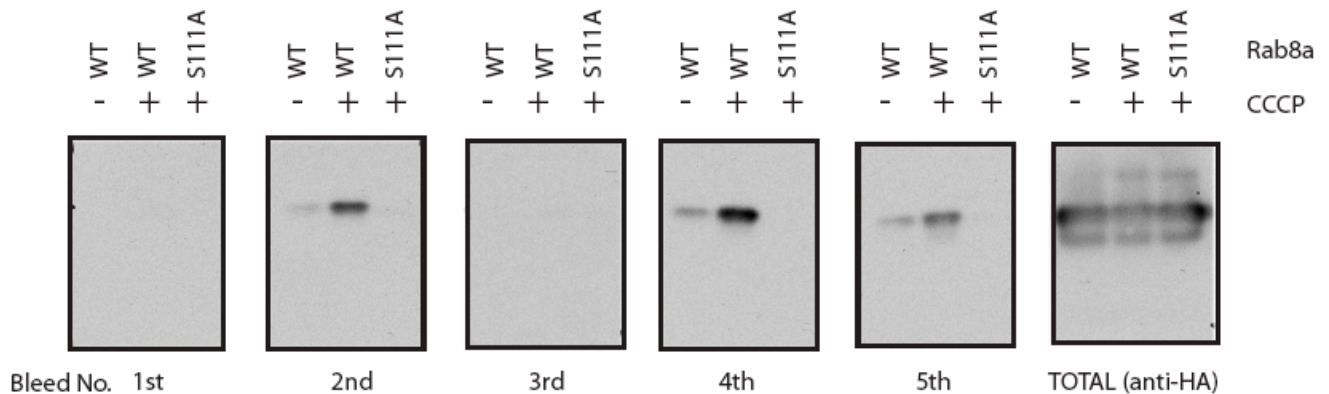


Antibody Datasheet

Product description:- Anti-Rab8a phospho Ser 111
Antigen:- RNIEEHAS*ADVEKMR [residues 104 - 117 of human]
Sheep Number:- S504D
Formulation:- Phosphate Buffered Saline
Storage temperature:- -20 °C
Purification Method:- Affinity purified against phospho specific peptide
Working Concentration:-
 For immunoblotting use at 0.2 ug/ml plus 2 ug/ml of non-phospho peptide
 Please note that Rab8a needs to be immunoprecipitated first before immunoblotting.

Immunoblotting:



HA-Rab8a was transiently transfected into Flp In HEK293 stable cell lines expressing PINK1-Flag tagged Wildtype (WT). Cells were induced for PINK1 expression with doxycyclin for 24 hrs and stimulated with and without CCCP (mitochondrial depolarizing agent) for 3 hours. HA-Rab8a S111A mutant was transfected into PINK1-Flag WT stable cell lines. Cells were induced for PINK1 expression with doxycyclin for 24 hours and stimulated with CCCP (mitochondrial depolarizing agent) for 3 hours. 1 mg of whole cell lysate was immunoprecipitated for Rab8a using 10 ul of HA-agarose affinity resin. Proteins were resolved by SDS-PAGE and subjected to western blot analysis. The blot was probed with the bleeds of anti-Rab8a phospho Ser 111 S503D antibody as indicated.

Antibody recognises HA-Rab8a when it is expressed in cells stably expressing PINK1 under conditions of mitochondrial depolarization and not under basal conditions (A faint band is observed under basal conditions which could be unphosphorylated Rab8a being picked up unspecifically). Also the specificity of the antibody is confirmed with loss of recognition of HA-Rab8a S111A mutant.