

Product no **AS16 3829****RING1 | E3 ubiquitin-protein ligase RING1****Product information**

<b>Immunogen</b>	Recombinant RING1 of <i>Drosophila melanogaster</i> , amino acids: 150-250, UniProt: <a href="#">Q9VB08</a>
<b>Host</b>	Rabbit
<b>Clonality</b>	Polyclonal
<b>Purity</b>	Immunogen affinity purified serum in PBS pH 7.4.
<b>Format</b>	Lyophilized
<b>Quantity</b>	50 µg
<b>Reconstitution</b>	For reconstitution add 50 µl, of sterile water
<b>Storage</b>	Store lyophilized/reconstituted at -20°C; once reconstituted make aliquots to avoid repeated freeze-thaw cycles. Please remember to spin the tubes briefly prior to opening them to avoid any losses that might occur from material adhering to the cap or sides of the tube.

**Application information**

<b>Recommended dilution</b>	3 µg of antibody (ChIP), 1 : 2000 (WB)
<b>Expected   apparent MW</b>	47   58 kDa
<b>Confirmed reactivity</b>	<i>Drosophila melanogaster</i>
<b>Not reactive in</b>	No confirmed exceptions from predicted reactivity are currently known
<b>Selected references</b>	To be added when available, antibody released in November 2020.

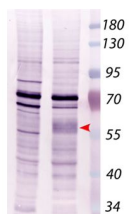
**Western Blot (WB)**

Figure 1. Western Blot (WB) result.

20 µg of total protein from Psc/Su(z)2-KO cells (Kahn et al., 2016. doi: 10.1093/nar/gkw701 , line1) and Ras3 (wild type, line2) cells lysed with 1x SDS page loading buffer were separated on 12% SDS-PAGE and blotted 2h to PVDF using tank transfer. Blot was dried and incubated in the primary antibody at a dilution of 1 : 2000 for 1h at RT with agitation. The antibody solution was decanted and the blot was rinsed briefly twice in 1x PBS. Blot was incubated in secondary antibody (anti-rabbit IgG AP conjugated, from Promega) diluted to 1:10 000 in for 30 minutes at RT with agitation. The blot was washed as above and developed with NBT/BCIP solution (SIGMA).

Courtesy of Dr. Alexander Glotov. , Umeå University, Sweden

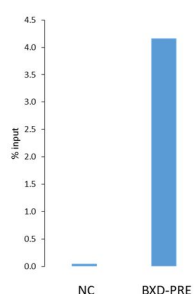
**Chromatin Ummunoprecipitation (ChIP)**

Figure 2. ChIP recovery.

ChIP and qPCR analysis were done as described [Schwartz YB, Kahn TG, Nix DA, Li XY, Bourgon R, et al. (2006) Genome-wide analysis of Polycomb targets in *Drosophila melanogaster*. Nat Genet 38: 700–705. doi: 10.1038/ng1817]. Chromatin from Ras3 cells was used for ChIP. Quantitative PCR was performed with primers specific for BXD-PRE of Ubx gene (Polycomb target gene in repressed state), used as positive controls, and for intergenic region, used as negative control (NC). Figure 2 shows the ChIP recovery measured by qPCR as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA). Chromatin from 5x10<sup>7</sup> cells and 3 µg of anti-RING1 antibody were used for ChIP reaction.

Courtesy of Dr. Tatyana Khan, Umeå University, Sweden.