

Antibody Datasheet

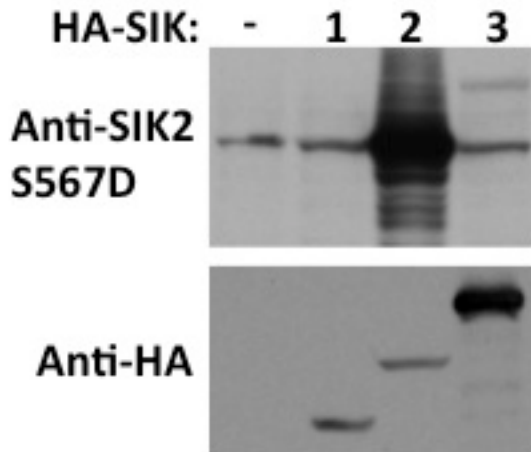
Product description:- Anti-SIK2 (272 – 445)
Antigen:- GST-SIK2 (272 – 445 mouse) [DU 43597]
Sheep Number:- S567D
Formulation:- Phosphate Buffered Saline
Storage temperature:- –20 °C
Purification Method:- Affinity purified against recombinant protein GST-SIK2 (272 – 445 mouse) [DU 42597] and then depleted against GST

Working Concentration:-

For immunoblotting use at 1 ug/ml (2nd and 3rd bleeds can detect endogenous)

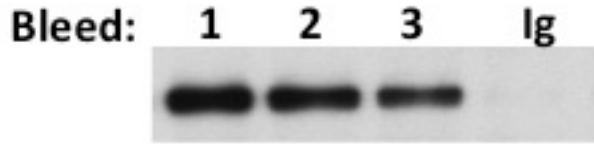
For immunoprecipitation use 10 ug antibody per mg of cell extract. (all three bleeds work equally as well).

Immunoblotting:



HEK293 cells were transfected with a vector encoding HA-SIK1, HA-SIK2, HA-SIK3 or the empty vector (-) and lysed 48 hours post-transfection. Cell lysates (20 ug protein extract) were separated by SDS-Page on an 8 % gel followed by immunoblotting with 1 ug/ml anti-SIK2 (S567D) and the rat monoclonal antibody (3F10) from Roche.

Immuniprecipitation:



Cells lysates (1 mg protein extract) from mouse RAW264.7 macrophages were incubated with 10 ug of purified antibody from bleeds 1, 2 or 3 of sheep anti-SIK2 (S567D) or control immunoglobulins from non-immunised sheep for 90 mins at 4 deg C. Immune complexes were recovered by adding 10 ul of protein G-Sepharose and incubating the sample for a further 30 mins at 4 deg C. The immunoprecipitates were washed with lysis buffer and proteins bound to the column were eluted with 1 % SDS, separated by SDS-Page (8 % gel) and immunoblotted using the anti-SIK2 antibody from Cell Signalling Technology.