

# 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*

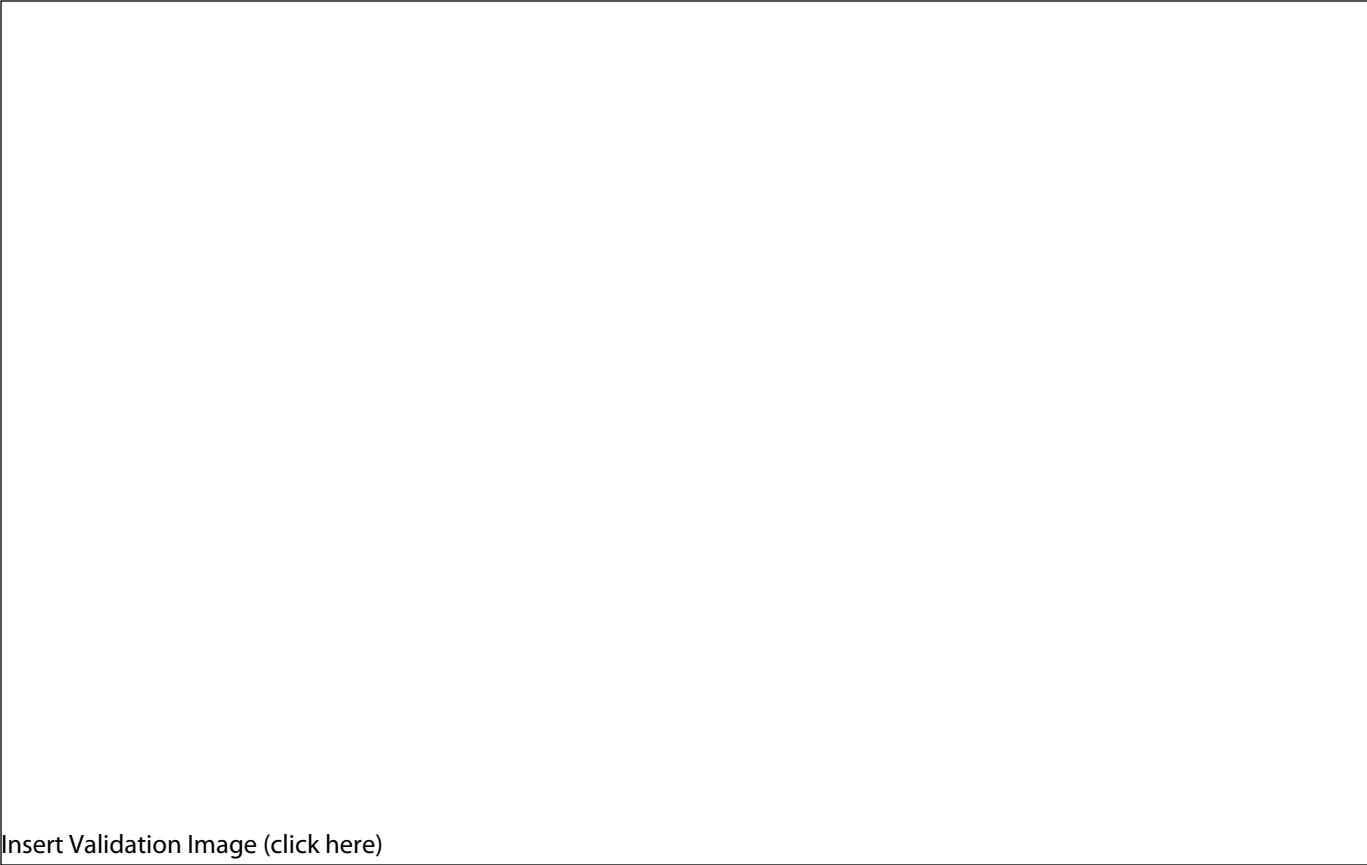
Histone Name

AA modified

AA Position

Modification

Validation #1  
Analysis



Insert Validation Image (click here)

Validation #2  
Analysis



Insert Validation Image (Click here)

