

Novex[®] AP Chemiluminescent Detection Kit

Cat. nos. SLF1021, SLF1022

Store at 2°C to 8°C

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Rev. 1.0

Description

Novex[®] AP Chemiluminescent Detection Kits are designed for use as part of the western detection protocol for the iBind[™] Western System. For details on performing western detection with the iBind[™] Western Device, refer to the manual at www.lifetechnologies.com.

Components

Item	SLF1021	SLF1022	Storage
Anti-Mouse Secondary Antibody Alk-Phosphatase	100 µL	—	2°C to 8°C
Anti-Rabbit Secondary Antibody Alk-Phosphatase	—	100 µL	2°C to 8°C
Novex [®] AP Chemiluminescent Substrate (CDP-Star [®])	25 mL	25 mL	2°C to 8°C
Chemiluminescent Substrate Enhancer (NitroBlock II [™])	2.5 mL	2.5 mL	2°C to 8°C

For Research Use Only. Not for use in diagnostic procedures.

General guidelines

- Alkaline phosphatase labeled anti-mouse, or anti-rabbit secondary antibodies are provided for use in the secondary antibody binding step of the iBind™ western detection protocol.
- Chemiluminescent detection is performed after blocking, antibody binding, and washes have been completed by the iBind™ Western Device.
- Add Chemiluminescent Substrate Enhancer to the Novex® AP Chemiluminescent Substrate when using nitrocellulose membranes.

Prepare solutions

Item	Nitrocellulose	PVDF
Novex® AP Chemiluminescent Substrate (CDP-Star®)	2.375 mL	2.5 mL
Chemiluminescent Substrate Enhancer (NitroBlock II™)	0.125 mL	—

Prepare membrane

After performing blocking, antibody binding, and washes with the iBind™ Western Device:

1. Remove the membrane from the iBind™ Card and place it in a tray containing 20 mL of distilled water.
2. Discard the used iBind™ Card.
3. Rinse the membrane with 20 mL of distilled water and then decant.
4. Proceed to “Chemiluminescent detection.”

Chemiluminescent detection

1. Place the membrane on a sheet of transparency plastic with the **protein-side up**. Do not allow the membrane to dry out.
2. With a clean pipette, apply 2.5 mL of the chemiluminescent substrate solution evenly across the membrane surface (do not touch the membrane surface with the pipette).
3. Incubate for 5 minutes.
4. Blot excess chemiluminescent substrate solution from the membrane surface with filter paper. Do not allow the membrane to dry out.
5. Cover the membrane with another clean piece of transparency plastic, or with plastic wrap.
6. Place a piece of X-ray film over the membrane sandwich and expose for 1 second to several minutes, and develop the X-ray film,

OR

Scan the membrane sandwich in a digital imager.

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