Supplementary Information

Intricate Effects of α-Amino and Lysine Modifications on Arginine Methylation on the Histone H4 N-terminal Tail

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[‡] *These authors made equal contribution to this work*

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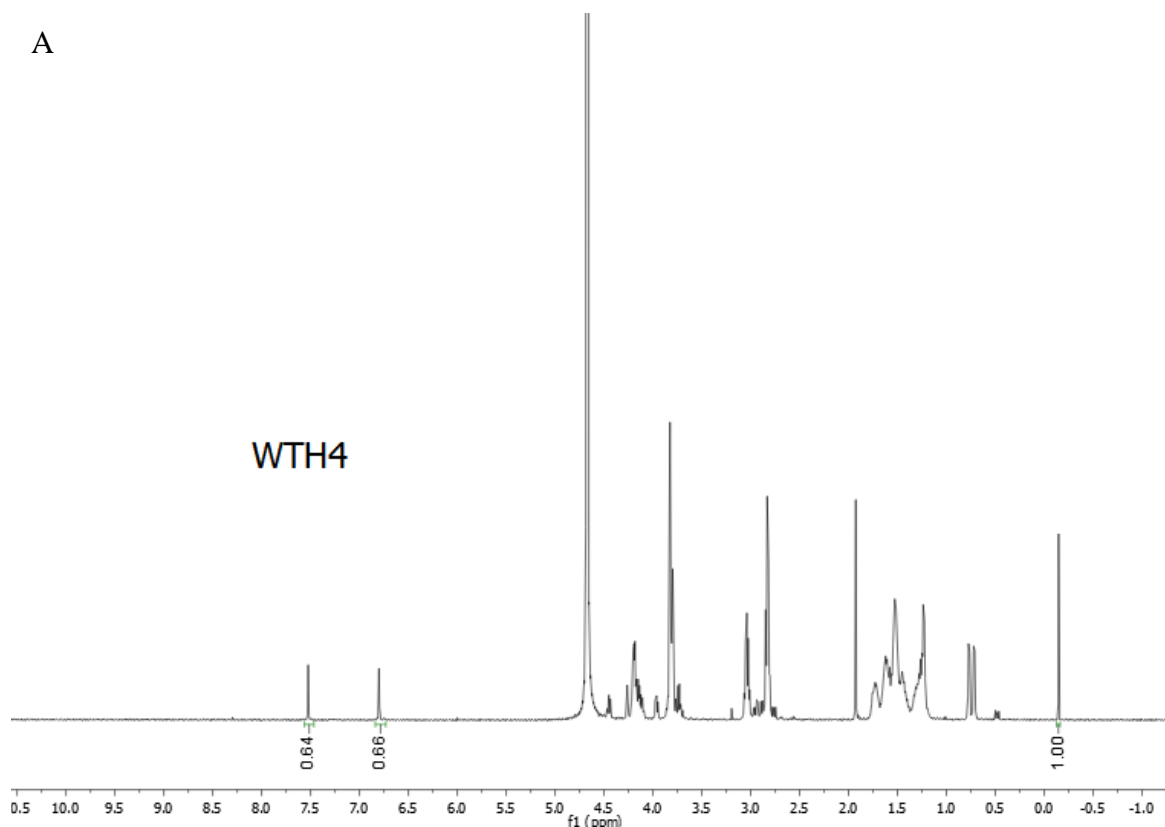
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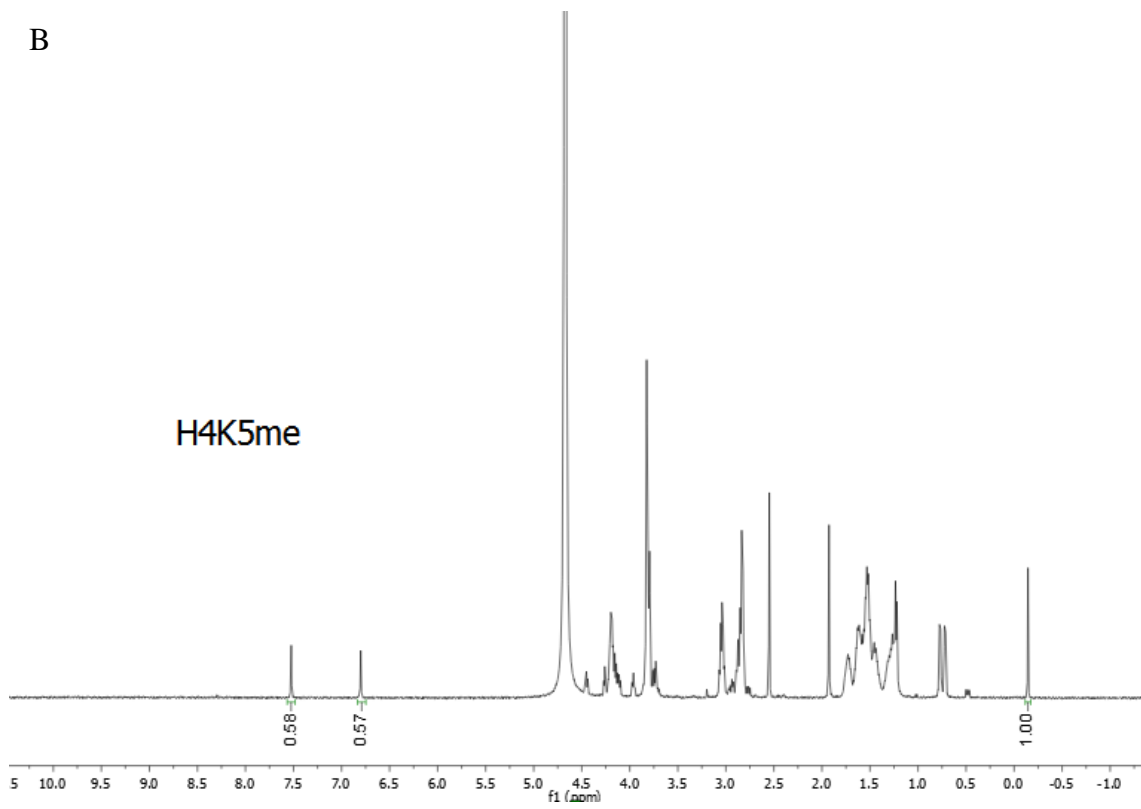
Table S1. Calibration of histone H4(1-20) peptide concentration by ^{1}H NMR. Adjustment factor = Mol (NMR) / Mol (weight).

