## Broad Institute - Encode3 Secondary Antibody Validation

# Cell Signaling 5246S lot # 6

Target: EZH2

Approved name: enhancer of zeste 2 polycomb repressive complex subunit 2

<u>Function</u>: EZH2 is the catalytic subunit of the polycomb related complex PRC2. It functions as the methyl transferase which writes the methyl mark on Histone H3 Lysine 27. Consequently, there is a highly correlated pattern in the genomewide distribution of H3K27me3 and the phosphorylated derivative of EZH2, or phopho-EZH2, the protein form recognized by this antibody.

<u>Member of complex</u>: EZH2 is a component of the PRC2 complex, which is a chromatin modifier that aids in the maintenance of transcriptional silencing.

Other complex members: EED, EZH1, RbBP4, RbBP7, SUZ12

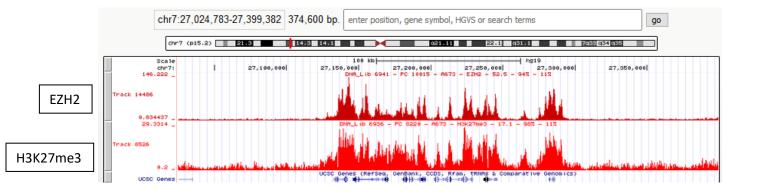
#### Reference:

1. Genes Dev. 2002. **16**:2893-2905. Histone methyltransferase activity associated with a human multiprotein complex containing the Enhancer of Zeste protein. Kuzmichev A, Nishioka K, Erdjument-Bromage H, Tempst P, Reinberg D. <a href="PMID:12435631">PMID:12435631</a>.

### Antibody being validated:

- 1. Cell Signaling Technology 5246S Lot: 6 [Rabbit monoclonal]
- 2. Broad Aliases: PchAb 1206 and PchAb 1290
- 3. Immunogen: a synthetic peptide corresponding to residues surrounding Arg354 of human Ezh2 protein.
- 4. https://www.encodeproject.org/antibodies/ENCAB913HCF/

This validation relies on the use of antibodies to a chromatin regulator and a functionally related histone modification in A673 cells, and the demonstration that highly similar patterns of enrichment are obtained with each antibody. The first track shown used an antibody to phosphorylated EZH2 (PchAb 1206, ENCAB913HCF), and the second track shown used an antibody to H3K27me3 previously characterized to Encode standards (PchAb 848, ENCAB036YAO).



## Antibody being compared:

- 1. Millipore 07-449 Lot: 2120130 [Rabbit polyclonal, Protein A purification]
- 2. Broad Alias: PchAb 848
- 3. Immunogen: KLH-conjugated, synthetic 2X-branched peptide containing the sequence ...AR(me3K)SAP... in which me3K corresponds to trimethyl-lysine at residue 27 of human histone H3.
- 4. https://www.encodeproject.org/antibodies/ENCAB036YAO/