | Action |
|---|---|
| Cool the buffers | Pre-cool the transfer buffer to 4°C overnight, before starting the electrotransfer run. |
| Operate in the cold room | Perform the electrotransfer run in a cold room. |

### Preparing materials for the transfer stack

**NOTICE**

Always use gloves and flat tweezers when handling the membrane, to minimize the risk of contamination.

Follow the instructions below to prepare the materials for one transfer stack.

| Step | Action |
|---|---|
| 1 | Cut the membrane to the same size as the TE76 blotting paper. |

| Step | Action |
|---|---|
| 2 | Place the membrane in a container with 100% methanol. Incubate the membrane for 5 minutes on an orbital shaker or rocking platform set to gentle agitation. |

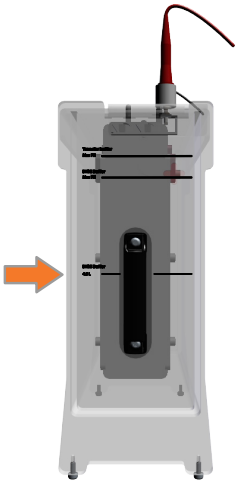
***Tip:***
*Ethanol can be used in the transfer buffer instead of methanol. In such case, replace methanol with 100% ethanol in this step.*

| | |
|---|---|
| 3 | Pour out the methanol. |
| 4 | Pour transfer buffer in the container with the membrane. |
| 5 | Incubate the membrane in transfer buffer for 15 minutes on an orbital shaker or rocking platform set to gentle agitation. |
| 6 | Soak two pieces of TE76 blotting paper and two sponges in transfer buffer. |

## Preparing the tank

Follow the steps below to prepare the tank for the electrotransfer run.

| Step | Action |
|---|---|
| 1 | Fill up the tank with transfer buffer to the **DIGE Buffer 4.5 L** fill line. |



## Assembling the transfer stack

**NOTICE**

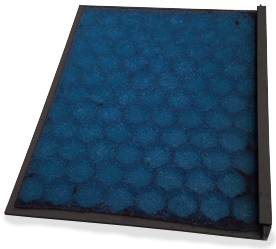
Always use gloves and flat tweezers when handling the membrane, to minimize the risk of contamination.

Follow the steps below to prepare the electrotransfer stack.

| Step | Action |
|---|---|
| 1 | If the gels were stored at 4°C, equilibrate the gels to room temperature. |
| 2 | Open the hinge of the transfer cassette. Separate the two transfer cassette halves. |
| 3 | Assemble the first half of the transfer stack:<br>**a.** Place the black half of the transfer cassette on the lab bench next to the sink. |

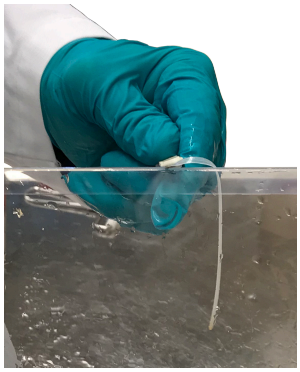
| Step | Action |
|---|---|

**b.** Place one pre-soaked sponge on the black half of the transfer cassette. Tuck in the edge of the sponge into the groove of the black half of the transfer cassette.



4 Take out the gel from the gel cassette:

**a.** Remove the silicone sealing on both short sides of the gel cassette with a small knife.



**b.** Place the gel cassette on the lab bench. Make sure that the notched glass plate is facing up.

| Step | Action |
|---|---|

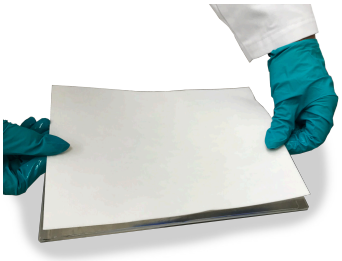
**c.** Gently pry open the gel cassette with a plastic wedge tool. Lift off the notched glass plate.

*Result:*
The gel rests on the bottom glass plate.

**d.** Remove plastic spacers from the glass plate.

**e.** Remove the IPG strip with the plastic wedge tool. Discard the IPG strip according to local regulations.

**f.** Place one pre-soaked blotting paper on top of the gel. Align the edges of the blotting paper with the edges of the gel.



*Result:*
A temporary stack is created of the glass plate, the gel, and one blotting paper.

**g.** Use a wet roller to remove any bubbles, and increase the contact between the blotting paper and the gel.

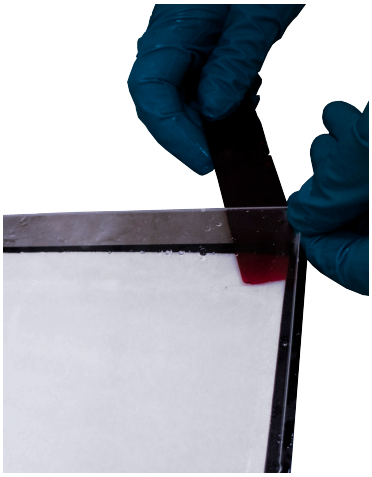
| Step | Action |
|---|---|

**h.** Lift up the temporary stack of the glass plate, the gel, and the blotting paper, then turn it over. Place it on the sponge with the blotting paper facing down.

| Placement | Transfer stack content at this step |
|---|---|
| Top | Glass plate |
| Layer 3 | Gel |
| Layer 2 | Blotting paper |
| Layer 1 | Sponge |
| Bottom | Black half of the transfer cassette |

*Result:*
The transfer stack now contains: the sponge, one blotting paper, the gel, and the glass plate.

