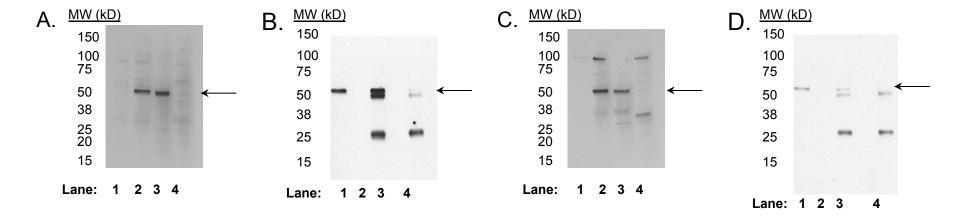
ENCODE DCC Antibody Validation Document

Date of Submission
Name: Email:
Lab
Lab
Antibody Name: Target:
Company/
Source:
Catalog Number, database ID, laboratory Lot Number
Antibody
Description:
Target
Target Description:
Species Toyant Species Hest
Species Target Species Host
Validation Method #1 Validation Method #2
Purification Polyclonal/
Method Monoclonal
Vicilia IIDI
Vendor URL:
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ease complete the following for antibodies to histone modifications:
your specifications are not listed in the drop-down box,
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Validation #1 Analysis					
Insert Validation Image (click here)					

Antibody ab24550 (TBLR1) Immunoblot/ immunoprecipitation

Antibody NB600-270 (TBLR1) Immunoblot/ immunoprecipitation



A. Western blot using antibody ab24550 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 ab24550. Lane1: Nuclear lysate. Lane 2: Unbound material from immunoprecipitation with ab24550. Lane 3: Bound material from immunoprecipitation with ab24550. Lane 4: Bound material from control immunoprecipitation with rabbit IgG. Arrow indicates band of expected size (56kD) that is highly enriched in the specifically immunoprecipitated fraction. **C.** Western blot using antibody NB600-270 on nuclear lysates from cell lines GM12878 (Lane1), K562 (Lane2), HeLaS3 (Lane3), and HepG2 (Lane4). **D.** Immunoprecipitation was performed on nuclear lysates from K562 cells using antibody NB600-270. Lane1: Nuclear lysate. Lane 2: Unbound material from immunoprecipitation with NB600-270. Lane 4: Bound material from control immunoprecipitation with rabbit IgG. Arrow indicates band of expected size (56kD) that is highly enriched in the specifically immunoprecipitated fraction.

Validation #2 Analysis					
nsert Validation Image (Click here)					
isere variation image (click fiele)					

Validation 2: ChIPseq with alternate antibodies to the same factor

	TBLR1 NB600-270	TBLR1 ab24550
Total peaks	65216	38496
% Peak overlap	82.4	85.8

Antibodies/Immunogens:

NB600-270: Peptide that maps to a region between residues 100 and 150 of human TBL1-related Protein 1 **ab24550**: Recombinant full length protein.

Comparison: K562 cells were used for ChIP-seq with antibody ab24550 or antibody NB600-270. 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.