

# Product Datasheet

## GW182 Antibody - BSA Free

### NBP1-57134

Unit Size: 100 ul

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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**NBP1-57134**

GW182 Antibody - BSA Free

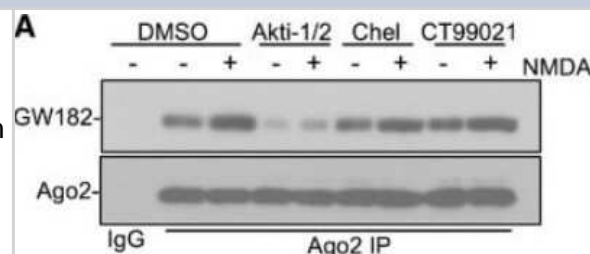
Product Information	
Unit Size	100 ul
Concentration	0.5 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.09% Sodium Azide
Isotype	IgG
Purity	Affinity purified
Buffer	PBS, 2% Sucrose

Product Description	
Description	The addition of 50% glycerol is optional for those storing this antibody at -20C and not aliquoting smaller units. However, please note that glycerol may interrupt some downstream antibody applications and should be added with caution.
Host	Rabbit
Gene ID	27327
Gene Symbol	TNRC6A
Species	Human, Rat
Reactivity Notes	Rat reactivity reported in scientific literature (PMID: 29712715).
Marker	P/GW Body Marker
Immunogen	Synthetic peptides corresponding to GW182 (trinucleotide repeat containing 6A) The peptide sequence was selected from the N terminal of GW182. Peptide sequence RELEAKATKDVERNLSRDLVQEEELMEEKKKKKDDKKKKKEAAQKKATEQ. The peptide sequence for this immunogen was taken from within the described region.

Product Application Details	
Applications	Western Blot
Recommended Dilutions	Western Blot 1.0 ug/ml

**Images**

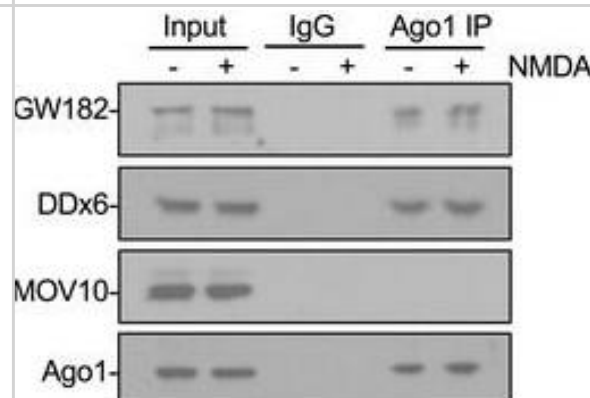
Western Blot: GW182 Antibody [NBP1-57134] - NMDAR-dependent increase in Ago2-GW182 interaction and Ago2 phosphorylation at S387 requires Akt activity. NMDAR-stimulated increase in Ago2-GW182 interaction is Akt-dependent. Cortical neuronal cultures were treated with Akti-1/2 (Akt inhibitor), chelerythrine (Chel, PKC inhibitor) or CT99021 (GSK-3beta inhibitor) 20 min before NMDA or vehicle application. Lysates were prepared 10 min after NMDA washout and immunoprecipitated with Ago2 antibodies or control IgG as shown. Proteins were detected by Western blotting. Image collected and cropped by CiteAb from the following publication ([www.onlinelibrary.wiley.com/doi/10.15252/embj.201797943](http://www.onlinelibrary.wiley.com/doi/10.15252/embj.201797943)) licensed under a CC-BY license.



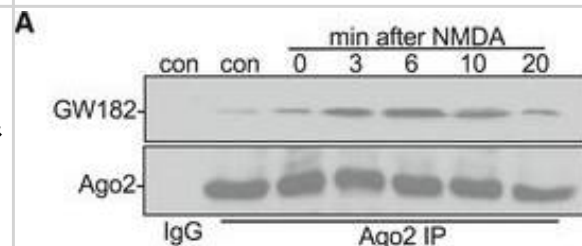
Western Blot: GW182 Antibody [NBP1-57134] - MCF-7 whole cell lysates, concentration 0.2-1 ug/ml.