## Validation 2: Mass Spectrometry Analysis

ENCODE data standards recognizes various methodologies for secondary validation of antibodies. Among these methodologies is immunoprecipitation followed by mass spectrometry analysis. Briefly, GM12878 whole cell lysates were immunoprecipitated using primary antibody, and the IP fraction was loaded on a 12% acrylamide gel and separated with a Bio-Rad PROTEAN II xi system. Gel was stained with Coomassie Blue in order to visualize marker bands. A gel fragment corresponding to the band indicated above in the western blot image was excised and sent to the University of Alabama at Birmingham Cancer Center Mass Spectrometry/Proteomics Shared Facility. There the sample was run on an LTQ XL Linear Ion Trap Mass Spectrometer with alternating collision-induced dissociation and electron-transfer dissociation. Peptides were identified using MASCOT (Matrix Science), with probability based matching at  $p < 0.05$ . As per ENCODE data standards, all MASCOT results are listed below, including common contaminants. Target protein is highlighted in bold font.

1. P35908 Keratin, type II cytoskeletal 2 epidermal n=1 Tax=Homo sapiens RepID=K22E\_HUMAN
2. P00558 Phosphoglycerate kinase 1 n=3 Tax=Homininae RepID=PGK1\_HUMAN
3. B4E3A4 cDNA FLJ57283, highly similar to Actin, cytoplasmic 2 n=1 Tax=Homo sapiens RepID=B4E3A4\_HUMAN
4. P04264 Keratin, type II cytoskeletal 1 n=1 Tax=Homo sapiens RepID=K2C1\_HUMAN
5. UPI00017BCE7F keratin, type I cytoskeletal 10 n=1 Tax=Homo sapiens RepID=UPI00017BCE7F
6. Q53GL5 Isocitrate dehydrogenase 2 (NADP+), mitochondrial variant (Fragment) n=1 Tax=Homo sapiens RepID=Q53GL5\_HUMAN
7. A8K3K1 cDNA FLJ78096, highly similar to Homo sapiens actin, alpha, cardiac muscle (ACTC), mRNA n=1 Tax=Homo sapiens RepID=A8K3K1\_HUMAN
8. Q53FG3 Interleukin enhancer binding factor 2 variant (Fragment) n=1 Tax=Homo sapiens RepID=Q53FG3\_HUMAN
9. P35527 Keratin, type I cytoskeletal 9 n=1 Tax=Homo sapiens RepID=K1C9\_HUMAN
10. P14618 Pyruvate kinase isozymes M1/M2 n=2 Tax=Homininae RepID=KPYM\_HUMAN
11. UPI0001AE6926 UPI0001AE6926 related cluster n=1 Tax=Homo sapiens RepID=UPI0001AE6926
12. B0S7Z4 Major histocompatibility complex, class I, C n=2 Tax=Homo sapiens RepID=B0S7Z4\_HUMAN
13. P30501 HLA class I histocompatibility antigen, Cw-2 alpha chain n=1 Tax=Homo sapiens RepID=1C02\_HUMAN
14. C5J3Q8 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=C5J3Q8\_HUMAN
15. Q29954 HLA-C protein (Fragment) n=1 Tax=Homo sapiens RepID=Q29954\_HUMAN
16. A4HQM5 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=A4HQM5\_HUMAN
17. Q9Y295 Developmentally-regulated GTP-binding protein 1 n=7 Tax=Eutheria RepID=DRG1\_HUMAN
18. UPI0001A5EC2B PREDICTED: actin, beta-like 3 n=1 Tax=Homo sapiens RepID=UPI0001A5EC2B
19. P06733 Alpha-enolase n=1 Tax=Homo sapiens RepID=ENOA\_HUMAN
20. A9QUT7 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=A9QUT7\_HUMAN
21. B2RB23 cDNA, FLJ95265, highly similar to Homo sapiens acetyl-Coenzyme A acyltransferase 2 (mitochondrial 3-oxoacyl-Coenzyme A thiolase) (ACAA2), nuclear gene encoding mitochondrial protein, mRNA n=1 Tax=Homo sapiens RepID=B2RB23\_HUMAN
22. D0VFI1 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=D0VFI1\_HUMAN
23. UPI0001AE669E UPI0001AE669E related cluster n=1 Tax=Homo sapiens RepID=UPI0001AE669E
24. C0KTQ5 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=C0KTQ5\_HUMAN
25. Q6S8J3 POTE ankyrin domain family member E n=1 Tax=Homo sapiens RepID=POTEE\_HUMAN
26. P22695 Cytochrome b-c1 complex subunit 2, mitochondrial n=1 Tax=Homo sapiens RepID=QCR2\_HUMAN
27. B0UY16 Major histocompatibility complex, class I, C (Fragment) n=2 Tax=Homo sapiens RepID=B0UY16\_HUMAN
28. Q15019 Septin-2 n=3 Tax=Homininae RepID=SEPT2\_HUMAN
29. C0M144 MHC class I antigen (Fragment) n=2 Tax=Homo sapiens RepID=C0M144\_HUMAN
30. B4DY90 cDNA FLJ56903, highly similar to Tubulin beta-7 chain n=1 Tax=Homo sapiens RepID=B4DY90\_HUMAN
31. UPI0001AE68FD UPI0001AE68FD related cluster n=1 Tax=Homo sapiens RepID=UPI0001AE68FD
32. B3KUZ8 Aspartate aminotransferase n=1 Tax=Homo sapiens RepID=B3KUZ8\_HUMAN
33. P06733-2 Isoform MBP-1 of Alpha-enolase n=1 Tax=Homo sapiens RepID=P06733-2
34. B3KPS3 cDNA FLJ32131 fis, clone PEBLM2000267, highly similar to Tubulin alpha-ubiquitous chain n=1 Tax=Homo sapiens RepID=B3KPS3\_HUMAN
35. Q0IIN1 Keratin 77 n=1 Tax=Homo sapiens RepID=Q0IIN1\_HUMAN
36. Q9BV20 Methylthioribose-1-phosphate isomerase n=1 Tax=Homo sapiens RepID=MTNA\_HUMAN
37. B4DQ51 cDNA FLJ57418, highly similar to Short/branched chain specific acyl-CoA dehydrogenase, mitochondrial (EC 1.3.99.-) n=1 Tax=Homo sapiens RepID=B4DQ51\_HUMAN

