

**Mononucleosome binding of GST-PHD: detection by Western**

**Binding buffers:** 300 mM NaCl, 0.1% NP40 and 50 mM Tris 7.5, 10% glycerol.  
200-400  $\mu$ l each binding.

Sample amount:

1  $\mu$ g of MonoNucleosome

10  $\mu$ g of GST-PHD

Binding: 4oC, O/N.

Add 30  $\mu$ l 50% glutathione beads, 4oC, 1 h.

Wash with 1 ml of binding buffer for 3 times.

Beads boiled in 50  $\mu$ l 2x SDS sample buffer.

Load 15-20  $\mu$ l for Western. For input, take 0.1  $\mu$ g nucleosome into 100  $\mu$ l, load 15-20  $\mu$ l, this is 5% input. Western detection with antibody to H3, or specific modifications.