4. Incubate the membrane for 45 minutes at room temperature with 10 ml WestVision working solution with gentle agitation.

5. Wash the membrane 3x 5 minutes each in 10 ml PBST with gentle agitation.

6. Prepare substrate working solution according to the substrate kit instructions. Following are protocols for signal development using either a chromogenic or chemiluminescent substrate.

Chromogenic signal development with Vector® TMB substrate (SK-4400):

7. Equilibrate membrane for 2 minutes in PBS in a clean vessel.

8. Incubate membrane in the substrate working solution at room temperature with gentle agitation for 5 minutes or until suitable staining develops. Briefly rinse the membrane in PBS and air-dry.

Chemiluminescent signal development using DuoLuX® Substrate (SK-6604):

7. Equilibrate membrane for 2 minutes in PBS in a clean vessel.

8. Remove excess buffer by holding the membrane vertically and touching the edge of the membrane to absorbent paper.

9. Place membrane target-side-up on plastic wrap on a level surface.

10. Pipette 5 ml of DuoLuX Substrate working solution onto the membrane surface.

11. Incubate for 5 minutes under subdued light. (If high background is present, rinse the membrane in PBS for a few seconds and remove excess as in Step 8.)

12. Place the membrane between two pieces of plastic wrap or a clear sheet protector. Acquire image with the gel imager or expose the membrane to x-ray film for the appropriate time.

Detailed product listings, specifications, protocols and additional information is available on our website: vectorlabs.com