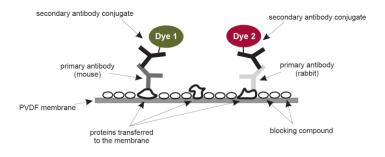


# **Multi-color IR Western Blotting Pack**

#### **Description:**

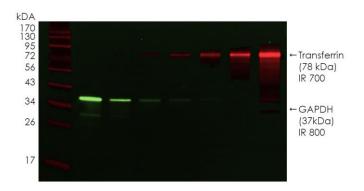
Multi-color IR Western Blotting Pack near infrared (IR) fluorescent Western blotting kits allow to two proteins at once, increasing the quality and quantity of information that can be gained from single blot. Assay a control alongside a protein of interest. Assay phosphorylated and un-phosphorylated isoforms of a protein simultaneously. The fluorescent dyes provided with the packs allow detection of low pg of protein. Multi-color IR Western Blotting Pack protocols save time and money since there is no need for disposable film, and the blot can be imaged immediately, without drying.

- MULTIPLEXING Visualize two proteins simultaneously
- SENSITIVITY Low pictogram detection
- QUALITY REAGENTS Includes Easy Protein-Free Blocking Buffer and Easy IR Western Wash Buffer
- FAST RESULTS 3.5 hours for entire protocol
- FLUORESCENT DETECTION Near IR (IR700 and IR800) kits with fluorescent secondary antibodies

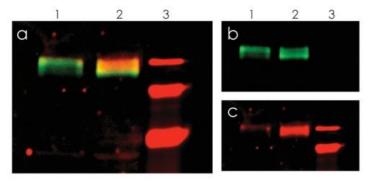


#### - Two colors, two proteins, simultaneously

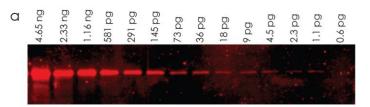
By using dyes that have different spectral properties (excitation and emission), you can visualize them in different channels at the same time on a digital imager.



Also known as multiplexing, simultaneous detection with different antibodies is ideal for certain types of experiments. For example, assay a loading control alongside a protein of interest; assay for two proteins you want to quantify relative to each other; or, assay phosphorylated and non-phosphorylated isoforms of a protein simultaneously.



- Picogram detection levels and Linear data



## **Short Protocol:**

- 1. Prepare your protein blot
- Prepare 20 ml of 1x Easy Protein-Free Blocking Buffer. Block membrane in 10 ml 1x Easy Protein-Free Blocking Buffer for 10 minutes at room temperature with gentle agitation.
- Incubate blot with primary antibody diluted in 10 ml of 1x Easy Protein-Free Blocking Buffer or the users preferred blocking buffer for one hour at RT with gentle agitation.
- 4. Prepare 100 ml 1x Easy IR Western Wash Buffer. Wash blot with 1x Easy IR Western Wash Buffer:
  - 1 x quickly with 10 ml
  - 1 x 15 min with 10 ml
  - 3 x 5 min with 10 ml each time
- 5. Incubate blot with secondary IR antibodies diluted 1:10,000 (1  $\mu l$  each) in 10 ml of 1x Easy IR Western Wash Buffer for 1 hour at room temperature with gentle agitation
- 6. Wash blot with 1x Easy IR Western Wash Buffer:
  - 3 x 5 min with 10 ml each time
  - 1 x 5 min with 20-50 ml PBS or TBS without detergent
- Place blot on background quenching sheet and drain excess liquid; blot may be imaged immediately, or stronger signal may be obtained by waiting 15 to 30 minutes for membrane to become semi-dry
- Image using CCD camera; for most instruments the settings for IR680 and IR800 dyes will work well for the Multi-color IR Western Blotting Pack conjugates

## For Orders:

Catalog Number	Product	Size
NB051202210	Multi-color IR Western Blotting Pack, Goat-anti-rabbit IgG IR700/ Goat-anti-mouse IgG IR800	10 assays
NB051202310	Multi-color IR Western Blotting Pack, Goat-anti-mouse IgG IR700/ Goat-anti-rabbit IgG IR800	10 assays

#### Kit Contents of NB051202210:

Item	Quantity
Low-fluorescence PVDF transfer membrane	10 pcs
Background Reduction Sheets	10 pcs
Easy Protein-Free Blocking Buffer	50 mL
Easy IR Western Wash Buffer	120 mL
Goat-anti-rabbit IgG IR700	20 µL
Goat-anti-mouse IgG IR800	20 µL

## Kit Contents of NB051202310:

Item	Quantity
Low-fluorescence PVDF transfer membrane	10 pcs
Background Reduction Sheets	10 pcs
Easy Protein-Free Blocking Buffer	50 mL
Easy IR Western Wash Buffer	120 mL
Goat-anti-mouse IgG IR700	20 µL
Goat-anti-rabbit IgG IR800	20 µL