

# Product Datasheet

## ASXL1 Antibody (6E2) - Azide and BSA Free H00171023-M05

Unit Size: 0.1 mg

Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.

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**H00171023-M05**

ASXL1 Antibody (6E2) - Azide and BSA Free

**Product Information**

|                      |                                                                                                                  |
|----------------------|------------------------------------------------------------------------------------------------------------------|
| <b>Unit Size</b>     | 0.1 mg                                                                                                           |
| <b>Concentration</b> | Concentrations vary lot to lot. See vial label for concentration. If unlisted please contact technical services. |
| <b>Storage</b>       | Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.                                                     |
| <b>Clonality</b>     | Monoclonal                                                                                                       |
| <b>Clone</b>         | 6E2                                                                                                              |
| <b>Preservative</b>  | No Preservative                                                                                                  |
| <b>Isotype</b>       | IgG2a Kappa                                                                                                      |
| <b>Purity</b>        | IgG purified                                                                                                     |
| <b>Buffer</b>        | In 1x PBS, pH 7.4                                                                                                |

**Product Description**

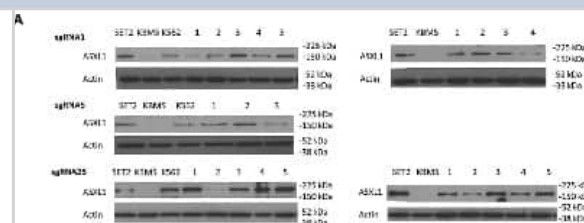
|                    |                                                                                                                                                                                                                                                                     |
|--------------------|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| <b>Description</b> | Novus Biologicals Mouse ASXL1 Antibody (6E2) - Azide and BSA Free (H00171023-M05) is a monoclonal antibody validated for use in WB, ELISA and IP. Anti-ASXL1 Antibody: Cited in 3 publications. All Novus Biologicals antibodies are covered by our 100% guarantee. |
| <b>Host</b>        | Mouse                                                                                                                                                                                                                                                               |
| <b>Gene ID</b>     | 171023                                                                                                                                                                                                                                                              |
| <b>Gene Symbol</b> | ASXL1                                                                                                                                                                                                                                                               |
| <b>Species</b>     | Human                                                                                                                                                                                                                                                               |
| <b>Immunogen</b>   | ASXL1 (AAH64984.1, 1 a.a. ~ 84 a.a) full-length recombinant protein with GST tag. MW of the GST tag alone is 26 KDa.<br>MKDKQKKKKERTWAEAAARLVLENYS DAPMTPKQILQVIEAEGLKEMSGTSPLA<br>CLNAMLHSNSRGGEGFLFYKLPGRISLFTLKR                                                 |
| <b>Notes</b>       | This product is produced by and distributed for Abnova, a company based in Taiwan.                                                                                                                                                                                  |

**Product Application Details**

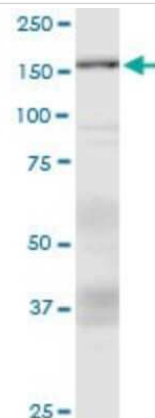
|                              |                                                                                                                                 |
|------------------------------|---------------------------------------------------------------------------------------------------------------------------------|
| <b>Applications</b>          | Western Blot, ELISA, Immunoprecipitation                                                                                        |
| <b>Recommended Dilutions</b> | Western Blot 1:100-1:2000, ELISA 1:100-1:2000, Immunoprecipitation                                                              |
| <b>Application Notes</b>     | This antibody is useful for ELISA, Western Blot. Use in immunoprecipitation reported in scientific literature (PMID: 24894717). |

**Images**

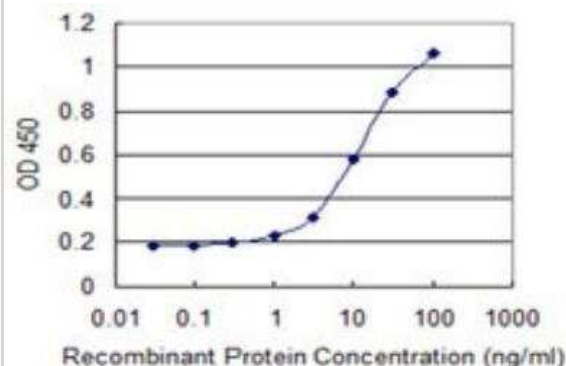
Western Blot: ASXL1 Antibody (6E2) [H00171023-M05] - Functional effects of CRISPR/Cas9-mediated ASXL1 mutation correction. Evaluation of ASXL1 protein expression by Western blotting. Left-hand side: ASXL1 protein expression in the SET2 leukemia cell line (wild-type for ASXL1), uncorrected KBM5 cells, the K562 leukemia cell line (carrying the Y591X heterozygous ASXL1 mutation), and KBM5 clones (labeled 1-5) with heterozygous precise correction of the ASXL1 mutation. Right-hand side: ASXL1 protein expression in the SET2 leukemia cell line (wild-type for ASXL1), uncorrected KBM5 cells, and KBM5 clones (labeled 1-5) with homozygous precise correction of the ASXL1 mutation. Beta-actin was used as loading control. Image collected and cropped by CiteAb from the following publication (<http://www.oncotarget.com/fulltext/6392>) licensed under a CC-BY license.



