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Western Blot Analysis

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Protocol status: Working
We use this protocol and it's working

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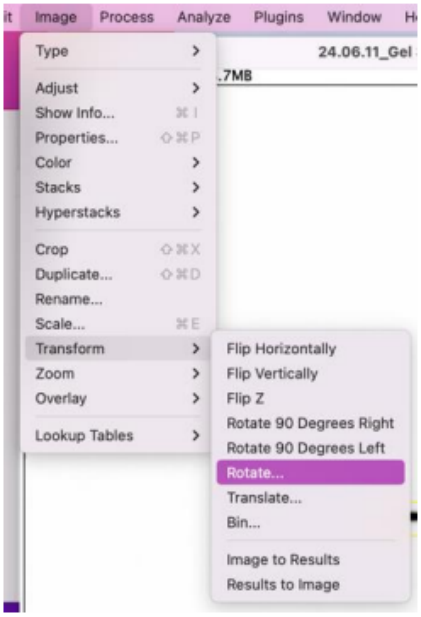
Abstract

This protocol details the western blot analysis.

Image J Western Blot

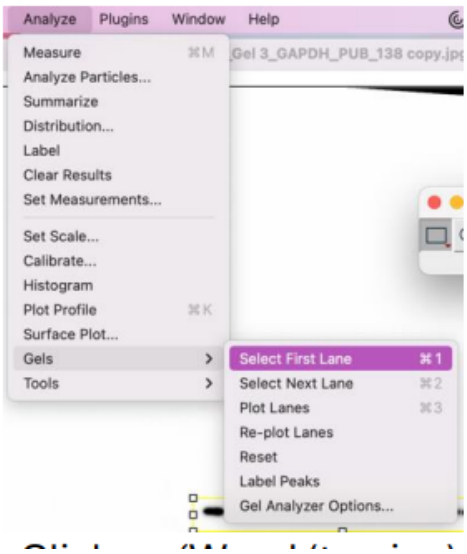
1 Drag Western Blot jpg file in Image J.

2 Click on ‘Image’ tab → ‘Transform’ → ‘Rotate’ and adjust image to make sure the lanes are horizontal.

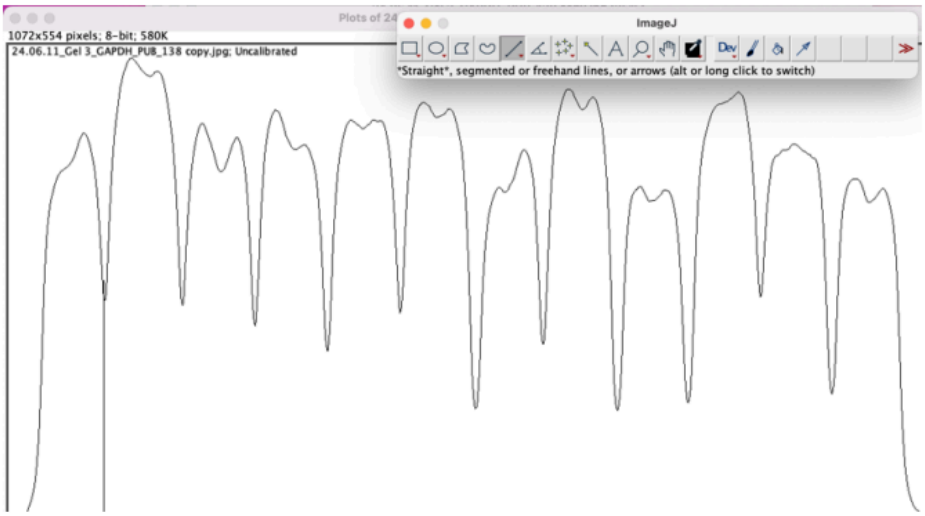


3 Click on the ‘rectangle tool’ and select the bands you want to analyze.

4 Click on the ‘Analyze’ tab →’Gels’ → ‘Select First Lane’ (or Control 1) → ‘Plot Lanes’ (or Control 3).



5 Click on ‘Straight’ tool to divide the peaks. Draw lines to divide peaks. Hold ‘Shift’ while drawing lines for a straight line.



6 Click on ‘Wand (tracing) tool’ and select each individual peak. A new window should pop up for the area under each peak.

7 Once you have all the areas (integrated densities) for each peak (band). Copy and paste the values into excel corresponding to their ID from the gel layout.

8 Do the same for the control protein.

9 Then divide the ID of your target protein/ID ctl.

10 Graph and do stats on these values in GraphPad.