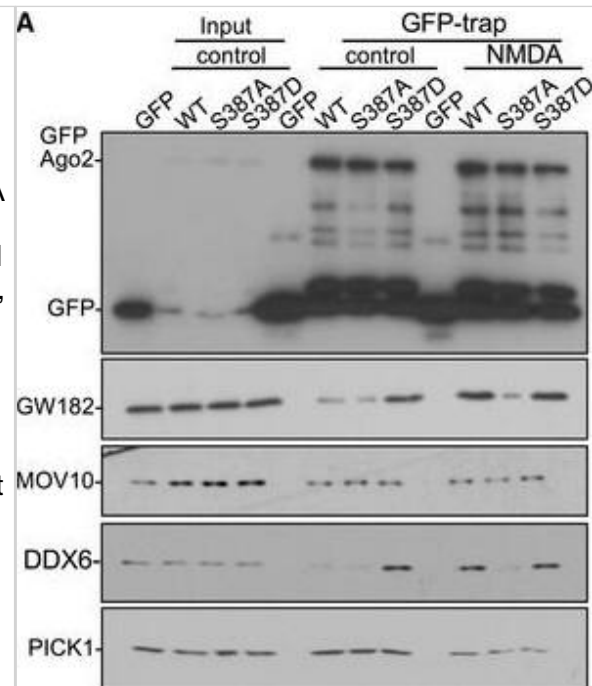
Western Blot: GW182 Antibody [NBP1-57134] - Endogenous Ago1□GW182 & Ago1□DDX6 interactions are unaffected by NMDAR stimulation (related to Fig 1) Cortical neuronal cultures were exposed to NMDA or vehicle for 3 min; lysates were prepared 10 min after NMDA washout & immunoprecipitated with Ago1 antibodies. Proteins were detected by Western blotting. Graph shows quantification of Ago1□GW182 interaction, normalised to vehicle control; n = 5; t□test. Source data are available online for this figure. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29712715>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



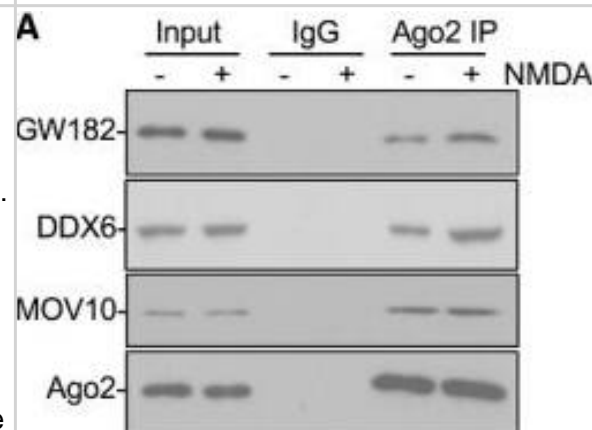
Western Blot: GW182 Antibody [NBP1-57134] - Transient increase in GW182□Ago2 interaction & S387 phosphorylation in response to NMDAR stimulation Transient increase in Ago2□GW182 interaction. Cortical neuronal cultures were exposed to NMDA or vehicle for 3 min, & lysates were prepared 0, 3, 6, 10, 20 min after NMDA washout & immunoprecipitated with Ago2 antibodies or control IgG. Proteins were detected by Western blotting. The inputs are shown in (B). Graph shows quantification of Ago2□GW182 interaction, normalised to vehicle control; n = 4. \*\*P < 0.01, \*\*\*P < 0.001; one□way ANOVA, Bonferroni post hoc test. Mean ± SEM. Transient increase in S387 phosphorylation & Akt activation. The same lysates from (A) (1% of input) were analysed by Western blotting using antibodies against pS387 Ago2, Ago2, pS473 Akt, Akt, GW182 & GAPDH as a loading control. Graphs show quantification of pS387 Ago2 levels normalised to total Ago2 (top) & pS473 Akt normalised to total Akt (bottom); n = 4. \*P < 0.05; two□way ANOVA, Bonferroni post hoc test. Mean ± SEM. Source data are available online for this figure. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29712715>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