**i.** Gently, detach the gel from the glass plate with a plastic wedge tool, and lift off the glass plate.



*Result:*
The transfer stack now contains: the sponge, one blotting paper, and the gel.

**j.** Center the gel on the transfer cassette. If necessary, use a wet roller to even out the gel.

5 Assemble the second half of the transfer stack:

**a.** Place the equilibrated membrane on top of the gel. Make sure that the membrane covers the entire gel.

| Step | Action |
|---|---|

**b.** Tuck in the edge of the membrane into the groove of the black half of the transfer cassette.

*Result:*
The transfer stack now contains: the sponge, one blotting paper, the gel, and the membrane.

**c.** Use a wet roller to remove any bubbles between the gel and the membrane.

**d.** Place one pre-soaked blotting paper on the membrane. Remove any bubbles between the layers with a wet roller.

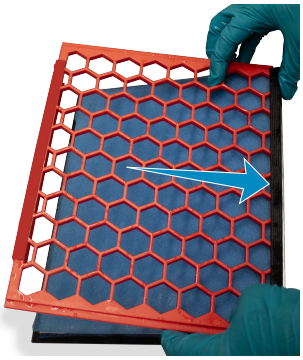
*Result:*
The transfer stack now contains: the sponge, the first blotting paper, the gel, the membrane, and the second blotting paper.

| Step | Action |
|---|---|

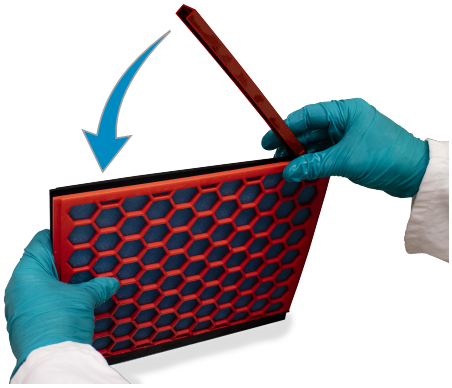
**e.** Place one pre-soaked sponge on top of the blotting paper. Tuck in the edge of the sponge into the groove of the black half of the transfer cassette.

*Result:*
The transfer stack now contains: the first sponge, the first blotting paper, the gel, the membrane, the second blotting paper, and the second sponge.

**f.** Slide the red half of the transfer cassette into the groove of the black half.



| Step | Action |
|---|---|
| 6 | Press the black half and the red half of the transfer cassette together. Close the hinge over both halves to lock the transfer cassette. |



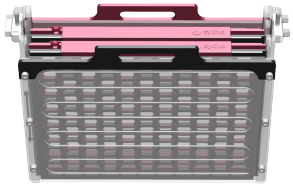
*Result:*
The transfer stack is ready.

To prevent the transfer cassette from drying out, immediately continue with *Place the transfer cassettes in the transfer cassette holder, on page 61*.

## Place the transfer cassettes in the transfer cassette holder

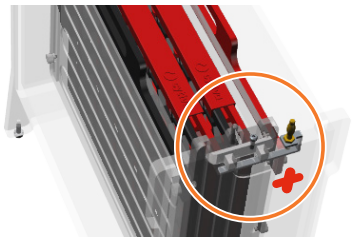
Follow the steps below to place the transfer cassettes in the transfer cassette holder.

| Step | Action |
|---|---|
| 1 | Insert assembled transfer cassettes into the transfer cassette holder. Make sure that the polarity of the transfer cassette matches the polarity of the transfer cassette holder. |



***Tip:***
*Write down the orientation and order in which the transfer stacks were loaded in the transfer cassette holder.*

| Step | Action |
|---|---|
| 2 | Align the polarity symbols on the transfer cassette holder with the polarity symbols on the tank. Then, lift the transfer cassette holder by its handle into the tank. |



**NOTICE**

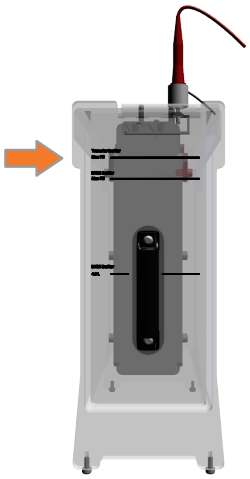
**Banana plug damage.** Handle the transfer cassette holder carefully to prevent damage to the banana plug on the tank.

| | |
|---|---|
| 3 | Optional: Prepare a second transfer stack by following step 2 to 6 in *Assembling the transfer stack, on page 55*. |

| Step | Action |
|---|---|
| 4 | Fill up the tank with transfer buffer to the **Transfer Buffer Max Fill** line. |

**WARNING**

**Shock hazard.** Do not exceed the **Transfer Buffer Max Fill** line when filling the tank with transfer buffer. Buffer overflow can cause an electric shock when touching the tank during a run.



Immediately continue with *Section 5.3.3 Running the electrotransfer, on page 64*.

