Storage Information

The AdvanBlock™-PF reagent is stable at 4°C for at least one year.

Warnings and Precautions

- AdvanBlock™-PF is for research use only.
- Always wear gloves when handling reagents.
- Refer to MSDS for additional safety information.
- The product is guaranteed to be free of manufacturer defects, and to function as described when the enclosed protocol is followed by properly trained personnel.

For More Information

visit **http://advansta.com/products/AdvanBlock-PF** or go directly to the web-page by scanning the QR-code with your favorite bar-code scanner app on your smart phone.

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AdvanBlock™-PF Blocking Solution

Non-protein blocking solution for fluorescent and chemiluminescent Western blots

For Catalog Number

R-03023-D20 AdvanBlock™-PF, 200 ml

Description

AdvanBlock™-PF is a protein-free blocking solution, optimized for use with the WesternBright™ MCF fluorescent Western blotting kit and also an excellent choice for chemiluminescent Western blot experiments. This fast-acting blocking solution stabilizes the fluorescence of the WesternBright™ MCF secondary antibody conjugates. AdvanBlock™-PF can reduce background when used with primary antibodies that have a high degree of cross-reactivity with protein blockers such as BSA, casein, or milk protein. With low-quality primary antibodies that may require protein-based blocking agents, BSA or non-fat dry milk can be dissolved directly in the AdvanBlock™-PF solution used to dilute the antibody. Provided as a 5X concentrate.

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Short Protocol

1. Prepare your protein blot on either a PVDF or Nitrocellulose membrane using your standard technique.
2. Block the membrane for 10 minutes at ambient temperature with gentle agitation using a sufficient volume of buffer to completely cover the membrane.
3. Incubate with the primary antibody diluted in AdvanBlock™-PF for one hour at ambient temperature with gentle agitation.
4. Wash the blot with AdvanWash™ or AdvanWash™-IR Washing Solution, PBST or TBST:
   - 2 x quickly (~5 seconds per rinse)
   - 3 x 5 minutes, with at least 0.3 mL/cm^2 membrane each time
5. Incubate with the secondary antibody diluted in AdvanBlock™-PF for one hour at ambient temperature with gentle agitation.
6. Wash the blot with AdvanWash™ or AdvanWash™-IR Washing Solution, PBST or TBST:
   - 2 x quickly (~5 seconds per rinse)
   - 3 x 5 minutes, with at least 0.3 mL/cm^2 membrane each time
7. Rinse the blot for 5 minutes with 1X PBS to remove detergent which may cause elevated fluorescent background.
8. For best results, image the blot dry.

Tips

- Western blotting requires optimization of primary and secondary antibody concentrations used in steps 3 and 5 of the Short Protocol. These must be determined empirically for every antigen-antibody pair.
- Fluorescent Western blotting can be performed as a common procedure utilizing a primary antibody and a fluorescently labeled secondary antibody. Alternatively, directly labeled primary antibodies may be used, which eliminates the need of secondary antibody and shortens the procedure. Adjust the protocol appropriately if using directly labeled primary antibodies.
- AdvanBlock™-PF increases sensitivity, so optimal antibody concentrations may be lower than with other blocking buffers.
- For IR dyes use AdvanWash™-IR Washing Solution for decreased non-specific background.
- Make sure not to touch the membrane with fingers or dirty forceps as this can result in non-specific background.

Notes

- Advansta provides a reliable technology to prepare fluorescently labeled antibodies in an easy to use SpectraDye Antibody Labeling Kit format.