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Smart-IP Series

# Anti-HA-tag mAb-Magnetic Beads

<b>CODE No.</b>	M180-11MS
<b>CLONALITY</b>	Monoclonal
<b>CLONE</b>	TANA2
<b>ISOTYPE</b>	Mouse IgG2b $\kappa$
<b>QUANTITY</b>	4 tests (Slurry: 200 $\mu$ L)
<b>SOURCE</b>	Purified IgG from hybridoma supernatant
<b>IMMUNOGEN</b>	KLH conjugated synthetic peptide, YPYDVPDYA (HA-tag)
<b>REACTIVITY</b>	This antibody reacts with N-terminal and C-terminal HA-tagged proteins.
<b>FORMULATION</b>	10 mg magnetic beads in 1 mL PBS/0.1% BSA/0.1% ProClin 150
<b>STORAGE</b>	This beads suspension is stable for one year from the date of purchase when stored at 4°C.

## APPLICATION-CONFIRMED

Immunoprecipitation 50  $\mu$ L of beads slurry/sample

\*The purification capacity of Anti-HA-tag mAb-Magnetic Beads varies depending upon the characteristics of a HA-tagged protein. For example, 50  $\mu$ L of beads slurry binds 4.8  $\mu$ g of a HA-tagged protein (35 kDa).

## APPLICATION-REPORTED

RNP immunoprecipitation Reference 3) and 4)

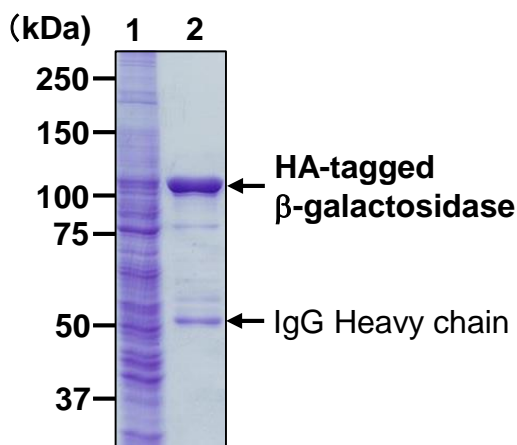
## REFERENCES

- 1) Kato, K., *et al.*, *Nat. Commun.* **9**, 2448 (2018) [IP]
- 2) Shichino, Y., *et al.*, *Elife* **7**, e32155 (2018) [IP]
- 3) Eiken, H. M., *et al.*, *Nat. Commun.* **8**, 1574 (2017) [RIP]
- 4) Jeong, H. W., *et al.*, *Nat. Commun.* **8**, 726 (2017) [RIP]

For more information, please visit our web site <http://ruo.mbl.co.jp/>

### **Immunoprecipitation**

- 1) Wash  $1 \times 10^7$  cells 3 times with PBS and suspend with 1 mL of cold Lysis buffer [50 mM Tris-HCl (pH 7.5), 150 mM NaCl, 1% NP-40] containing appropriate protease inhibitors.
- 2) Centrifuge the tube at  $12,000 \times g$  for 5 min. at  $4^\circ\text{C}$  and transfer the supernatant to another tube.
- 3) Add magnetic beads as suggested in the **APPLICATION** into 300  $\mu\text{L}$  of the supernatant prepared in step 2). Mix well and incubate with gentle agitation for 1 hr. at  $4^\circ\text{C}$ .
- 4) Place the tube on the magnetic rack (MBL; code no. 3190) for a few seconds.
- 5) Remove the supernatant.
- 6) Add 1 mL of cold Wash buffer [50 mM Tris-HCl (pH 7.5), 150 mM NaCl, 0.05% NP-40] and resuspend the magnetic beads.
- 7) Place the tube on the magnetic rack for a few seconds.
- 8) Remove the supernatant.
- 9) Repeat Steps 6)-8) 3 times.
- 10) Resuspend the magnetic beads in 20  $\mu\text{L}$  of Laemmli's sample buffer, boil for 5 min., and place the tube on the magnetic rack for a few seconds.
- 11) Load 20  $\mu\text{L}$  of the sample per lane in a 1-mm-thick SDS-polyacrylamide gel and carry out electrophoresis.
- 12) Visualize the protein bands by CBB staining.



### ***Immunoprecipitation of HA-tagged protein***

Sample: HA-tagged  $\beta$ -galactosidase/293T whole cell lysate

Lane 1: Input (10  $\mu\text{L}$ /lane)

Lane 2: Post-IP beads of Anti-HA-tag mAb (M180-11)