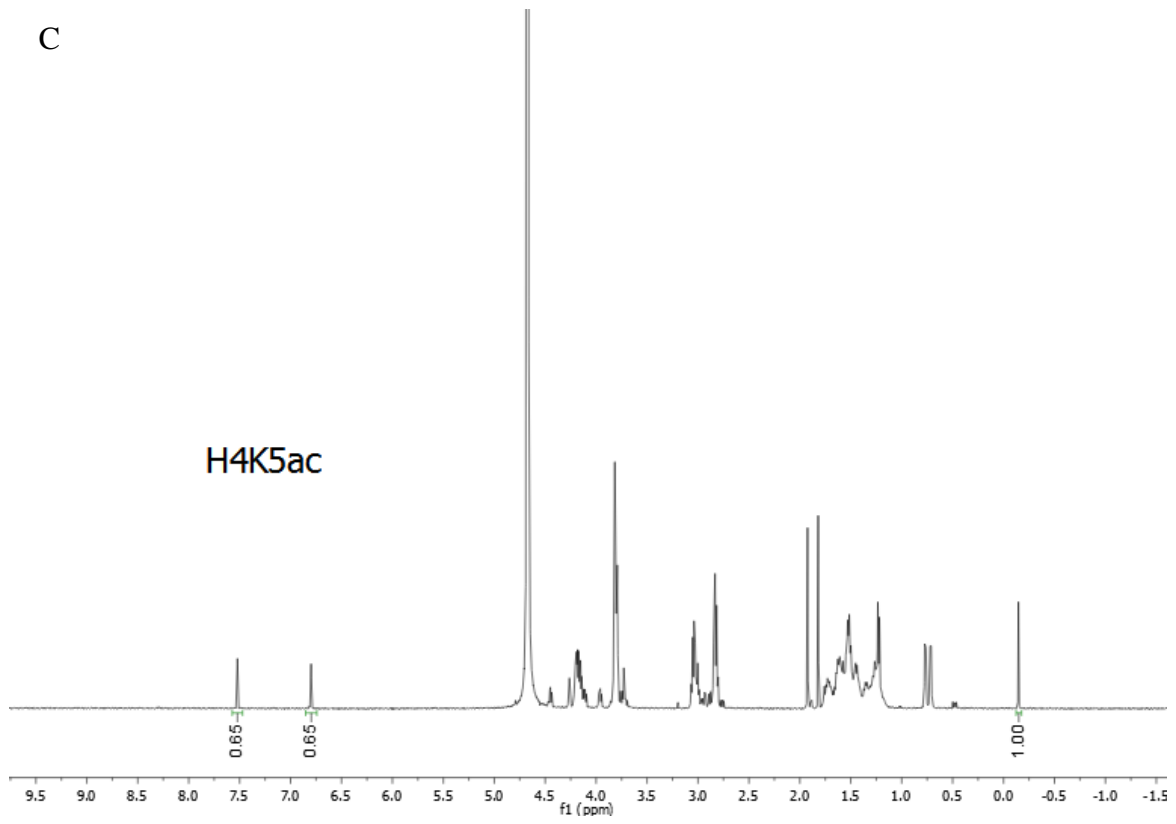| Peptide	Code for NMR Spectrum	Adjustment Factor
Ac-H4(1-20)	WTH4	0.64
Ac-H4(1-20)K5$_{me}$	H4K5me	0.58
Ac-H4(1-20)K5$_{ac}$	H4K5ac	0.65
Ac-H4(1-20)K5$_{pr}$	H4K5pr	0.64
Ac-H4(1-20)K5$_{bu}$	H4K5bu	0.65
Ac-H4(1-20)K5$_{cr}$	H4K5cr	0.77
Ac-H4(1-20)K5$_{hib}$	H4K5hib	0.67

