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GST pull down assay

minghao chen¹, Xuefeng Ren¹

¹Department of Molecular and Cell Biology, University of California, Berkeley, Berkeley, CA 94720, USA.

ASAP Collaborative Research Network



Minghao Chen

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ABSTRACT

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

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- 1 Mix the GST tagged protein and fluorescent protein with 30 μ l Glutathione Sepharose beads (Cytiva) at final 1 μ M concentration in the buffer of 25 mM HEPES at pH 7.5, 150 mM NaCl, 1 mM MgCl₂ and 1 mM TCEP. The total volume is 200 μ l.
- 2 Rocking at  4 °C  Overnight
- 3 Wash the beads four times, then eluted in 50 μ l of buffer with 25 mM glutathione
- 4 Mix 18 μ l eluent in lithium dodecylsulfate (LDS)/BME buffer and subjected to SDS/PAGE gel without heating samples. The gel was scanned at 488 or 550 nm in ChemiDoc MP imaging system (Bio-Rad).