

Update by Dr. Wahseng Lim on Immunoprecipitations (TrueBlot)
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I recently used a secondary from eBioscience (called Trueblot) that detects only IgG in its native confirmation and not denatured form. So when you use it in western blot of the protein that you have immunoprecipitated, it will only detect the IgG you use as the primary antibody and not the IgG you used for IP since the later is denatured by the denaturing gel. This means your blot will not show the heavy and light chains, which very often prevent you from seeing bands in their vicinities. It works really well.

Check out the following website:

<http://www.ebioscience.com/ebioscience/whatsnew/trueblot.htm>

Make sure you read the following paragraphs:

1. use of anti-Ig beads for precipitation - never use Protein A or Protein G beads when using Rabbit TrueBlot.
/*Protein A or G should not be used for IP in conjunction with Rabbit TrueBlot because protein A and G contaminants bind to rabbit IgG with high affinity - causing strong contaminating bands.* If a rabbit, mouse, rat or goat antibody is used for IP, use anti-rabbit Ig, anti-mouse Ig, anti-rat Ig or anti-goat Ig beads (respectively) for immunoprecipitation./
2. complete reduction of immunoprecipitate.
3. effective blocking of the immunoblot using milk as blocking protein, rather than BSA. BSA is not an effective blocker