Figure S1. NMR spectra of the histone H4(1-20) peptides. A) Ac-H4(1-20), code WTH4. B) Ac-H4(1-20)K5$_{me}$, code H4K5me. C) Ac-H4(1-20)K5$_{ac}$, code H4K5ac. D) Ac-H4(1-20)K5$_{pr}$, code H4K5pr. E) Ac-H4(1-20)K5$_{bu}$, code H4K5bu. F) Ac-H4(1-20)K5$_{cr}$, code H4K5cr. G) Ac-H4(1-20)K5$_{hib}$, code H4K5hib.
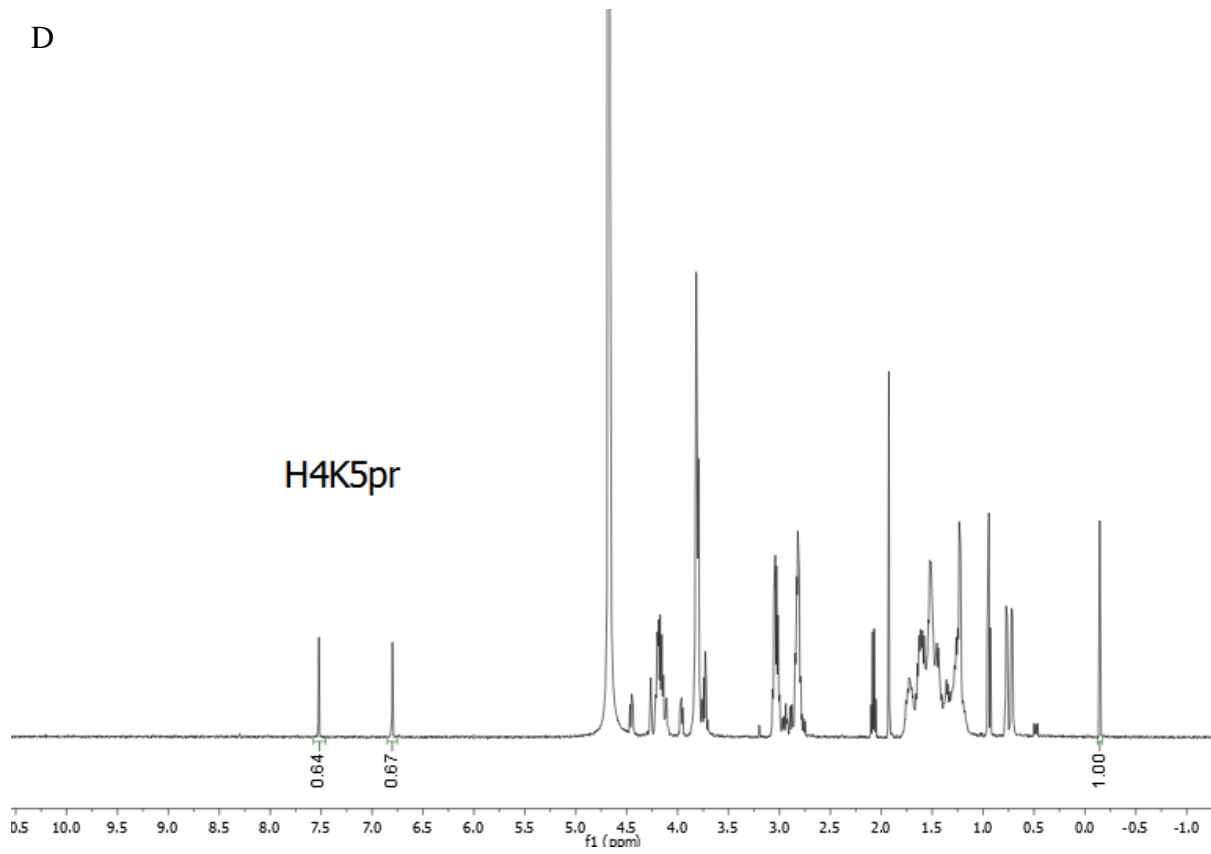