## 5.3.3 Running the electrotransfer

Follow the steps below to run the electrotransfer.

| Step | Action |
|---|---|
| 1 | Place the safety lid on the tank. Make sure that the place holder pins keep the safety lid in place, as indicated in the image below. |

**WARNING**

**Electric shock.** Do not hold the lid by the power connectors or power leads. Damaged power connectors or power leads can cause electric shock.

| | |
|---|---|
| 2 | If necessary, plug in the power supply adapter (see image below) to the power lead. |

**WARNING**

**Shock hazard:** When using power supply adapters, make sure that they have the appropriate rating. Electric shock can occur when using a power supply adapter with the incorrect rating.

| Step | Action |
|---|---|
| 3 | Plug in the power leads to the electrophoresis power supply. Make sure that the polarity of the power leads matches the polarity of the electrophoresis power supply. |

| Step | Action |
|---|---|
| 4 | Set up the program on the electrophoresis power supply with the run conditions in the table below. |

| Current (mA) | Time (min) |
|---|---|
| 400[1] | 90[2] |

[1] The maximum electrical input for the product is 800 V, 400 mA, and 100 W.
[2] Depending on the molecular weight of the proteins in the sample, the running time can require optimization.

**WARNING**

**Shock hazard.** Do not set the electrophoresis power supply to an electrical input above 800 VDC, 400 mA and 100 W during any part of the run. Exceeding the electrical input limits can result in fire or electric shock.

**CAUTION**

**Fire hazard.** Make sure that the buffer temperature does not exceed 30°C when using ethanol in the transfer buffer. Heating flammable liquids above their flash point can cause fire.

***Note:***

*Electrotransfer generates heat that can damage heat-sensitive proteins in the sample.*

| Step | Action |
|---|---|
| 5 | Press the **Run** button on the electrophoresis power supply. |
| 6 | At the end of the electrotransfer run, turn off the electrophoresis power supply. Disconnect the power leads from the electrophoresis power supply. |

Remove the membrane from the transfer cassette as described in *Section 5.3.4 After the electrotransfer run, on page 67*.

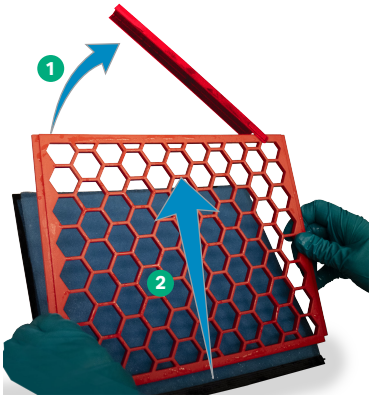
## 5.3.4 After the electrotransfer run

Follow the steps below after the electrotransfer run.

| Step | Action |
|---|---|
| 1 | Lift off the safety lid. |

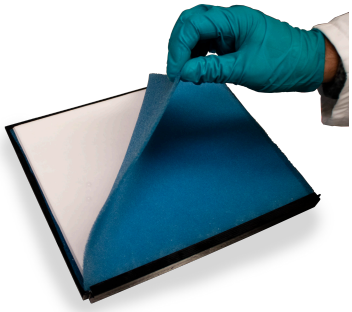
> **WARNING**
>
> **Electric shock.** Do not hold the lid by the power connectors or power leads. Damaged power connectors or power leads can cause electric shock.

| | |
|---|---|
| 2 | Remove the transfer cassette holder by its handles from the tank. Hold the transfer cassette holder over the sink, and drain out the buffer. |
| 3 | Remove the transfer cassettes from the transfer cassette holder. |
| 4 | Remove the membrane from the transfer stack:<br>**a.** Place the transfer cassette on the lab bench, with the black side down.<br>**b.** Open the hinge of the transfer cassette (**1**), then remove the red half of the transfer cassette (**2**). |



| Step | Action |
|---|---|

**c.** Remove the sponge.



**d.** Carefully, remove the blotting paper.

**e.** Carefully, lift off the membrane with tweezers. See step 5 for membrane processing.

**f.** If desired, the post-transfer gel can be scanned to give a qualitative estimate of the electrotransfer efficiency.

**g.** Discard the gel, and the blotting paper according to local regulations.

| Step | Action |
|---|---|
| 5 | Process the membrane: |

| Detection method | Action |
|---|---|
| Protein detection, such as DIBE coverage analysis | a. Keep the membrane in transfer buffer.<br>b. Follow the instructions from the manufacturer of the protein detection method. |
| Fluorescence | a. Air dry the membrane.<br>b. Scan the membrane for fluorescence. |

| | |
|---|---|
| 6 | For long term storage, air dry the membrane in a box protected from light. Store the dried membrane protected from light. |

Clean the electrotransfer module as described in *Section 6.4 Cleaning after a run, on page 74*.

# 6 Maintenance

## About this chapter

This chapter provides information about maintenance that must be performed by users of the product.

## In this chapter

## 6.1 Safety precautions

**WARNING**

**Electrical shock hazard.** Always disconnect power to the product before performing any maintenance task.

**WARNING**

**Decontaminate before maintenance.** To avoid personnel being exposed to potentially hazardous substances, make sure that the product is properly decontaminated and sanitized before maintenance or service.

**NOTICE**

**Banana plug damage.** Handle the tank carefully to prevent damage to the banana plug on the tank.

**NOTICE**

**Overheating.** Do not operate the product with buffer temperatures above the maximum specified technical specifications. Overheating causes irreparable damage to the product.

# 6.2 Cleaning before planned service

## Cleaning before planned return

Complete the checklist in the *Health and Safety Declaration Form for Product Return or Servicing*, when the product is going to be returned.

## Health and safety declaration forms

Health and safety declaration forms are available for copying or printing in the *Reference information* chapter of this manual, or on digital media supplied with the user documentation.

