ENCODE DCC Antibody Validation Document

Date of Submission
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Antibody Name: Target:
Company/
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Catalog Number, database ID, laboratory Lot Number
Antibody
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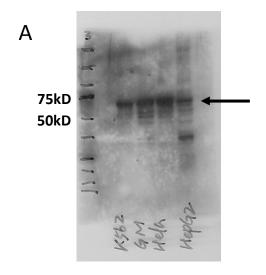
Validation #1 Analysis			
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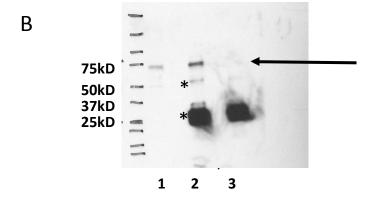
Antibody: RFX5 Source: Rockland #200-401-194

Figure 1: Immunoblotting/Immunoprecipitation

Arrow indicates band detected by Rockland antibody 200-401-194 consistent with expected size (65kD) of RFX5 in nuclear lysates from (left to right): K562, GM12878, HeLaS3, and HepG2 cell lines.

Arrow indicates immunoprecipitated (IP) band of expected size from HeLaS3 nuclear lysates. Lane 1= HeLaS3 nuclear lysate, Lane 2= bound material from HeLaS3 IP with 200-401-194, Lane 3= bound material from control IgG IP from HeLaS3. IPs were performed with the ExactaCruz reagent (Santa Cruz Biotechnology), so the intensities of IgG bands (indicated by *), particularly the heavy chain at 55kD, are diminished.



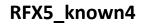


Validation #2 Analysis	
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Table 1.	RFX5	motif	enrichment.
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Cell Line	Motif Enrichment	Enrichment p-value	Fraction of peaks
	(log2)	(-log10)	containing motif
GM12878	2.64148	203.846	0.195876
H1-hESC	3.36223	237.855	0.378431
HepG2	2.96118	415.584	0.239672

Figure 2. Motif consensus sequences and position weight matrices for highly enriched RFX5 motif.





>RFX5_known4 Rfx_5#known Rfx4_bulyk_sc09primary +

Position/ Consensus	<u>A</u>	<u>C</u>	<u>G</u>	I
В	0.143522	0.316811	0.206641	0.333027
H	0.363838	0.275175	0.143142	0.217845
V	0.167173	0.561442	0.177495	0.093890
C	0.002395	0.959751	0.032345	0.005509
H	0.400559	0.250652	0.120082	0.228707
T	0.002331	0.011423	0.004255	0.981991
R	0.774005	0.006875	0.218576	0.000544
G	0.042920	0.001813	0.953809	0.001458
C	0.005434	0.952584	0.001106	0.040876
A	0.957141	0.000549	0.031944	0.010366
A	0.990768	0.002859	0.005499	0.000874
C	0.002052	0.990864	0.001010	0.006074
S	0.114722	0.349363	0.464141	0.071774
V	0.280483	0.181532	0.373684	0.164300
N	0.282246	0.183429	0.218756	0.315568