## SUPPLEMENTARY

### Table S1. List of primers.

|  |  |
| --- | --- |
| Primera | Sequenceb |
| ASHH2-CW-D870A FW  ASHH2-CW-D870A RV  ASHH2-CW-insC866-SFPN-C871 FW  ASHH2-CW-insC866-SFPN-C871 RV  ASHH2-CW-I915A FW  ASHH2-CW I915A RV  ASHH2-CW L919A FW  ASHH2-CW L919A RV | 5’-TCGATGTGATgccTGCTTTAAATGGCGAC  5’-ACCCAGGCACTCTCTGTG  5’-cctaacTGCTTTAAATGGCGACGAATAC  5’-aaatgaACATCGAACCCAGGCACT  5’-AGAGTTGGGCATAGGACAGGATG  5’-TCATTagcTTCTTCATTTGACATCTCTTGAGATTTTGAGC  5’-AGAGgctGGCATAGGACAGGATGAAG  5’-TCATTAATTTCTTCATTTGACATCTCTTGAG |

aFW and RV indicate forward and reverse primers, respectively.

bLower case indicates mutated codons.

### Table S2. Histone mimicking peptides.

|  |  |  |
| --- | --- | --- |
| Name | Sequence | Manufacturer |
| H3  H3K4me1  H3K4me2  H3K4me3 | ARTKQTARY\*  ARTK(me1)QTARY\*  ARTK(me2)QTARY\*  ARTK(me3)QTARY\* | LifeTein LLC (95% purity)  LifeTein LLC (95% purity)  LifeTein LLC (95% purity)  LifeTein LLC (95% purity) |

\*An additional tyrosine was introduced for concentration determination by absorption at 280 nm (NanoDrop), and an extinction coefficient of 1490 M-1 cm-1 was used.

### Table S3. Summary of ITC data.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Ligand | Temperature, K | ΔH, kJ/mol | TΔS, kJ/mol | ΔG, kJ/mol | Kd, μM |
| CW wild type  H3K4me1  H3K4me2  H3K4me3  D870A  H3K4me1  H3K4me2  H3K4me3  L919A  H3K4me1 | 298.15  298.15  298.15  298.15  298.15  298.15  298.15 | -89.25±8.58  -83.64±3.11  -60.14±1.38  -79.22±4.81  -93.33±3.65  -57.47±2.37  -76.92±2.31 | -57.37±7.14  -53.18±3.23  -32.47±1.49  -45.70±4.10  -64.03±3.59  -30.45±2.65  -48.84±1.97 | -31.88±2.78  -30.46±0.15  -27.67±0.13  -33.52±0.71  -29.29±0.23  -27.02±0.29  -28.08±0.42 | 1.31±0.32  4.61±0.28  14.18±0.74  1.37±0.32  7.48±0.72  18.53±2.17  12.17±2.10 |

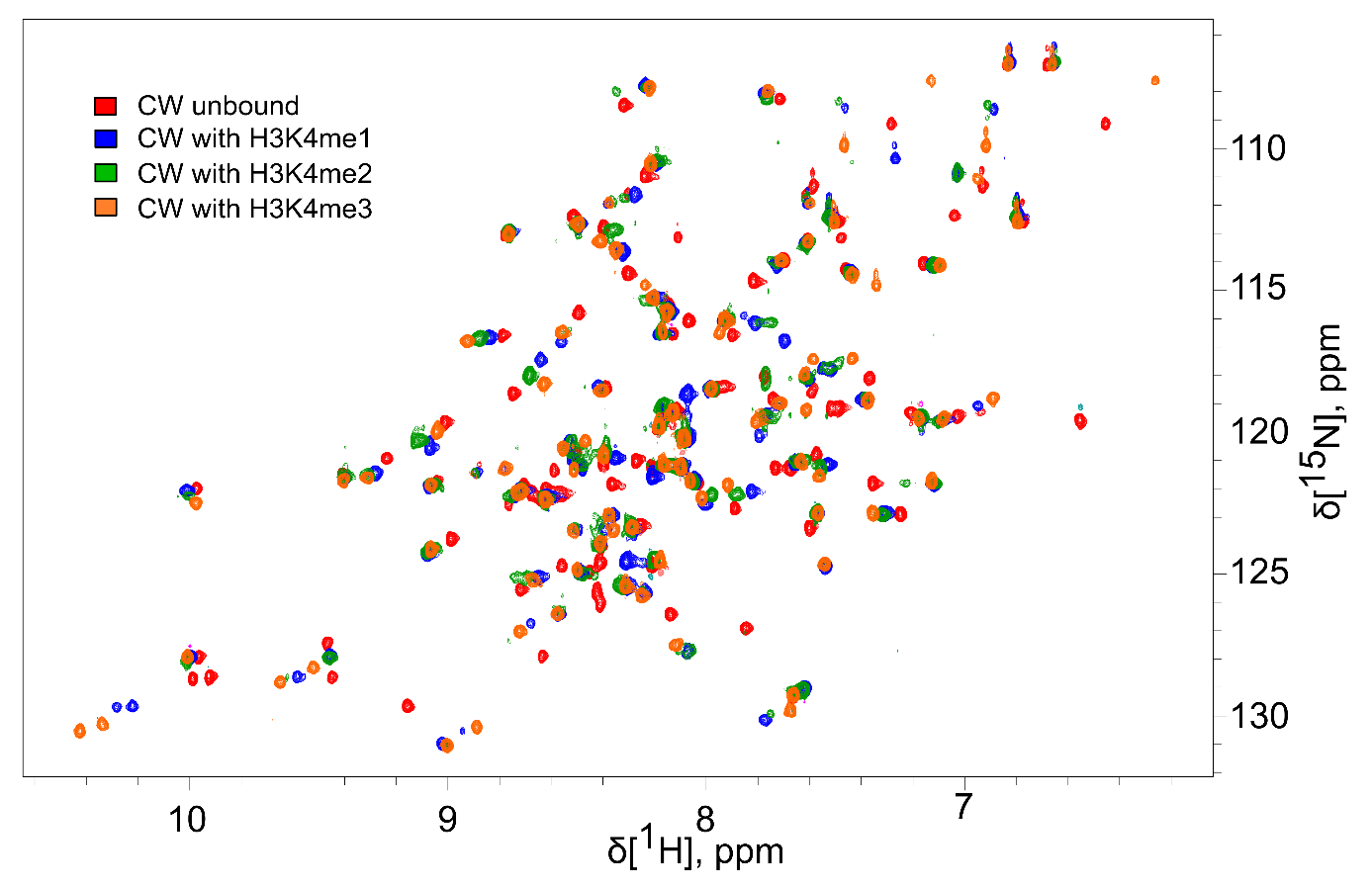


Figure S1.

Overlaid HSQC spectra of CW wild type in free state (red) and bound to H3K4me1 (blue), H3K4me2 (green) and H3K4me3 (orange) peptides.

A picture containing calendar

Description automatically generated

Figure S2

Titration curves and integrated data of: A-C – CW wt with H3K4me1/2/3 peptides; D-F – D870A with H3K4me1/2/3 peptides; G-I – L919A with H3K4me1/2/3 peptides; J – CW wt with H3 unmodified peptide; K – insC866-SFPN-C871 with H3K4me1 peptide; L – I915A with H3K4me1 peptide. In H-L no sigmoidal curve could be fitted.