WSE-7056

# EzRun ClearNative Instruction Manual

September 18, 2025 3rd edition

## 1.Precautions for safe use of this product

To use this product safely, please read this instruction manual carefully first. Please refrain from operating the product until you fully understand the contents of this instruction manual. This instruction manual describes only how to use this product for the specified purpose. Please refrain from using the product for purposes or in ways not described in this instruction manual. If you use the product for purposes or in ways not described in this instruction manual, you are solely responsible for all necessary safety measures and unforeseen circumstances. Also, please carefully read and understand the instruction manuals of any devices you will be using at the same time.

## 2.Purpose of use

This product is electrode buffer for High-Resolution-Clear-Native PAGE using SDS-free Tris-Glycine polyacrylamide precast gels and homemade gels (Laemmli compliant).

## 3.Product configuration

| Name | | Volume | Quantity |
|---|---|---|---|
| **EzRun ClearNative** | For Cathode | 500mL | 1 |
| | For Anode | 500mL | 1 |

## 4.Composition

| Name | | Main components |
|---|---|---|
| **EzRun ClearNative** | For Cathode | Tris, Anionic surfactant |
| | For Anode | Tris |

This Product does not contain any poisonous or deleterious substances under the Poisonous and Deleterious Substances Control Law, or any substances subject to notification that exceed the exemption amounts stipulated under the Industrial Safety and Health Law or the PRTR Law. For details, please download the SDS for this product from the ATTO website (https://www.atto.co.jp/).

## 5.Storage

- Store this product room temperature (15–30°C), protected from direct sunlight. Unopened products are stable until the expiration date.
- Solutions prepared by diluting this product should be stored tightly closed at room temperature (15–30°C), protected from direct sunlight.
- When this product is stored under low-temperature conditions, some components may precipitate. If precipitation occurs, warm the product in water at approximately 40–50 °C before use and ensure that all precipitated components are completely dissolved.
- Electrophoresis buffer that has been used once cannot be reused.

## 6.Disposal method

- Dispose of each reagent in accordance with the disposal method of your affiliated institution.
- Bottle Material Body and lid : Polypropylene

## 7.Items required other than this product

- Magnetic stirrer
- Stirrer bar
- Beaker
- Graduated cylinder
- Medium bottles or other containers
- Distilled water
- Tris-Glycine polyacrylamide gel (recommended: **u-PAGEL H**)
- Filter paper (e.g., **CB-09A**)
- Blotting membrane (e.g., **WSE-4051**)
- Blotting buffer (recommended: **WSE-7210 EzFastBlot HMW**)
- Electrophoresis tank: (recommended: **AE-6530**, **WSE-1165 Mini-Slab**, etc.)
- Blotting apparatus: (recommended: **WSE-4025 HorizBlot 2M,** **WSE-1115 Powered Blot Ace** etc.)
- Power supply: (recommended: **WSE-3100 Power Station Ghibli I**)

## 8.Precations for use

- This product is a 5x stock solution. Dilute according to the instructions before use.
- This product is intended for High-Resolution-Clear-Native PAGE and cannot be used for SDS-PAGE.
- This product is supplied as a set for the cathode and anode.
- Use the anode buffer in the lower chamber of the electrophoresis tank and the cathode buffer in the upper chamber.
- This product can be used with Tris-Glycine gels that do not contain SDS. It is not compatible with Bis-Tris gels or Tris-Acetate gels.
- Some components of this product may precipitate. If this occurs, warm the product in water at approximately 40–50 °C before use and ensure that all precipitated components are completely dissolved.

## 9.How to use

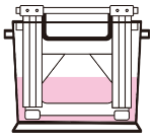
A. Preparation

For Cathode:
Dilute **EzRun ClearNative for Cathode (the bottle with a blue label)** 5 times with distilled water.
For Anode:
Dilute **EzRun ClearNative for Anode (the bottle with a red label)** 5 times with distilled water.

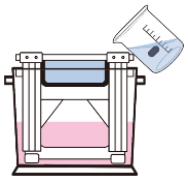
B. Electrophoresis

**Note**: When using this product, the procedure differs from the instructions provided with the device. Please follow the steps below.

1. Set the gel in the electrophoresis tank.
2. Add an appropriate volume of the diluted **EzRun ClearNative for Anode (the bottle with a red label)** to the lower chamber (anode side). Add the anode buffer to the electrophoresis tank while tilting the tank to prevent air bubbles from remaining at the bottom of the gel.