**Western Blot: GW182 Antibody [NBP1-57134] - NMDA-induced increase in interaction with GW182 is caused by Ago2 phosphorylation at S387**  
 Ago2 S387A mutation blocks, & S387D mutation occludes NMDA-induced increases in GW182 & DDX6 interactions. Cortical neurons were transfected with molecular replacement constructs expressing Ago2 shRNA plus shRNA-resistant GFP-Ago2 (WT, S387A or S387D). Lysates were prepared 10 min after NMDA washout, & GFP-Ago2 complexes were precipitated using GFP-trap beads. Bound proteins were detected by Western blotting using GFP, GW182, MOV10, DDX6 or PICK1 antibodies as shown. Graphs show quantification of GFP-Ago2 interactions, normalised to untreated WT condition;  $n = 5$ . \* $P < 0.05$ , \*\* $P < 0.01$ ; two-way ANOVA, Bonferroni post hoc test. Mean  $\pm$  SEM. Ago2 S387A mutation blocks, & S387D mutation occludes NMDA-induced increase in GW182 co-localisation in neuronal dendrites. Cortical neurons were transfected with molecular replacement constructs expressing Ago2 shRNA plus shRNA-resistant GFP-Ago2 (WT, S387A or S387D), fixed 10 min after NMDA washout, permeabilised & stained with GW182 & GFP antibodies. Graph shows Pearson's co-localisation coefficients;  $n = 4$  independent experiments (11 cells per condition). \* $P < 0.05$ , \*\* $P < 0.01$ ; two-way ANOVA, Bonferroni post hoc test. Scale bar = 10  $\mu$ m. Mean  $\pm$  SEM. Source data are available online for this figure. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29712715>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



**Western Blot: GW182 Antibody [NBP1-57134] - Ago2 association with GW182 in neuronal dendrites increases in response to NMDAR stimulation**  
 Endogenous Ago2-GW182 & Ago2-DDX6 interactions increase in response to NMDAR stimulation. Cortical neuronal cultures were exposed to NMDA or vehicle for 3 min, & lysates were prepared 10 min after NMDA washout & immunoprecipitated with Ago2 antibodies. Proteins were detected by Western blotting. Graph shows quantification of Ago2-GW182 interaction, normalised to vehicle control;  $n = 5$ . \* $P < 0.05$ ; \*\*\* $P < 0.001$ ;  $t$ -test; mean  $\pm$  SEM. Analysis of endogenous Ago2-GW182 co-localisation in cortical neuronal cultures. Cortical neuronal cultures were exposed to NMDA or vehicle for 3 min, fixed 10 min after NMDA washout, permeabilised & co-stained with Ago2 & GW182 antibodies. Representative whole-cell images are shown. Scale bar = 50  $\mu$ m. Endogenous GW182-Ago2 co-localisation increases in response to NMDAR stimulation in neuronal dendrites. Images show dendrites taken from boxed region in (B), above. Graph shows Pearson's co-localisation coefficients;  $n = 4$  independent experiments (18–24 cells per condition). \* $P < 0.05$ ,  $t$ -test. Scale bar = 10  $\mu$ m. Mean  $\pm$  SEM. Line-scan analyses of Ago2 & GW182 fluorescence intensities in control & NMDA-stimulated dendrites shown in (C). NMDAR stimulation has no effect on endogenous Ago2-GW182 co-localisation in neuronal cell bodies. Images show cell bodies taken from boxed region in (B). Graph shows Pearson's co-localisation coefficients;  $n = 4$  independent experiments (18–20 cells per condition),  $t$ -test. Scale bar = 10  $\mu$ m. Mean  $\pm$  SEM. Source data are available online for this figure. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/29712715>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



## Publications

Seth P, Hsieh PN, Jamal S et al. Regulation of MicroRNA Machinery and Development by Interspecies S-Nitrosylation Cell 2019-02-21 [PMID: 30794773] (WB, Human)

Rajgor D, Sanderson TM, Amici M et al. NMDAR-dependent Argonaute 2 phosphorylation regulates miRNA activity and dendritic spine plasticity EMBO J. 2018-06-01 [PMID: 29712715] (WB, Rat)





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### **Products Related to NBP1-57134**

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HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP]
NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control
NBP1-88231PEP	GW182 Recombinant Protein Antigen

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### **Limitations**

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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