38. C51WV5 Trypsinogen n=1 Tax=Sus scrofa RepID=C51WV5\_PIG

39. B4DJ77 cDNA FLJ53047, highly similar to TNF receptor-associated factor 1 n=1 Tax=Homo sapiens RepID=B4DJ77\_HUMAN

40. UPI0001A5EAC9 PREDICTED: similar to cytoskeletal beta actin n=1 Tax=Homo sapiens RepID=UPI0001A5EAC9

41. Q0P6N9 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=Q0P6N9\_HUMAN

42. A6NE09 Putative uncharacterized protein ENSP00000346598 n=1 Tax=Homo sapiens RepID=A6NE09\_HUMAN

43. UPI000179EC85 Serum albumin precursor (Allergen Bos d 6) (BSA). n=1 Tax=Bos taurus RepID=UPI000179EC85

44. B7Z708 cDNA FLJ58698, highly similar to cAMP-dependent protein kinase, alpha-catalytic subunit (EC 2.7.11.11) n=1 Tax=Homo sapiens RepID=B7Z708\_HUMAN

45. Q1M309 HLA-B antigen n=1 Tax=Homo sapiens RepID=Q1M309\_HUMAN

46. D3JW64 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=D3JW64\_HUMAN

47. UPI000173A15C UPI000173A15C related cluster n=1 Tax=Homo sapiens RepID=UPI000173A15C

48. Q29988 HLA-C protein n=1 Tax=Homo sapiens RepID=Q29988\_HUMAN

49. B7Z760 cDNA FLJ54780, highly similar to HLA class I histocompatibility antigen, alpha chain F n=1 Tax=Homo sapiens RepID=B7Z760\_HUMAN

