www.rockland.com
tech@rockland.com
+1 484.791.3823

Protocol

Revitablot™ Stripping Buffer

The basics of cell culture for patient-derived melanoma cell lines share certain similarities, however, cell culture conditions vary typically for each melanoma cell line. Deviating from the culture conditions required for a particular melanoma cell line can result in different phenotypes being expressed. We recommend that you acquaint yourself with each cell line of interest and carefully follow the cell line specific instructions provided with each product.

I. Procedure for HRP-Conjugated & Fluorescent-Conjugated Antibodies

Note: Blots may be stripped up to four times for HRP-conjugated antibodies and up to two times for fluorescent-conjugated antibodies.

1. Image for the primary target and then wash blot for at least 10 minutes in 1X TBS-T on an orbital shaker at room temperature. This process removes any remaining HRP substrate.
2. Decant TBS-T and apply 10 mL of Revitablot™
   Note: Volume depends on size of blot; apply enough buffer to sufficiently cover the membrane
3. Incubate from 5–20 minutes (user determined) on an orbital shaker at room temperature.
4. Decant Revitablot™ buffer and wash blot for at least 10 minutes in 1X TBS-T as in Step 1.
5. Decant TBS-T and block blot in 5–10 mL of 5% BLOTTO (B501-0500) for 2 hours on an orbital shaker at room temperature.
6. Decant the BLOTTO and apply the next primary antibody target.
7. Incubate for the optimum time and conditions per provided instructions.
8. Complete the Western blot procedure (washing, application of secondary antibody, substrate addition, and imaging).