

Rapid Total Protein Staining Kit

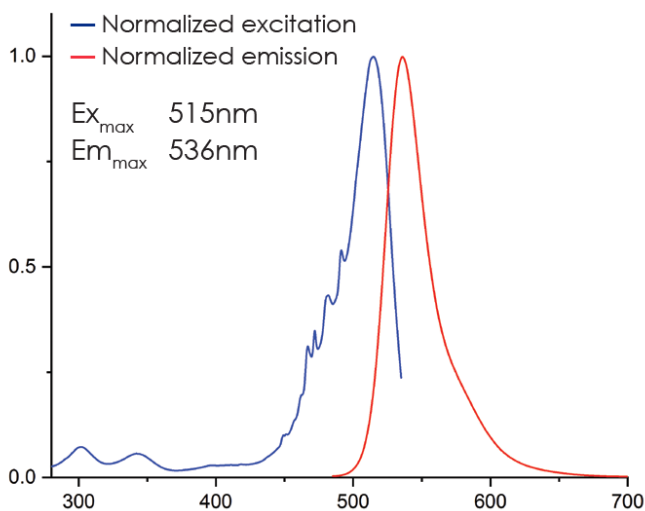
Description:

Rapid Total Protein Staining Kit is a fluorescent protein stain for PVDF and Nitrocellulose membranes that allows for sensitive and quantitative visualization of protein bands.

For precise quantitative Western blot data, it is critical to perform normalization to account for sample inconsistencies, sample loading errors and uneven transfer. In an effort to increase transparency, total protein normalization is quickly becoming the standard for Western blot normalization as it is now the preferred method for top tier journals. The Rapid Total Protein Staining Kit provides a convenient method for total protein normalization that is compatible with digital imaging systems equipped with a Cy3-compatible filter.

- **CONVENIENT** – Simple protocol that takes less than 20 minutes, no de-staining required
- **VERSATILE** – Compatible with chemiluminescent and IR700/IR800 fluorescent Western blotting
- **ACCURATE** – Linear across a broad range from 1-50µg of lysate, ideal for total protein normalization

- Rapid Total Protein Staining can be imaged with a CCD or laser based imaging system using green-light excitation.



Workflow:

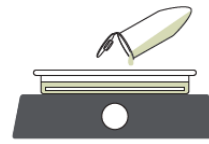
1



After electrotransfer is complete, transfer blot to incubation tray filled with water

Incubate membrane for 5 minutes at RT with gentle agitation.

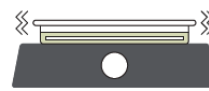
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Decant water and dispense 5–10mL of AdvanStain Total Staining Solution into incubation tray

Incubate PVDF membranes for 5 minutes and Nitrocellulose membranes for 10 minutes at RT with gentle agitation.

3



Wash membrane

3X for 3 minutes with 5–10mL Washing Solution with gentle agitation.

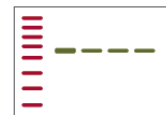
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Image blot for total protein analysis

PVDF membranes may be imaged wet or dry using green-light excitation. Nitrocellulose membranes must be imaged wet.

5



Prepare for Western blotting

If the PVDF membrane is imaged wet, rinse blot with water before proceeding with Western blot. If imaged dry, pre-wet the blot with 100% Methanol then rinse with water before proceeding with Western blot.

Short Protocol for PVDF Membranes:

1. Prepare the solutions:

- **Washing Solution:** Before using the product for the first time, add 135ml of Ethanol directly to the bottle of Washing Solution and mix well. Check the box on the bottle's label. This only needs to be done one time.

Note: This protocol is for staining one full-size mini-blot. If using a larger blot, adjust the volume of staining and washing solution appropriately.

2. Following the transfer, immerse the blot in at least 10mL of water for 5 minutes with gentle agitation.
3. Decant water and dispense 5–10mL of PVDF Staining Solution for 5 minutes with gentle agitation.
4. Decant the PVDF Staining Solution and wash the membrane three times for 3 minutes per wash with 5–10mL Washing Solution.
5. Image the blot for permanent record. If imaging wet, immediately place the blot in a Imaging Folder before imaging. If imaging dry, allow the blot to completely dry (no visible streaks should be observed).
6. Following imaging, immerse the blot in at least 10mL of water for 5 minutes with gentle agitation before proceeding with Western blotting. If the blot was imaged dry, rewet with methanol prior to rinsing with water.

Short Protocol for NC Membranes:

1. Prepare the solutions:

- **NC Staining Solution:** Before using the product for the first time, add 30mL of Ethanol directly to the bottle of NC Staining Solution and mix well. Check the box on the bottle's label. This only needs to be done one time.
- **Washing Solution:** Before using the product for the first time, add 135ml of Ethanol directly to the bottle of Washing Solution and mix well. Check the box on the bottle's label. This only needs to be done one time.

Note: This protocol is for staining one full-size mini-blot. If using a larger blot, adjust the volume of staining and washing solution appropriately.

2. Following the transfer, immerse the blot in at least 10mL of water for 5 minutes with gentle agitation.
3. Decant water and dispense 5–10mL of Working NC Staining Solution for 10 minutes with gentle agitation.
4. Decant the NC Staining Solution and wash the membrane three times for 3 minutes per wash with 5–10mL Washing Solution.
5. Immediately place the blot in a Imaging Folder and image the wet blot for permanent record. Do not allow the blot to dry prior to imaging.
6. Following imaging, immerse the blot in at least 10mL of water for 5 minutes with gentle agitation before proceeding with Western blotting.

For Orders:

Catalog Number	Product	Size
NB051106115	Rapid Total Protein Staining Kit for PVDF membranes	15 blots
NB051106215	Rapid Total Protein Staining Kit for NC membranes	15 blots

For orders: destek@nepenthe.com.tr

This kit is for research use only. Not for use in vitro diagnostics.

www.nepenthe.com.tr