## 6.3 User maintenance schedule

Verify the following aspects before each run. Use the corrective action if required.

| Verify | Corrective action |
|---|---|
| All parts are clean. | Clean the product. |
| All parts are intact and free of cracks and other damage. | Replace the damaged part or the entire product. |
| The power leads are intact and free of cracks and other damage. | Replace the safety lid. |
| The hinge of the transfer cassette is free of cracks and other damage. | Replace the hinge, see *Section 6.6 Replace the hinge of the transfer cassette, on page 76*. |

## 6.4 Cleaning after a run

Follow the steps below to clean the tank and all other components used during the run.

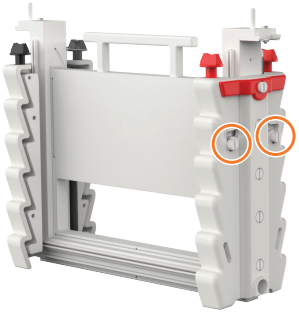
| Step | Action |
|---|---|
| 1 | Tilt the tank over the sink along the short side of the tank, and pour out the buffer. |
| | **CAUTION**<br>**Heavy.** Use the handles on the side of the tank to lift the tank with both hands. The tank is heavy when filled with buffer. Dropping the tank can cause injury. |
| | **NOTICE**<br>**Banana plug damage.** Handle the tank carefully to prevent damage to the banana plug on the tank. |
| 2 | Rinse all the used components with distilled or deionized water. |
| 3 | Periodically, the components can be cleaned with a dilute solution of a mild detergent. |

# 6.5 Tighten the screws of the gel cassette holder

Follow the steps below to tighten the screws on the gel cassette holder.

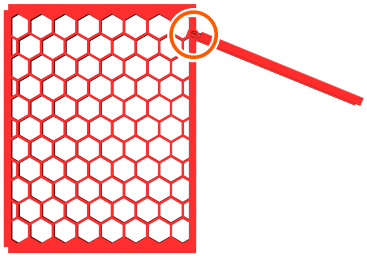
**NOTICE**

Do not remove the polarity markers from the sides of the gel cassette holder.

| Step | Action |
|---|---|
| 1 | If the plastic clamp does not stay in the open position, tighten the white screws connected to the plastic clamps.  |

# 6.6 Replace the hinge of the transfer cassette

Follow the steps below to replace the hinge of the transfer cassette.

| Step | Action |
|---|---|
| 1 | Remove the broken hinge, or any remaining part of the hinge, from the red half of the transfer cassette. |
| 2 | Clip the replacement hinge into the circular opening of red half of the transfer cassette.  |

# 7 Troubleshooting

## About this chapter

This chapter provides information to assist users to identify and correct problems that can occur when operating the product.

If the suggested actions in this guide do not solve the problem, or if the problem is not covered by this guide, contact your Cytiva representative for advice.

## In this chapter

# 7.1 Electrophoresis troubleshooting guide

| Error description | Cause | Corrective action |
|---|---|---|
| Dye front is irregular. | Contaminants in the first dimension. | Contaminants in the sample can cause distortions or swollen regions in the IPG strip after IEF. Modify sample preparation to limit these contaminants. For more information, see *Handbook, CY14825*. |
| | The top surface of the gel damaged during application of the IPG strip. | Take care during application of the IPG strip to the gel, so that neither the IPG nor the gel is damaged. |
| Low voltage, wattage, and current during the 2-D electrophoresis run. | Incorrect buffer. | Only use the DIGE buffer kit. |
| | Old buffer. | Only use the DIGE buffer kit. Do not reuse buffers. |
| No current at start of the 2-D electrophoresis run. | Insufficient volume of buffer in the inner chamber. | Add buffer to the inner chamber. Make sure that the buffer is in contact with the upper electrode and the buffer level in the inner chamber reaches the top of the gel plates. |
| Second-dimension separation proceeds slowly with high current. | Inner buffer chamber leaks. | • Tighten the red and black clamp screws fully to seal the inner buffer chamber.<br>• After fitting the gel cassettes or the dummy plate in the gel cassette holder, check that the inner buffer chamber does not leak. |

## 7.2 Stained gel troubleshooting guide

| Error description | Cause | Corrective action |
|---|---|---|
| Distortion in the second dimension pattern. | Contaminants in the first dimension. | Contaminants in the sample can cause distortions or swollen regions in the IPG strip after IEF. Modify sample preparation to limit these contaminants. For more information, see *Handbook, CY14825*. |
| High background after silver staining. | Poor quality reagents. | • Use reagents of the highest purity, preferably electrophoresis grade.<br>• Use deionized, double-distilled water.<br>• Use CyDye DIGE minimal stain for DIGE/DIBE experiments. |
| Horizontal stripes across the gel. | Impurities in IPG equilibration buffer. | Prepare fresh IPG equilibration buffer. See *Electrophoresis buffers, on page 37*. |
| No distinct protein spots are visible in the gel. | Insufficient sample amount. | Increase the amount of protein used. |
| | Poor CyDye labeling of the sample. | Make sure that the pH of the sample is between 8.0 to 9.0 before labeling the sample. Use 400 nmol CyDye per 50 µg protein. |
| Poor representation of higher molecular weight proteins. | Incorrectly prepared IPG equilibration buffer. | Prepare IPG equilibration buffer according to *Electrophoresis buffers, on page 37*. Add DTT or iodoacetamide to the IPG equilibration buffer before use, according to see step 2 in *Preparing the gel, on page 40*. |
| | Poor transfer of protein from IPG strip to second-dimension gel. | • Run the second-dimension electrophoresis step with a lower power or current setting during the sample entry phase (phase 1 in *Section 5.2.3 Running the gel, on page 43*).<br>• Prolong the sample entry phase. |
| Protein spots are diffuse or broader than usual. | Gel surface damaged by IPG strip. | Make sure the IPG strip rests on the gel surface without damaging the gel surface. |
| | Incorrect equilibration time of IPG strips. | Check the equilibration time of IPG strips. Too long IPG equilibration can lead to diffusion, and too short IPG equilibration can lead to incomplete equilibration. |
| | Poor quality reagents. | Use only analytical grade reagents. |

