# Your tool for the trouble-free detection of IP samples



# Are antibody heavy and light-chains complicating your IP experiments?



Then our VeriBlot for IP secondary antibodies are just what you need:

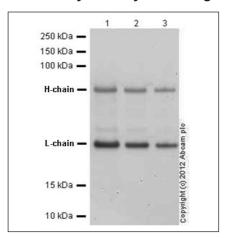
- No detection of denatured primary antibodies
- Clean IP sample detection without obstruction by heavy and light-chains



# Common problems with IP sample detection

Using secondary antibodies, which recognize the heavy and light-chains of the primary antibody for WB detection of IP samples will always result in two bands (the heavy-chain at 50kDa and the light-chain at 25kDa). These bands are due to the secondary antibody binding to the denatured/reduced primary antibody released with the antigen during the immunoprecipitation procedure. They can also result from the recognition of endogenous IgGs from the lysate itself.

### AbExcel Goat polyclonal Secondary Antibody to Mouse IgG – H&L (HRP) (ab97023)



#### WB legend:

Lane 1:  $0.5~\mu g$  Mouse monoclonal; Goat anti-mouse IgG – H&L (HRP) (ab97023) at 1/50,000 Lane 2:  $0.25~\mu g$  Mouse monoclonal; Goat anti-mouse IgG – H&L (HRP) (ab97023) at 1/50,000 Lane 3:  $0.125~\mu g$  Mouse monoclonal; Goat anti-mouse IgG – H&L (HRP) (ab97023) at 1/50,000

#### Problem:

- Protein of interest/immunoprecipitated protein is ~50kDa heavy-chain will obstruct its detection
- Protein of interest/immunoprecipitated protein is ~25kDa light-chain will obstruct its detection

#### Solutions:

- VeriBlot for IP secondary antibodies (for IP products <30kDa and co-immunoprecipitation experiments)
- 2. Light-chain specific secondary antibodies (for IP products >30kDa)

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#### Solution 1 - VeriBlot for IP

VeriBlot for IP secondary antibodies - only recognize native/non-reduced primary antibodies and therefore the detection of heavy and light-chains is highly minimized.

## VeriBlot for IP (for IP products <30kDa and co-immunoprecipitation experiments)

Product name	Conjugate	Number of blots	Product code
Anti-mouse IgG VeriBlot for IP secondary antibody	Biotin	~20	ab131367
Anti-mouse IgG VeriBlot for IP secondary antibody	HRP	~20	ab131368
Anti-rabbit IgG VeriBlot for IP secondary antibody	AP	~20	ab131365
Anti-rabbit IgG VeriBlot for IP secondary antibody	HRP	~20	ab131366

Tip: To completely reduce/denature the immunoprecipitate and to use milk as a blocking agent.

# Solution 2 - Light-chain specific secondary antibodies

As light-chain specific secondary antibodies do not recognize heavy-chains they are ideal for the detection of IP products >30kDa.

#### Alkaline phosphatase (AP) conjugated light-chain secondary antibodies

Product name	Number of blots	Product code
Mouse [JDC-12] to <b>Human</b> lambda - light chain	~500	ab99810
Rat [187.1] to Mouse kappa - light chain	~500	ab99616
Rat [JC5-1] to Mouse lambda - light chain	~500	ab99624
Rat [H139-52.1] to Mouse kappa - light chain	~500	ab99631
Mouse [SB62a] to Rabbit IgG - light chain	~500	ab99696
Mouse [K4F5] to <b>Rat</b> kappa - light chain	~500	ab99691

# Horseradish peroxidase (HRP) conjugated light-chain secondary antibodies

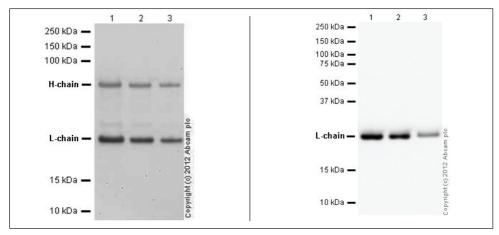
Number of blots	Product code
~500	ab99811
~500	ab99617
~500	ab99625
~500	ab99632
~500	ab99697
~500	ab99692
	~500 ~500 ~500 ~500 ~500

# Light-chain specific secondary antibodies

#### Detect IP products >30kDa without obstruction by the heavy-chain

H&L secondary antibody (ab97023), 1/50,000

Light-chain specific secondary antibody (ab99632), 1/10,000



Comparison of an anti-mouse secondary antibody recognizing H&L chains (ab97023) versus a mouse light-chain specific secondary antibody (ab99632)

# AbExcel secondary antibodies

This flyer lists key advantages of AbExcel secondary antibodies in western blotting:

- AbExcel products are extensively tested
- One vial of AP AbExcel secondary gives 1500+ blots
- One vial of HRP AbExcel secondary gives 600+ blots



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