A

WTH4

B

H4K5me

0.58　0.57　1.00

5　10.0　9.5　9.0　8.5　8.0　7.5　7.0　6.5　6.0　5.5　5.0　4.5　4.0　3.5　3.0　2.5　2.0　1.5　1.0　0.5　0.0　-0.5　-1.0
f1 (ppm)

C

H4K5ac

0.65　0.65　1.00

9.5　9.0　8.5　8.0　7.5　7.0　6.5　6.0　5.5　5.0　4.5　4.0　3.5　3.0　2.5　2.0　1.5　1.0　0.5　0.0　-0.5　-1.0　-1.5
f1 (ppm)

D

H4K5pr

E

H4K5bu

F

H4K5cr

0.77   0.75   1.00

10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 -0.5
                                          f1 (ppm)

G

H4K5hib

0.67   0.64   1.00

.5 10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0 -0.5 -1
                                          f1 (ppm)

S5

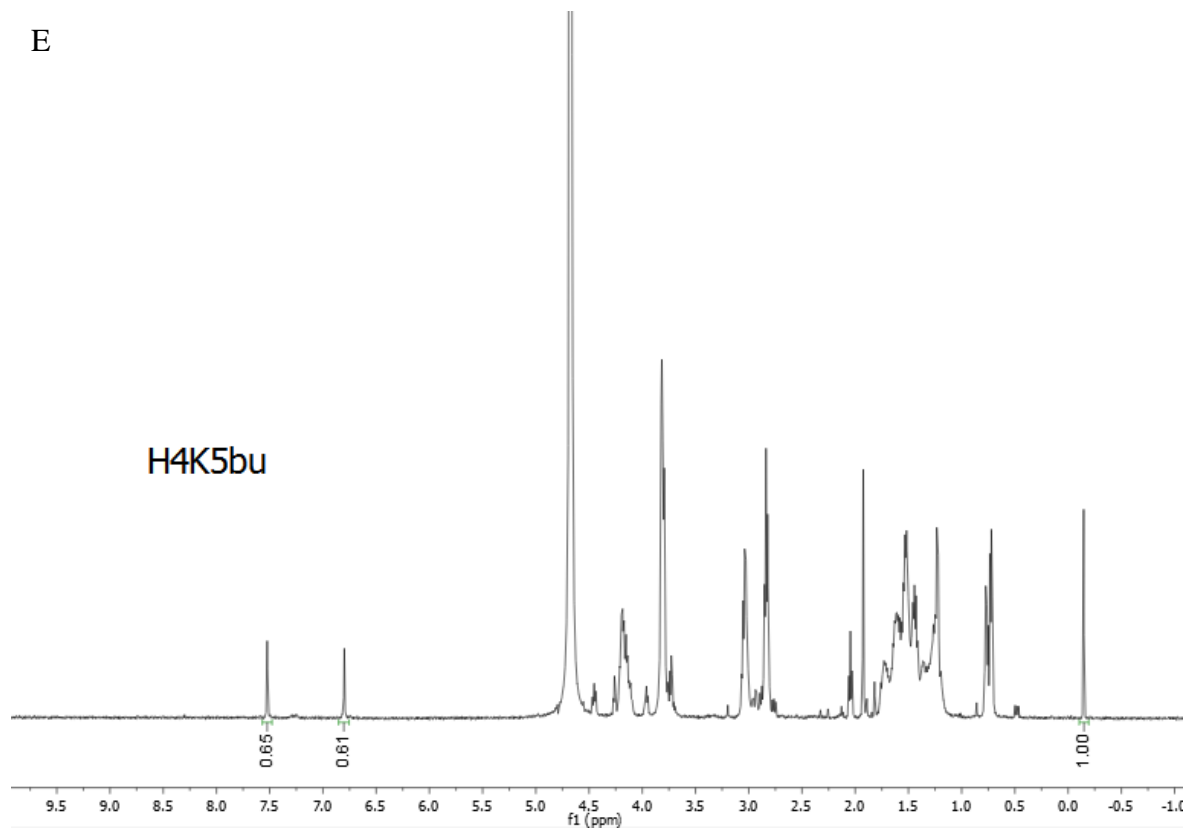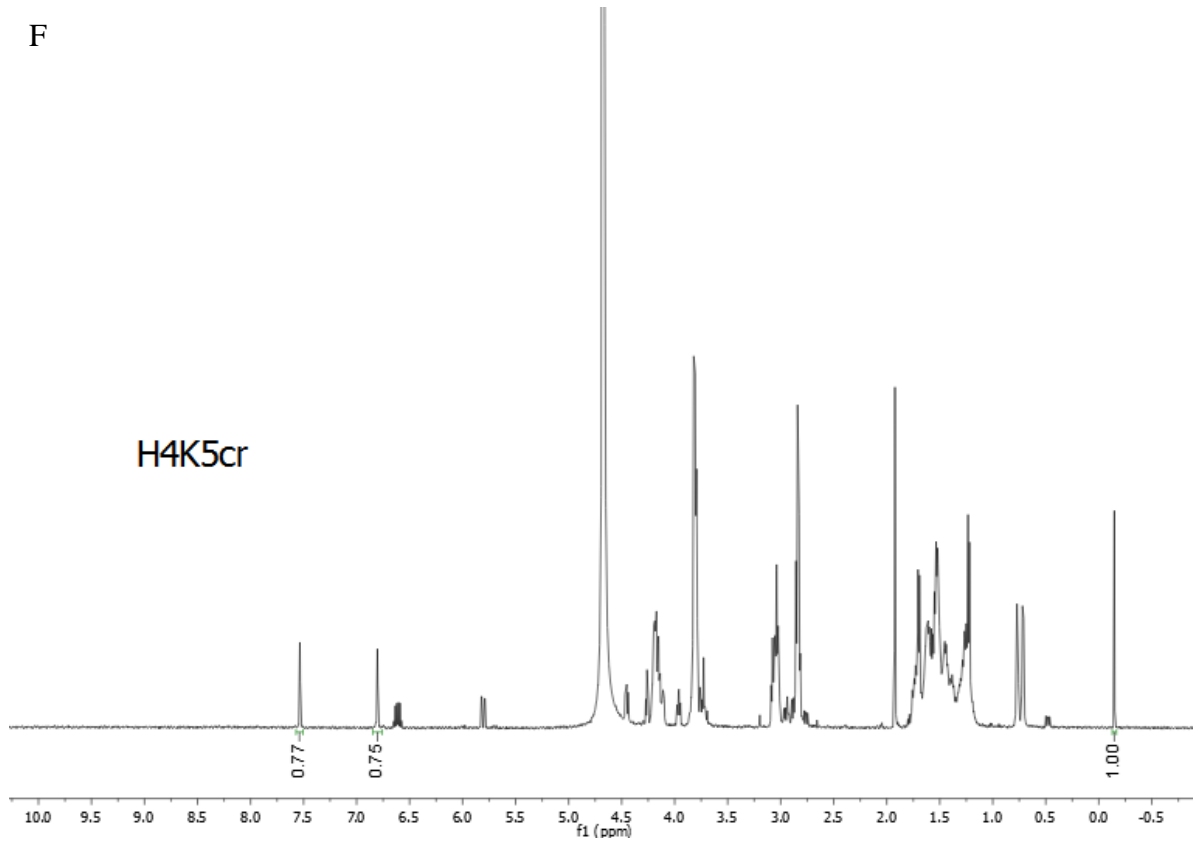