50. UPI0001AE718C UPI0001AE718C related cluster n=1 Tax=Homo sapiens RepID=UPI0001AE718C

51. B0V1N4 HLA-G histocompatibility antigen, class I, G n=2 Tax=Homo sapiens RepID=B0V1N4\_HUMAN

52. C0JT29 MHC class I antigen (Fragment) n=5 Tax=Homo sapiens RepID=C0JT29\_HUMAN

53. O19589 MHC class I histocompatibility antigen HLA-B (Fragment) n=1 Tax=Homo sapiens RepID=O19589\_HUMAN

54. B4DMA2 cDNA FLJ54023, highly similar to Heat shock protein HSP 90-beta n=1 Tax=Homo sapiens RepID=B4DMA2\_HUMAN

55. D5FHF9 MHC class I antigen (Fragment) n=2 Tax=Homo sapiens RepID=D5FHF9\_HUMAN

56. B3KQW1 cDNA FLJ33181 fis, clone ADRGL2003684, highly similar to HLA class I histocompatibility antigen, alpha chain H n=1 Tax=Homo sapiens RepID=B3KQW1\_HUMAN

57. B4E1T1 cDNA FLJ54081, highly similar to Keratin, type II cytoskeletal 5 n=1 Tax=Homo sapiens RepID=B4E1T1\_HUMAN

58. Q96FG8 ACAT1 protein n=1 Tax=Homo sapiens RepID=Q96FG8\_HUMAN

59. P24752 Acetyl-CoA acetyltransferase, mitochondrial n=1 Tax=Homo sapiens RepID=THIL\_HUMAN

60. UPI00001FD556 26S protease regulatory subunit S10B n=1 Tax=Homo sapiens RepID=UPI00001FD556

61. Q4QZC0 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=Q4QZC0\_HUMAN

62. P19474 52 kDa Ro protein n=1 Tax=Homo sapiens RepID=RO52\_HUMAN

63. Q4R7F0 Testis cDNA, clone: QtsA-15489, similar to human RNA binding motif protein, X-linked (RBMX), n=1 Tax=Macaca fascicularis RepID=Q4R7F0\_MACFA

64. Q562R1 Beta-actin-like protein 2 n=1 Tax=Homo sapiens RepID=ACTBL\_HUMAN

65. UPI0001AE791F UPI0001AE791F related cluster n=1 Tax=Homo sapiens RepID=UPI0001AE791F

66. Q2VPJ6 HSP90AA1 protein (Fragment) n=1 Tax=Homo sapiens RepID=Q2VPJ6\_HUMAN

67. Q0QER2 Isocitrate dehydrogenase 1 (Fragment) n=1 Tax=Homo sapiens RepID=Q0QER2\_HUMAN

68. B4DXS4 cDNA FLJ57227, highly similar to Isocitrate dehydrogenase n=1 Tax=Homo sapiens RepID=B4DXS4\_HUMAN

69. A8K8A6 cDNA FLJ76931, highly similar to Homo sapiens testes-specific heterogenous nuclear ribonucleoprotein G-T, mRNA n=1 Tax=Homo sapiens RepID=A8K8A6\_HUMAN

70. Q96E39 RNA-binding motif protein X-linked-like 1 (Kynurenine aminotransferase III) (Protein RBM1) n=2 Tax=Homo sapiens RepID=Q96E39\_HUMAN

71. B1AQP1 Upstream transcription factor 1 n=1 Tax=Homo sapiens RepID=B1AQP1\_HUMAN

72. Q7Z3Y8 Keratin, type I cytoskeletal 27 n=1 Tax=Homo sapiens RepID=K1C27\_HUMAN

73. Q9H6X9 cDNA: FLJ21717 fis, clone COL10322 n=1 Tax=Homo sapiens RepID=Q9H6X9\_HUMAN

74. UPI0001A5E585 PREDICTED: similar to protein expressed in prostate, ovary, testis, and placenta 2 n=1 Tax=Homo sapiens RepID=UPI0001A5E585