Western Blot: ASXL1 Antibody (6E2) [H00171023-M05] - Analysis of ASXL1 expression in MCF-7 (Cat # L046V1).



ELISA: ASXL1 Antibody (6E2) [H00171023-M05] - Detection limit for recombinant GST tagged ASXL1 is 0.3 ng/ml as a capture antibody.



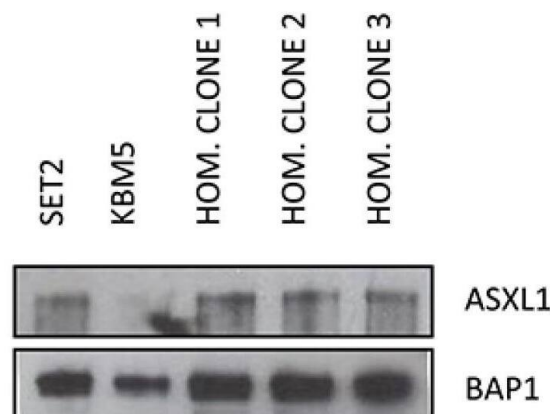
Western Blot: ASXL1 Antibody (6E2) [H00171023-M05] - Functional effects of CRISPR/Cas9-mediated ASXL1 mutation correction (A) Evaluation of ASXL1 protein expression by WB. Left: ASXL1 protein expression in SET2 leukemia cell line (wild-type for ASXL1), uncorrected KBM5 cells, the K562 leukemia cell line (carrying the Y591X heterozygous ASXL1 mutation), & KBM5 clones (labeled 1–5) w/ heterozygous precise correction of ASXL1 mutation. Right: ASXL1 protein expression in SET2 leukemia cell line (wild-type for ASXL1), uncorrected KBM5 cells, & KBM5 clones (labeled 1–5) w/ homozygous precise correction of ASXL1 mutation.  $\beta$ -actin used as loading control.

(B) Evaluation of expression levels of HOXA genes using quantitative real-time PCR (q-RT-PCR). Expression levels of HOXA5, 6, 7, 9, 10 & 13 measured in three ASXL1 homozygous corrected KBM5 clones compared w/ uncorrected cells. Results shown obtained from six independent experiments for each clone. Values in ASXL1 homozygous corrected cells are relative to uncorrected cells. Bar graphs show mean + standard error of mean (s.e.m.) (\* =  $P < 0.05$ , \*\* =  $P < 0.01$ , \*\*\* =  $P < 0.001$ , paired t-test).

(C) Evaluation of H3K27me3 levels & expression of PRC2 components by WB in uncorrected KBM5 cells & three KBM5 clones w/ homozygous correction of ASXL1 mutation. H3K27me3 levels & total H3 levels evaluated using purified histone fractions. Expression levels of two PRC2 components (EZH2, SUZ12) determined using whole cell lysates.  $\beta$ -actin used as loading control. (D) Immunoprecipitation of BAP1 in SET2 leukemia cell line (wild-type for ASXL1), uncorrected KBM5 cells, & three KBM5 clones w/ homozygous correction of ASXL1 mutation. The BAP1 protein fraction immunoprecipitated using a BAP1 antibody & stained for ASXL1 & BAP1. Image collected & cropped by CiteAb from the following publication

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**D**



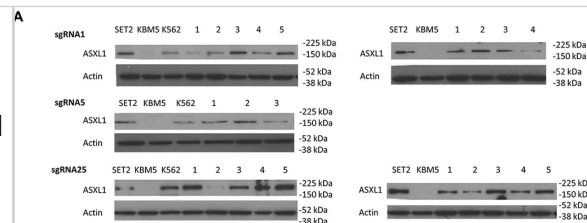
**Western Blot: ASXL1 Antibody (6E2) [H00171023-M05] - Functional effects of CRISPR/Cas9-mediated ASXL1 mutation correction(A)**

**Evaluation of ASXL1 protein expression by WB.** Left: ASXL1 protein expression in SET2 leukemia cell line (wild-type for ASXL1), uncorrected KBM5 cells, the K562 leukemia cell line (carrying the Y591X heterozygous ASXL1 mutation), & KBM5 clones (labeled 1–5) w/ heterozygous precise correction of ASXL1 mutation. Right: ASXL1 protein expression in SET2 leukemia cell line (wild-type for ASXL1), uncorrected KBM5 cells, & KBM5 clones (labeled 1–5) w/ homozygous precise correction of ASXL1 mutation.  $\beta$ -actin used as loading control.

