

ENCODE DCC Antibody Validation Document

Date of Submission

Name:

Email:

Lab

Antibody Name:

Target:

Company/
Source:

Catalog Number, database ID, laboratory

Lot Number

Antibody
Description:

Target
Description:

Species Target

Species Host

Validation Method #1

Validation Method #2

Purification
Method

Polyclonal/
Monoclonal

Vendor URL:

Reference (PI/
Publication
Information)

Please complete the following for antibodies to histone modifications:
*if your specifications are not listed in the drop-down box,
please write-in the appropriate information*

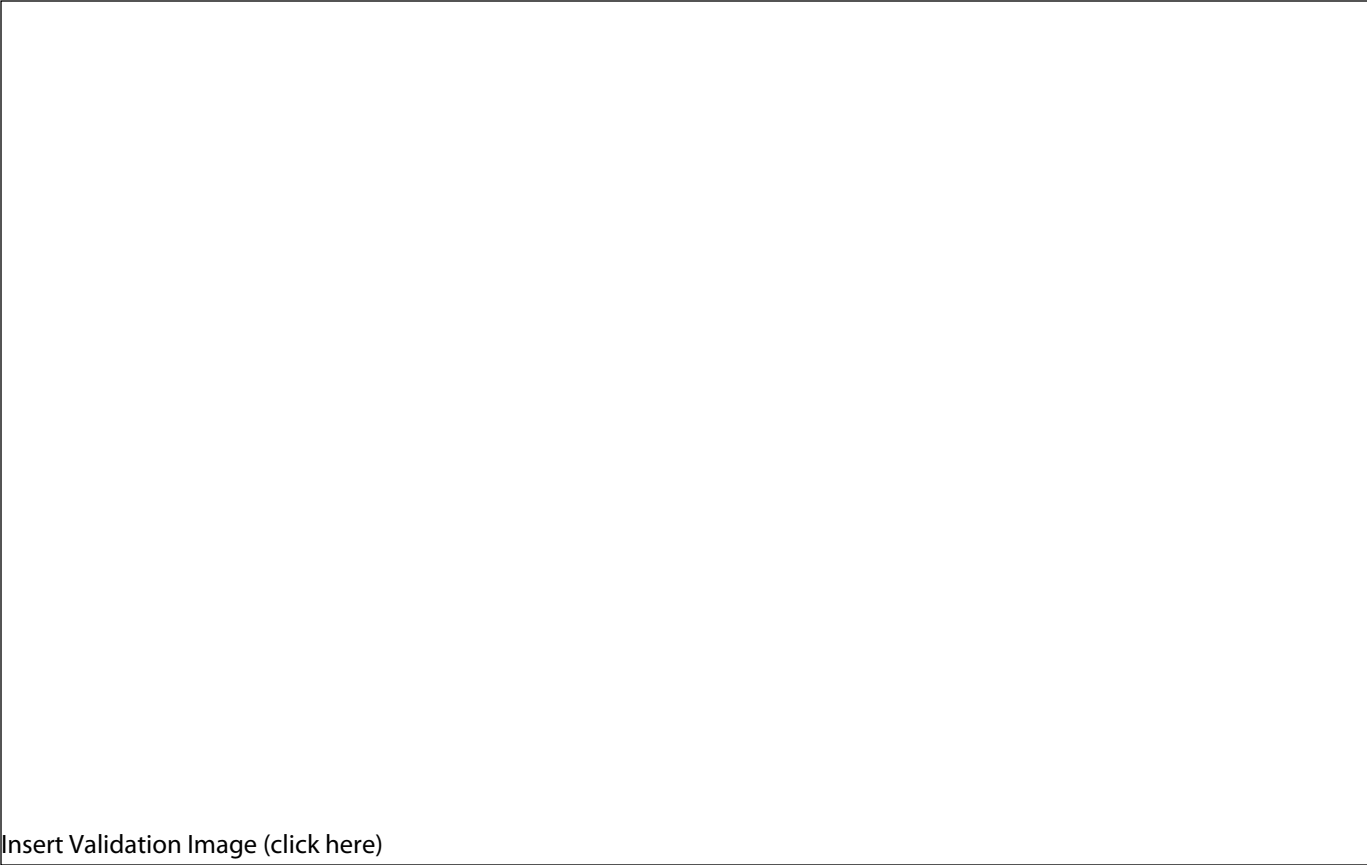
Histone Name

AA modified

AA Position

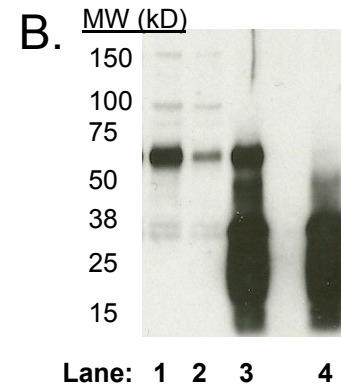
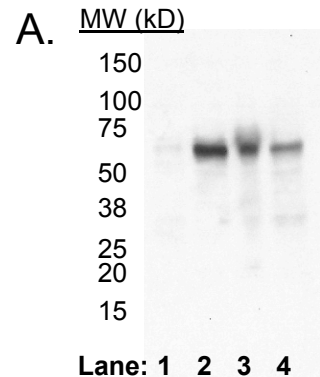
Modification

Validation #1
Analysis



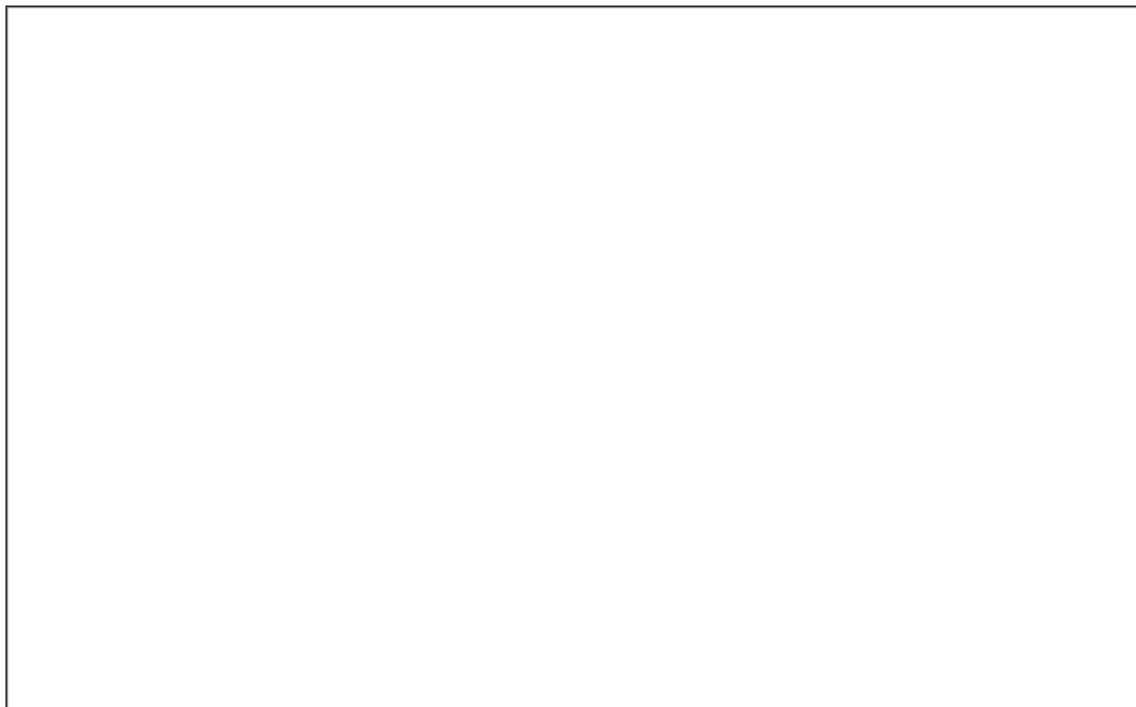
Insert Validation Image (click here)