| Error description | Cause | Corrective action |
|---|---|---|
| | Problems with the first-dimension separation. | Refer to the troubleshooting guides for the IPGphor system, or *2-D Electrophoresis Principles and Methods Handbook (CY14825)*. |
| Protein spots are poorly resolved. | Diffusion of proteins. | Begin electrophoresis as soon as the IPG strips are equilibrated, to prevent diffusion of low molecular-weight proteins. |
| | Problems with the first-dimension separation. | Refer to troubleshooting guides for IPGphor or Multiphor™ units, or *2-D Electrophoresis Principles and Methods Handbook (CY14825)*. |
| | Too fast second-dimension separation. | Follow the recommended running conditions, see *Section 5.2.3 Running the gel, on page 43*. |
| | Too high temperature. | • Pre-chill the buffer and carry out the electrophoresis run in a cold room.<br>• Alternatively, reduce the power. |
| Spots are vertically doubled, or "twinned". | IPG strip is not placed properly. | Make sure that the plastic back of the IPG strip is placed against the back glass plate of the gel cassette. |
| Spots skewed or distorted. | Gels run too fast, causing an uneven migration. | • Run at a lower power setting.<br>• Use a two-step program: Start at a low power setting until the proteins enter the gel, then increase the power for the remainder of the run.<br>• Follow the recommended running conditions, see *Section 5.2.3 Running the gel, on page 43*. |
| Vertical gap in the 2-D pattern. | Bubbles between IPG strip and top surface of second-dimension gel. | Make sure that no bubbles are trapped between the IPG strip and the top surface of the second-dimension gel. |
| Vertical streaking. | Incorrectly prepared equilibration buffer. | Prepare equilibration buffers according to *Electrophoresis buffers, on page 37*. Add DTT or iodoacetamide to the IPG equilibration buffer before use, according to see step 2 in *Preparing the gel, on page 40*. |
| | Insufficient equilibration of IPG strips. | Equilibrate the IPG strip with each IPG equilibration buffer for 15 minutes each. See step 2 in *Preparing the gel, on page 40*. |
| | IPG strip not properly placed on gel surface. | Make sure IPG strip uniformly contacts the gel surface along its entire length. Make sure to not cause any damage to the surface of the gel. |

| Error description | Cause | Corrective action |
|---|---|---|
| | Poor transfer of proteins from IPG strip to second-dimension gel. | • Run the second-dimension electrophoresis step with a lower power or current setting during the sample entry phase (phase 1 in *Section 5.2.3 Running the gel, on page 43*).<br>• Prolong the sample entry phase. |
| | The IPG strip was not equilibrated with iodoacetamide in a second IPG equilibration step. | Equilibrate the IPG strip in two steps. In the first step with DTT, and the second step with iodoacetamide, see *Preparing the gel, on page 40*. |

## 7.3 Electrotransfer troubleshooting guide

| Error description | Cause | Corrective action |
|---|---|---|
| Diffuse spots. | Assembly of electro-transfer stack. | Make sure that the gel is held firmly against the membrane and does not move once contact is made. |
| Inefficient binding to membrane. | Membrane parame-ters. | Wear gloves when handling membranes. |
| | | Store membranes in a clean, dry atmosphere away from noxious fumes. Keep membranes in the original package material. Protect the membrane from exposure to sunlight. Store at 18 to 25, and at 40% to 60% relative humidity. |
| | | Check if the protein amount exceeds the binding capacity of the membrane by using two membranes instead of one in a transfer stack. If proteins pass through the first membrane, use a membrane with a smaller pore size (0.10 to 0.20 µm), or use a different membrane type. |
| | Transfer buffer. | • Verify that the optimal amount of methanol is used in the transfer buffer.<br>• Prepare the transfer buffer without SDS. |
| Incomplete or incon-sistent transfer pattern. | Areas of the membrane have dried out before the electro-transfer run. | Avoid drying of the membrane when preparing the transfer stack. |
| | Bubbles trapped in the transfer stack. | Remove all trapped air pockets in the transfer stack assembly. For instructions, see *Assembling the transfer stack, on page 55*. |
| | Damaged electrode. | Check electrode continuity. During the transfer, a continuous stream of gas is released along the entire length of the electrodes. If bubbles do not form along the entire length of the electrode, replace the transfer cassette holder. |
| Low voltage, wattage, and current during electrotransfer run. | The transfer buffer is old. | Use a freshly prepared buffer. See *Electrotransfer buffer, on page 52*. |