**(B) Evaluation of expression levels of HOXA genes using quantitative real-time PCR (q-RT-PCR).** Expression levels of HOXA5, 6, 7, 9, 10 & 13 measured in three ASXL1 homozygous corrected KBM5 clones compared w/ uncorrected cells. Results shown obtained from six independent experiments for each clone. Values in ASXL1 homozygous corrected cells are relative to uncorrected cells. Bar graphs show mean + standard error of mean (s.e.m.) (\* =  $P < 0.05$ , \*\* =  $P < 0.01$ , \*\*\* =  $P < 0.001$ , paired t-test).

**(C) Evaluation of H3K27me3 levels & expression of PRC2 components by WB** in uncorrected KBM5 cells & three KBM5 clones w/ homozygous correction of ASXL1 mutation. H3K27me3 levels & total H3 levels evaluated using purified histone fractions. Expression levels of two PRC2 components (EZH2, SUZ12) determined using whole cell lysates.  $\beta$ -actin used as loading control.

**(D) Immunoprecipitation of BAP1** in SET2 leukemia cell line (wild-type for ASXL1), uncorrected KBM5 cells, & three KBM5 clones w/ homozygous correction of ASXL1 mutation. The BAP1 protein fraction immunoprecipitated using a BAP1 antibody & stained for ASXL1 & BAP1. Image collected & cropped by CiteAb from the following publication (<https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.6392>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



## Publications

Valletta S, Dolatshad H, Bartenstein M et al. ASXL1 mutation correction by CRISPR/Cas9 restores gene function in leukemia cells and increases survival in mouse xenografts. *Oncotarget* 2015-12-29 [PMID: 26623729] (WB)

Davies C, Yip BH, Fernandez-Mercado M et al. Silencing of ASXL1 impairs the granulomonocytic lineage potential of human CD34(+) progenitor cells. *Br J Haematol.* 2013-01-08 [PMID: 23294243]

Ismail IH, Davidson R, Gagne JP et al. Germ-line Mutations in BAP1 Impair its Function in DNA Double-Strand break Repair. *Cancer Res.* 2014-06-03 [PMID: 24894717] (IP, WB, Human)

### Details:

U2OS cells were transfected with control/ASXL1 shRNA for 48 h and the nuclear extracts were processed for WB using ASXL1 and BAP1 antibodies. In another set of experiment, U2OS cells were exposed or not to radiation (6 Gy) followed by recovery for 2 hr and the nuclear extracts were processed for immunoprecipitation using BAP1 or ASXL1 antibodies followed by WB of flow through (FT) and elute (EI) and 20% of the input (Supplementary Figure 2 C and D).



### **Novus Biologicals USA**

10730 E. Briarwood Avenue  
Centennial, CO 80112  
USA  
Phone: 303.730.1950  
Toll Free: 1.888.506.6887  
Fax: 303.730.1966  
nb-customerservice@bio-techne.com

### **Bio-Techne Canada**

21 Canmotor Ave  
Toronto, ON M8Z 4E6  
Canada  
Phone: 905.827.6400  
Toll Free: 855.668.8722  
Fax: 905.827.6402  
canada.inquires@bio-techne.com

### **Bio-Techne Ltd**

19 Barton Lane  
Abingdon Science Park  
Abingdon, OX14 3NB, United Kingdom  
Phone: (44) (0) 1235 529449  
Free Phone: 0800 37 34 15  
Fax: (44) (0) 1235 533420  
info.EMEA@bio-techne.com

### **General Contact Information**

[www.novusbio.com](http://www.novusbio.com)  
Technical Support: nb-technical@bio-techne.com  
Orders: nb-customerservice@bio-techne.com  
General: novus@novusbio.com

### **Products Related to H00171023-M05**

|                  |                                                    |
|------------------|----------------------------------------------------|
| NBP2-33376H      | Blue Marker Antibody (6F4-F6) [HRP]                |
| HAF007           | Goat anti-Mouse IgG Secondary Antibody [HRP]       |
| NB7539           | Goat anti-Mouse IgG (H+L) Secondary Antibody [HRP] |
| NBP1-96981-0.5mg | Mouse IgG2a Kappa Isotype Control (M2AK)           |

### **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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