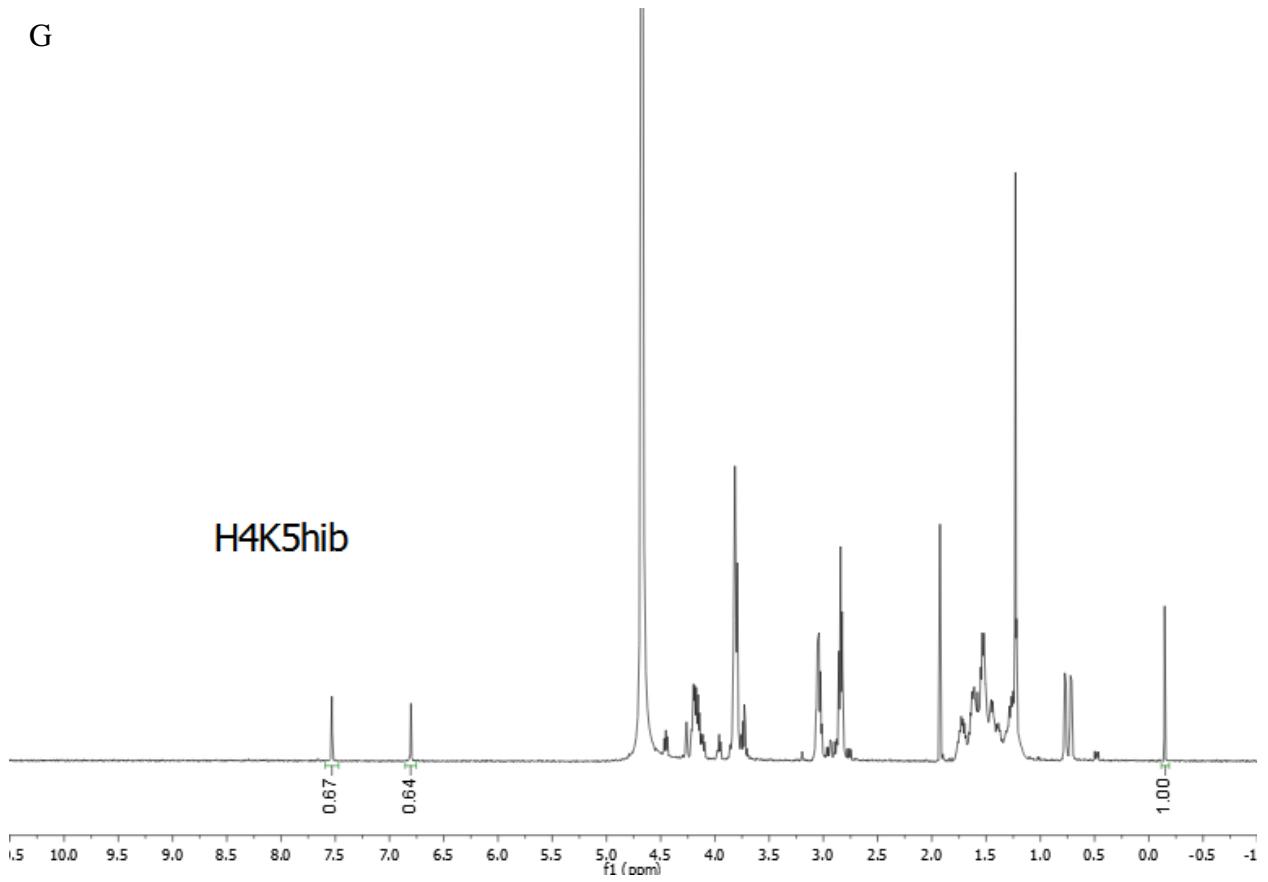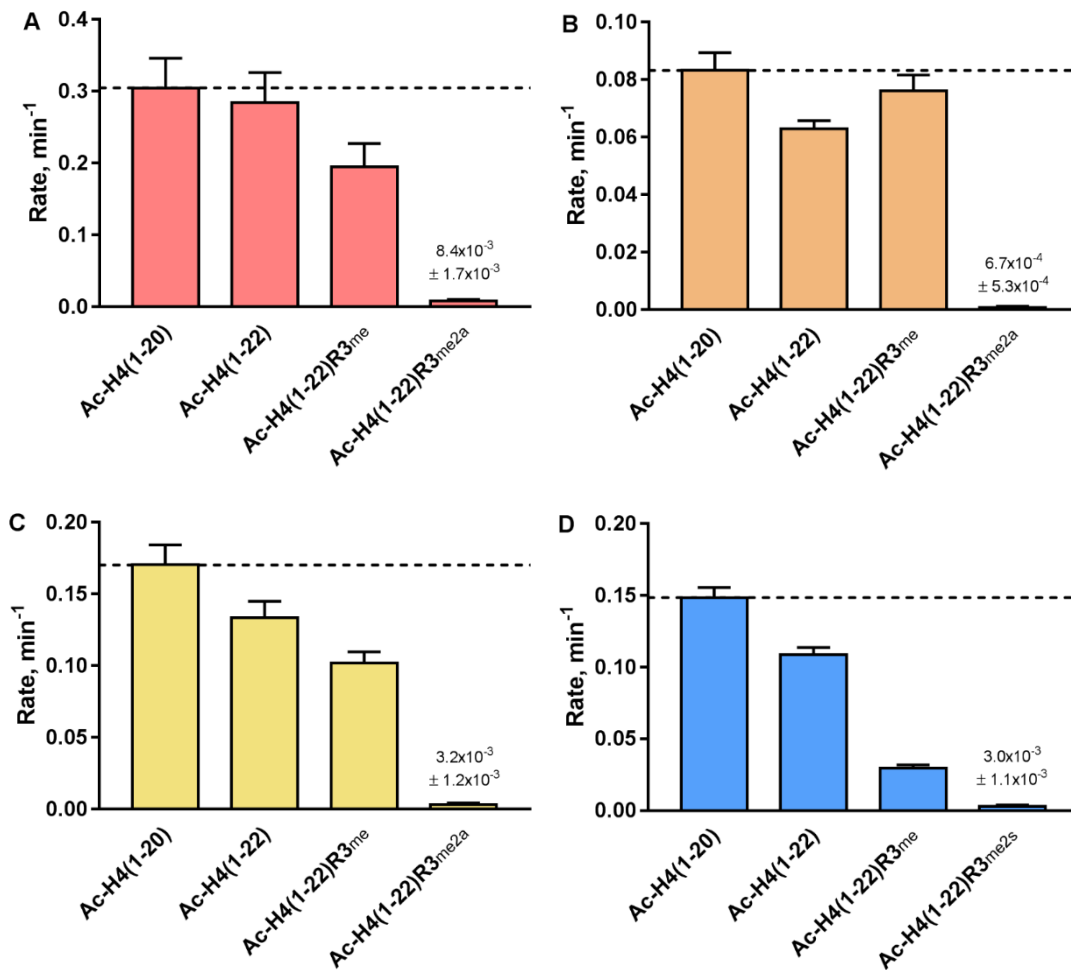**Figure S2.** The impact of monomethylated (MMA) and dimethylated (ADMA or SDMA) H4R3 on hPRMT activity. Single-point radioactive methylation assay performed with 0.05 µM hPRMT, 5 µM [^{14}C]-SAM, and 10 µM of peptide substrate at 30 ^{0}C over a period of 35 min. Average rates of arginine methylation by A) hPRMT1, B) hPRMT3, C) hPRMT8, and D) hPRMT5-MEP50 with peptide substrates Ac-H4(1-20), Ac-H4(1-22), Ac-H4(1-22)R3$_{me}$ (MMA), Ac-H4(1-22)R3$_{me2a}$ (ADMA), and Ac-H4(1-22)R3$_{me2s}$ (SDMA) are presented in each bar graph. Error bars represent standard deviation.