| Error description | Cause | Corrective action |
|---|---|---|
| No protein spots on membrane after the electrotransfer. | Buffer quality. | Do not add acid or base to the buffer to adjust the pH. This causes conductivity issues, which can disrupt the electrotransfer run and severely damage the product. Use only fresh transfer buffer, see *Electrotransfer buffer, on page 52*. |
| | Incomplete electro-transfer. | Increase the electrotransfer run time. |
| | Insufficient contact between the gel and the membrane.<br>The transfer cassette is not properly assem-bled or closed. | • After the electrotransfer run, stain the gel to determine the electrotransfer efficiency. Alternatively, use a pre-stained molecular weight marker to monitor the efficiency of the electrotransfer.<br>• After the electrotransfer run, stain a strip of membrane with a total protein dye, such as Ponceau S staining, or alternatively use a reversible stain on the whole membrane.<br>• Replace the sponges when they have become too thin to keep the transfer stack in place in the closed transfer cassette. Alternatively, use thicker blotting paper or more layers of blotting paper in the transfer stack.<br>• Make sure that the membrane is on the anode (+) side of the stack and the gel is on the cathode (-) side of the stack. See *Assembling the transfer stack, on page 55*. |
| | Proteins fixed to gel. | Do not use staining dyes or fixing agents on the gel before the electrotransfer. |
| | Wrong orientation of the transfer stack in the tank. | • Check that the power leads are correctly inserted into the electrophoresis power supply according to their polarity.<br>• Check that the membrane is on the anode (+) side of the gel in the transfer stack.<br>• Check that the transfer cassette holder is placed correctly in the tank. |

| Error description | Cause | Corrective action |
|---|---|---|
| Poor transfer of small proteins. | Insufficient protein retention to the membrane. | • Optimize the electrotransfer run time with relevant molecular weight markers.<br>• Use a PVDF membrane instead of a nitrocellulose membrane. PVDF membranes have a higher protein binding capacity than nitrocellulose membranes.<br>• Use a membrane with smaller pore size.<br>• Use two membranes, one on top of the other, to minimize the risk that proteins pass through the membrane without binding. |
| | SDS in the transfer buffer interferes with protein binding to the membrane. | Prepare the transfer buffer without SDS. |
| Poor transfer of large proteins. | Incomplete electrotransfer. | Prolong the run time. |
| | High molecular weight proteins stay in the gel. | • Add 0.1% SDS to the transfer buffer. Note that SDS in the transfer buffer will reduce the efficiency of proteins binding to nitrocellulose membrane.<br>• Reduce the methanol concentration to 10% or less.<br>***Note:***<br>*SDS in transfer buffer can inhibit protein binding to the membrane.* |
| Protein spots smeared across the membrane. | Excessive heat generated during the electrotransfer. | • Use a freshly prepared transfer buffer.<br>• Fill the tank with enough transfer buffer to cover the top of the transfer cassette. This prevents temperature gradients across the transfer cassette.<br>• Pre-chill the transfer buffer and carry out the electrotransfer in a cold room.<br>• Alternatively, reduce the current and run the electrotransfer for a longer time. |
| Significant proportion of proteins remain in the gel after the electrotransfer. | Too high concentration of methanol in the transfer buffer. | Reduce the concentration of methanol in the transfer buffer to 15%. |

# 8 Reference information

## About this chapter

This chapter lists the technical specifications of DIGE Unit LF24 instrument. The chapter also includes a chemical resistance guide, recycling information, regulatory information, ordering information, and Health and Safety Declaration form for return.

## In this chapter

# 8.1 Specifications

## Technical specification

| Parameter | Specification |
|---|---|
| Gel cassette size (W × H × D) | 277 × 217 × 7 mm |
| Gel size (W × H × D) | 255 × 186 × 1 mm |
| Number of gels | 1 to 2 gels |
| Number of transfer cassettes | 1 to 2 transfer cassettes |
| Maximum voltage | 800 VDC |
| Maximum amperage | 400 mA |
| Maximum wattage | 100 W |
| Maximum buffer temperature | 35°C[1] |
| Buffer volume (electrophoresis) | 4.5 L anode buffer<br>1.2 L cathode buffer |
| Buffer volume (electrotransfer) | Approximately 7.0 L transfer buffer, depending on the number of transfer stacks in place. |
| Dimensions | See *Section 4.2.2 Dimensions and weight, on page 29*. |
| Weight | See *Section 4.2.2 Dimensions and weight, on page 29*. |

[1] When using ethanol in the transfer buffer, make sure that the buffer temperature does not exceed 30°C. Heating flammable liquids above their flash point can cause fire.

**CAUTION**

**Fire hazard**. Do not exceed the maximum buffer temperature. Heating flammable liquids above their flash point can cause fire.

## Environmental requirements

The installation site must comply with the following specifications.

| Parameter | Requirement |
|---|---|
| Allowed location | Indoor use only |
| Ambient temperature, operating | 4°C to 35°C |

| Parameter | Requirement |
|---|---|
| Ambient temperature, storage | 4°C to 40°C |
| Relative humidity, operating | 20% to 95%, non-condensing |
| Relative humidity, non-operating | 20% to 95%, non-condensing |
| Altitude, operating | Up to 2000 m |
| Pollution degree of the intended environment | Pollution degree 2 |

# 8.2 Chemical resistance

## Introduction to chemical resistance

This section gives general guidelines concerning chemical resistance for the product. Regarding exposure to solutions not covered by these guidelines, contact your Cytiva representative for recommendations.

| Chemical | Concentration | CAS no. /EEC no. |
|---|---|---|
| Decon 90 | 10% | 1310-58-3/215-181-3 |
| Ethanol | 20% | 64-17-5/200-578-6 |
| Methanol | 20% | 67-56-1/200-659-6 |
| Spray with commercial house cleaning detergent. | 5% | N/A |

# 8.3 Recycling information

## Introduction

This section contains information about the decommissioning of the product.