Antibody sc-30189 (COREST) Immunoblot/immunoprecipitation

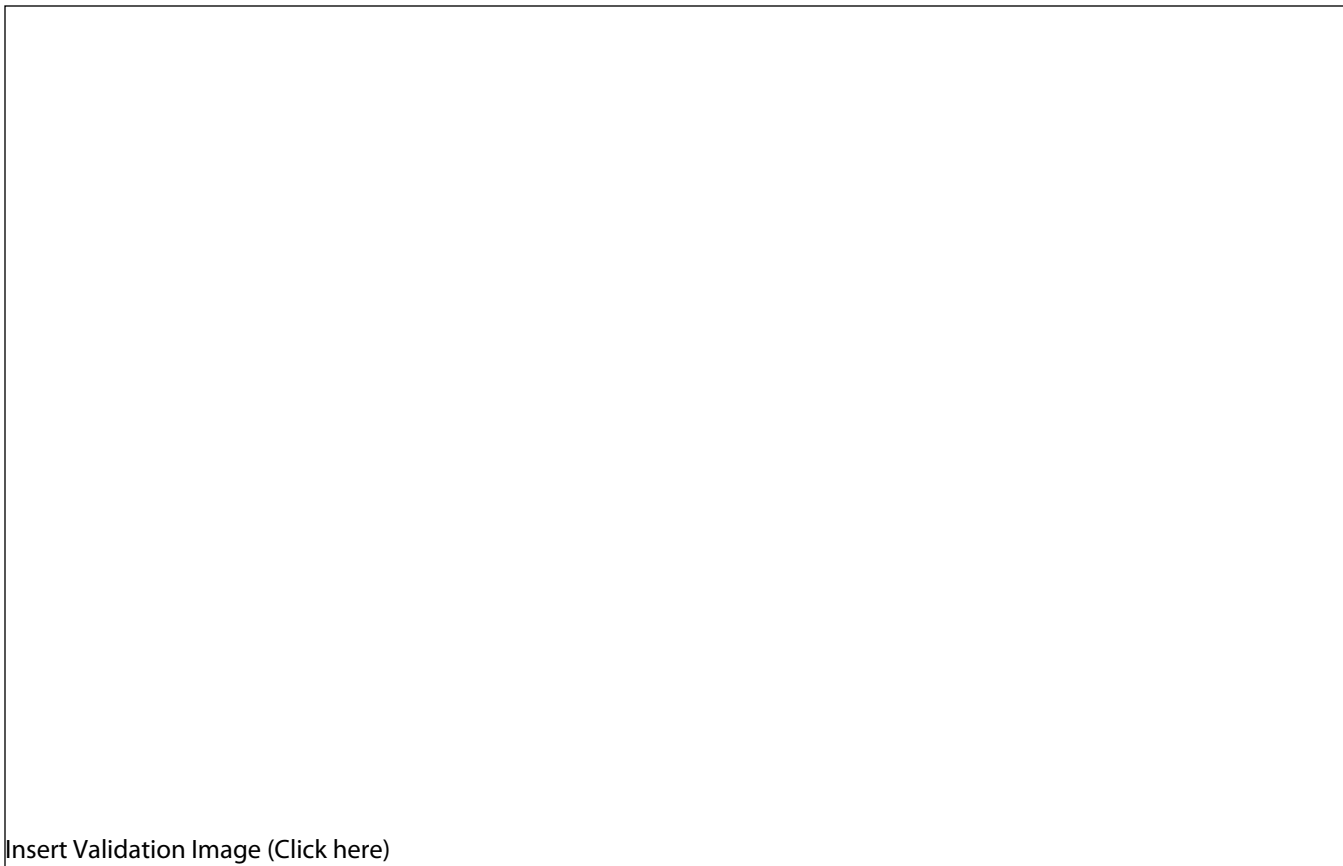


A. Western blot using antibody sc-30189 on nuclear lysates from cell lines GM12878 (Lane1), K562 (Lane2), HeLaS3 (Lane3), and HepG2 (Lane4). **B.** Immunoprecipitation was performed on nuclear lysates from K562 cells using antibody sc-30189. Lane1: Nuclear lysate. Lane 2: Unbound material from immunoprecipitation with sc-30189. Lane 3: Bound material from immunoprecipitation with sc- sc-30189. Lane 4: Bound material from control immunoprecipitation with rabbit IgG. Arrow indicates band of expected size (18kD) that is highly enriched in the specifically immunoprecipitated fraction.

Validation #2
Analysis



Insert Validation Image (Click here)



Validation 2: ChIPseq with alternate antibodies to the same factor

	COREST sc30189	COREST ab24166
Total peaks	111692	30269
% Peak overlap	87.6	93.2

Antibodies:

sc30189: epitope corresponding to amino acids 246-310 within CoREST of human origin

ab24166: immunogen is synthetic peptide conjugated to KLH derived from residues 400 to C-terminus of human COREST.

Comparison: K562 cells were used for ChIP-seq with antibody sc-30189 or antibody ab24166. Peaks were called from replicate experiments using PeakSeq with a .01 q-value cut-off. Comparisons between experiments were made using these peaks according to standard ENCODE replicate comparison parameters (http://genome.ucsc.edu/ENCODE/protocols/dataStandards/ChIP_DNase_FAIRE_DNAme_v2_2011.pdf; reported is the fraction of the top 40% of peaks in one list that are found in the full list of peaks obtained with the other antibody.