75. P0CG39 POTE ankyrin domain family member J n=1 Tax=Homo sapiens RepID=POTEJ\_HUMAN

76. D3DWP7 Heterogeneous nuclear ribonucleoprotein A/B, isoform CRA\_b n=2 Tax=Homo sapiens RepID=D3DWP7\_HUMAN

77. Q8IWP6 Class IVb beta tubulin n=1 Tax=Homo sapiens RepID=Q8IWP6\_HUMAN

78. O60884 DnaJ homolog subfamily A member 2 n=1 Tax=Homo sapiens RepID=DNJA2\_HUMAN

79. Q96B07 EIF4A2 protein n=5 Tax=Eutheria RepID=Q96B07\_HUMAN

80. UPI000186DC1C tubulin beta-1 chain n=1 Tax=Pediculus humanus corporis RepID=UPI000186DC1C

81. UPI000186CFD7 tubulin beta chain, putative n=1 Tax=Pediculus humanus corporis RepID=UPI000186CFD7

82. B3KS31 Tubulin, beta 6, isoform CRA\_a n=1 Tax=Homo sapiens RepID=B3KS31\_HUMAN

83. UPI000186CCD1 tubulin beta-3 chain, putative n=1 Tax=Pediculus humanus corporis RepID=UPI000186CCD1

84. UPI000186D1CD adenosylhomocysteinase, putative n=1 Tax=Pediculus humanus corporis RepID=UPI000186D1CD

85. B3KUN3 Adenosylhomocysteinase n=1 Tax=Homo sapiens RepID=B3KUN3\_HUMAN

86. UPI000186EB2A tubulin alpha-1 chain, putative n=1 Tax=Pediculus humanus corporis RepID=UPI000186EB2A

87. Q6L6T0 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=Q6L6T0\_HUMAN

**88. Q15853 Upstream stimulatory factor 2 n=1 Tax=Homo sapiens RepID=USF2\_HUMAN**

89. UPI0000D9FD95 keratin, type II cytoskeletal 4 n=1 Tax=Homo sapiens RepID=UPI0000D9FD95

90. UPI000186D735 heat shock protein 75 kDa, putative n=1 Tax=Pediculus humanus corporis RepID=UPI000186D735

91. B4DR68 cDNA FLJ58608, highly similar to Heat shock protein 75 kDa, mitochondrial n=1 Tax=Homo sapiens RepID=B4DR68\_HUMAN
92. D6RAF8 Putative uncharacterized protein HNRNPD n=3 Tax=Catarrhini RepID=D6RAF8\_HUMAN
93. Q12771 P37 AUF1 n=1 Tax=Homo sapiens RepID=Q12771\_HUMAN
94. B7Z1Y2 Fructose-bisphosphate aldolase n=2 Tax=Homo sapiens RepID=B7Z1Y2\_HUMAN
95. B3KU96 cDNA FLJ39433 fis, clone PROST2004142, highly similar to GDP-L-fucose synthetase (EC 1.1.1.271) n=1 Tax=Homo sapiens RepID=B3KU96\_HUMAN
96. D3DVX3 Isocitrate dehydrogenase 3 (NAD+) beta, isoform CRA\_b n=1 Tax=Homo sapiens RepID=D3DVX3\_HUMAN
97. B7Z8J7 cDNA FLJ57466 n=1 Tax=Homo sapiens RepID=B7Z8J7\_HUMAN
98. P00762 Anionic trypsin-1 n=1 Tax=Rattus norvegicus RepID=TRY1\_RAT
99. B4DLC0 cDNA FLJ58476, highly similar to Poly(rC)-binding protein 2 n=1 Tax=Homo sapiens RepID=B4DLC0\_HUMAN
100. C9WCV4 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=C9WCV4\_HUMAN
101. C0M111 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=C0M111\_HUMAN
102. Q9MYC1 Leucocyte antigen B (Fragment) n=1 Tax=Homo sapiens RepID=Q9MYC1\_HUMAN
103. B3GFZ6 Truncated MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=B3GFZ6\_HUMAN
104. B4Z1E1 MHC class I antigen (Fragment) n=1 Tax=Homo sapiens RepID=B4Z1E1\_HUMAN
105. UPI000173A5BC UPI000173A5BC related cluster n=1 Tax=Homo sapiens RepID=UPI000173A5BC
106. UPI0001A5ED94 PREDICTED: similar to CYorf16 protein n=1 Tax=Homo sapiens RepID=UPI0001A5ED94
107. Q6IPN6 Elongation factor 1-alpha n=1 Tax=Homo sapiens RepID=Q6IPN6\_HUMAN