**CAUTION**

Always use appropriate personal protective equipment when decommissioning the equipment.

## Decontamination

The product must be decontaminated before decommissioning. All local regulations must be followed with regard to scrapping of the equipment.

## Disposal of the product

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

## Disposal of electrical components

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

## Disposal instructions

Follow the instructions below before disposing of the product.

| Step | Action |
|---|---|
| 1 | Decontaminate the product. |
| 2 | Separate all the electric components (power leads, electrodes and banana connectors) from the product. |
| 3 | Dispose or recycle the cardboard boxes, the plastic parts and the electronic parts according to local regulations. |

# 8.4 Regulatory information

## Introduction

This section lists the regulations and standards that apply to the product. Your system is marked or listed according to the applicable regulatory requirements for your region. Local language translations are provided according to regulatory requirements.

## In this section

# 8.4.1 Contact information

## Contact information for support

To find local contact information for support and sending troubleshooting reports, visit *cytiva.com/contact*.

## Manufacturing information

The table below summarizes the required manufacturing information.

| Requirement | Information |
|---|---|
| Name and address of manufacturer | Cytiva Sweden AB<br>Björkgatan 30<br>SE 751 84 Uppsala<br>Sweden |
| Telephone number of manufacturer | + 46 771 400 600 |

# 8.4.2 European Union and European Economic Area

## Introduction

This section describes regulatory information for the European Union and European Economic Area that applies to the product.

## Conformity with EU Directives

See the EU Declaration of Conformity for the directives and regulations that apply for the CE marking.

If not included with the product, a copy of the EU Declaration of Conformity is available on request.

## CE marking

The CE marking and the corresponding EU Declaration of Conformity is valid for the product when it is:

- used according to the *Operating Instructions* or user manuals, and
- used in the same state as it was delivered, except for alterations described in the *Operating Instructions* or user manuals.

## Regulatory compliance of connected equipment

Any equipment connected to the product must meet the safety requirements of IEC/EN/UL/CSA 61010-1 or other relevant national safety regulations and standards. Within the European Union, connected equipment must be CE-marked.

**NOTICE**

Any equipment connected to the product must be installed and used according to the manufacturer's instructions, and the environmental requirements and the electrical power requirements of the equipment.

# 8.4.3 Great Britain

## Introduction

This section describes regulatory information for Great Britain that applies to the equipment.

## Conformity with UK Regulations

See the UK Declaration of Conformity for the regulations that apply for the UKCA marking.

If not included with the product, a copy of the UK Declaration of Conformity is available on request.

## UKCA marking


The UKCA marking and the corresponding UK Declaration of Conformity is valid for the instrument when it is:

- used according to the *Operating Instructions* or user manuals, and
- used in the same state as it was delivered, except for alterations described in the *Operating Instructions* or user manuals.

## 8.4.4 Eurasian Economic Union (Евразийский экономический союз)

This section describes the information that applies to the product in the Eurasian Economic Union (the Russian Federation, the Republic of Armenia, the Republic of Belarus, the Republic of Kazakhstan, and the Kyrgyz Republic).

### Introduction

This section provides information in accordance with the requirements of the Technical Regulations of the Customs Union and (or) the Eurasian Economic Union.

### Введение

В данном разделе приведена информация согласно требованиям Технических регламентов Таможенного союза и (или) Евразийского экономического союза.

### Manufacturer and importer information

The following table provides summary information about the manufacturer and importer, in accordance with the requirements of the Technical Regulations of the Customs Union and (or) the Eurasian Economic Union.

| Requirement | Information |
|---|---|
| Name, address and telephone number of manufacturer | See *Manufacturing information* |
| Importer and/or company for obtaining information about importer | Cytiva RUS LLC<br>109004, Moscow<br>internal city area Tagansky municipal district<br>Stanislavsky str., 21, building 5, premises I, offices 24,25,29<br>Russian Federation<br>Telephone: +7 985 192 75 37<br>E-mail: *rucis@cytiva.com* |

### Информация о производителе и импортере

В следующей таблице приводится сводная информация о производителе и импортере, согласно требованиям Технических регламентов Таможенного союза и (или) Евразийского экономического союза.

| Требование | Информация |
|---|---|
| Наименование, адрес и номер телефона производителя | См. *Информацию об изготовлении* |
| Импортер и/или лицо для получения информации об импортере | ООО "Цитива РУС"<br>109004, г. Москва<br>вн. тер. г. муниципальный округ Таганский<br>ул. Станиславского, д. 21 стр. 5, помещ. I, ком. 24,25,29<br>Российская Федерация<br>Телефон: +7 985 192 75 37<br>Адрес электронной почты: *rucis@cytiva.com* |

## Description of symbol on the system label
## Описание обозначения на этикетке системы

This Eurasian compliance mark indicates that the product is approved for use on the markets of the Member States of the Customs Union of the Eurasian Economic Union

Данный знак о Евразийском соответствии указывает, что изделие одобрено для использования на рынках государств-членов Таможенного союза Евразийского экономического союза

## 8.4.5 Declaration of Hazardous Substances (DoHS)

This section describes the information that applies to the product in China.

根据 SJ/T11364-2014《电子电气产品有害物质限制使用标识要求》特提供如下有关污染控制方面的信息。

The following product pollution control information is provided according to SJ/T11364-2014 Marking for Restriction of Hazardous Substances caused by electrical and electronic products.