3. Add an appropriate volume of the diluted **EzRun ClearNative for Cathode (the bottle with a blue label)** to the upper chamber (cathode side).
   * The required volume of buffer depends on the tank. Please refer to the table below.



| Gel size | apparatus | Required Volume of Buffer |
|---|---|---|
| Mini | **AE-6530**<br>**WSE-1150** | Cathode: 75 mL<br>Anode: 250 mL |
| | **WSE-1165** | Cathode: 250 mL<br>Anode: 250 mL |
| Compact | **WSE-1010**<br>**WSE-1025** | Cathode: 120 mL<br>Anode: 70 mL |
| | **WSE-1030**<br>**WSE-1040** | Cathode: 135 mL<br>Anode: 110 mL |
| Wide | **WSE-1170** | Cathode: 400 mL<br>Anode: 400 mL |

4. Apply the samples.
5. Connect the electrophoresis tank to the power supply and perform electrophoresis. Run the gel until the dye (BPB), which indicates the electrophoretic front, or the standard reaches 5 -10mm above the bottom of the gel.
   For one mini-size gel, electrophoresis can be performed at 20 mA constant current for 70–80 minutes, or at 150 V constant voltage for 80–90 minutes.
   - If using **PageRunAce (WSE-1150)**: run in STD mode for 70–80 minutes.
   - If using **Compact PAGE Ace (WSE-1010/25)**: run in STD mode for 20–30 minutes.
   - If using **Compact PAGE Neo (WSE-1030/40)**: run at 150 V constant voltage for 35–40 minutes.
   - For wide-size gels: run at 30 mA constant current or 150 V constant voltage.

   *Follow the instructions provided with each device for proper use

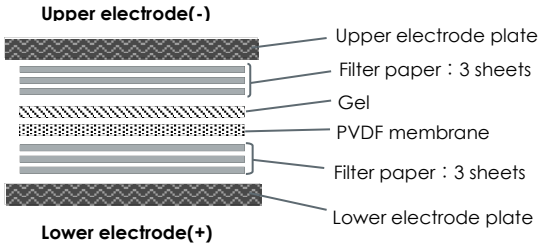
Gel staining

After electrophoresis, immerse the gel in a staining solution or a fixing solution, and perform staining and destaining. CBB staining, reverse staining, or fluorescent staining can be used. For blotting, proceed to step C without staining the gel.

C Blotting

*Semidry using **EzFastBlot HMW (WSE-7210)** or conventional wet transfer is recommended.
*Prepare the following buffers before starting blotting
*Pre-wet the PVDF membrane with methanol, and then equilibrate it in 1x **EzFastBlot HMW** (transfer buffer).

1. Prepare SDS-containing buffer by adding sodium dodecyl sulfate (SDS) to 1×**EzFastBlot HMW** so that the final concentration is 0.1%. For example, dissolve 50 mg of SDS in 50 mL of 1×**EzFastBlot HMW** to prepare the buffer solution. Approximately 50 mL of buffer is required per one mini-size gel (85 × 90 mm).
2. After electrophoresis, Immerse the gel in the 0.1% SDS solution prepared in step1, gently agitate to equilibrate for 20 minutes.
3. Immerse the equilibrated gel in 1×EzFastBlot HMW, agitate gently, and rinse for 10 minutes.
4. Assemble the blotting sandwich in the following order, referring to the diagram below:.




   1) Drop a few milliliters of 1×EzFastBlot HMW onto the lower electrode plate to pre-wet it.
   2) Place 3 sheets of filter paper soaked in 1×EzFastBlot HMW on the lower electrode plate.
   3) Place the PVDF membrane on top of the filter paper.
   4) Drop a few milliliters of 1×EzFastBlot HMW onto top of the PVDF membrane.
   5) Carefully place the gel on the PVDF membrane, ensuring that no air bubbles are trapped between the gel and the membrane.
   6) Place 3 sheets of filter paper soaked in 1×EzFastBlot HMW on top of the gel.
   7) Using a blotting roller or similar tool, gently remove any excess buffer or air bubbles between the gel and the PVDF membrane, ensuring tight contact.
   8) Gently lower the upper electrode plate onto the stacked filter paper.
5. Connect the blotting apparatus and power supply using the leads. Follow the instructions provided with each device for proper operation.
6. Apply a constant voltage of 12 V for 30–60 minutes, or a constant voltage of 24 V for 15–30 minutes,
7. Perform blocking, antibody reactions, and detection as usual.
   *When using transfer buffers other than **EzFastBlot HMW**, follow the same blotting procedure described above.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