### 电子信息产品污染控制标志说明
### Explanation of Pollution Control Label

该标志表明本产品不含有超过中国标准 GB/T 26572《电子信息产品中有毒有害物质的限量要求》中限量的有毒有害物质,报废后可以进行回收处理,不能随意丢弃。

This symbol indicates that this electrical and electronic product does not contain any hazardous substances above the maximum concentration value established by the Chinese standard GB/T 26572 , and can be recycled after being discarded, and should not be casually discarded.

# 有害物质的名称及含量
# Name and Concentration of Hazardous Substances

产品中有害物质的名称及含量
Table of Hazardous Substances' Name and Concentration

| 部件名称<br>Component name | 有害物质<br>Hazardous substance | | | | | |
|---|---|---|---|---|---|---|
| | 铅<br>(Pb) | 汞<br>(Hg) | 镉<br>(Cd) | 六价铬<br>(Cr(VI)) | 多溴联苯<br>(PBB) | 多溴二苯醚<br>(PBDE) |
| 29701935 | 0 | 0 | 0 | 0 | 0 | 0 |
| 29701936 | 0 | 0 | 0 | 0 | 0 | 0 |

**0:** 表示该有害物质在该部件所有均质材料中的含量均在 GB/T 26572 规定的限量要求以下。

**X:** 表示该有害物质至少在该部件的某一均质材料中的含量超出 GB/T 26572 规定的限量要求。

- 此表所列数据为发布时所能获得的最佳信息.

**0:** Indicates that this hazardous substance contained in all of the homogeneous materials for this part is below the limit requirement in GB/T 26572.

**X:** Indicates that this hazardous substance contained in at least one of the homogeneous materials used for this part is above the limit requirement in GB/T 26572

- Data listed in the table represents best information available at the time of publication.

# 8.5 Ordering information

## Introduction

This section lists the main products, accessories and user replaceable spare parts that are available for the product.

Visit *cytiva.com*, and locate the Electrophoresis and isoelectric focusing product page, to find the latest ordering information.

## Main product

| Product | Quantity | Product code |
|---|---|---|
| DIGE Unit LF24 | 1 | 29701935 |

## Accessories

| Product | Quantity | Product code |
|---|---|---|
| Transfer Unit LF24 | 1 | 29701936 |
| EPS 3501 XL | 1 | 18113005 |
| Power supply adapter (2 mm M/4 mm F) | 2 | 18112959 |

## Consumables

| Product | Quantity | Product code |
|---|---|---|
| DIGE Gels LF24 | 3 | 29706670 |
| DIGE Buffer Kit | 1 | 28937452 |

## User-replaceable spare parts

| Product | Quantity | Product code |
|---|---|---|
| Transfer Sponges DIGE LF24 | 4 | 29709626 |
| Safety lid DIGE Unit LF24 | 1 | 29701940 |

# 8.6 Health and Safety Declaration Form

## Product return or servicing

cytiva

### Health & Safety Declaration Form for Product Return or Servicing

| | | | |
|---|---|---|---|
| **Return authorization number:** | | *and/or* **Service Ticket/Request:** | |

To make sure the mutual protection and safety of Cytiva personnel, our customers, transportation personnel and our environment, all equipment must be clean and free of any hazardous contaminants before shipping to Cytiva. To avoid delays in the processing of your equipment, complete this checklist and include it with your return.

1. Note that items will NOT be accepted for servicing or return without this form
2. Equipment which is not sufficiently cleaned prior to return to Cytiva may lead to delays in servicing the equipment and could be subject to additional charges
3. Visible contamination will be assumed hazardous and additional cleaning and decontamination charges will be applied

| Yes | No | Specify if the equipment has been in contact with any of the following: | |
|---|---|---|---|
| ◯ | ◯ | Radioactivity (specify) | |
| ◯ | ◯ | Infectious or hazardous biological substances (specify) | |
| ◯ | ◯ | Other Hazardous Chemicals (specify) | |

**Equipment must be decontaminated prior to service / return. Provide a telephone number where Cytiva can contact you for additional information concerning the system / equipment.**

| | | |
|---|---|---|
| **Telephone No:** | | |
| **Liquid and/or gas in equipment is:** | ☐ | **Water** |
| | ☐ | **Ethanol** |
| | ☐ | **None, empty** |
| | ☐ | **Argon, Helium, Nitrogen** |
| | ☐ | **Liquid Nitrogen** |
| | **Other, specify** | |

| | | | |
|---|---|---|---|
| **Equipment type / Product No:** | | **Serial No:** | |

**I hereby confirm that the equipment specified above has been cleaned to remove any hazardous substances and that the area has been made safe and accessible.**

| | | | |
|---|---|---|---|
| **Name:** | | **Company or institution:** | |
| **Position or job title:** | | **Date (YYYY/MM/DD)** | |
| **Signed:** | | | |



For local office contact information, visit cytiva.com/contact.
28980027 AD 04/2020

**To receive a return authorization number or service number, call local technical support or customer service.**

# Index

## G



## I



## L



## M



## N



## O



## P



## R



## S



## T



## U

Page intentionally left blank

cytiva

**Give feedback on this document**

Visit cytiva.com/techdocfeedback or scan the QR code.

cytiva.com



Melanie has been developed by the Swiss Institute of Bioinformatics in collaboration with GeneBio and Cytiva.



For local office contact information, visit cytiva.com/contact

29701249 AA V:13 11/2022