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Supplemental Material to:

Yasunao F Kamikawa and Mary E Donohoe

The localization of histone H3K27me3 demethylase Jmjd3 is dynamically regulated

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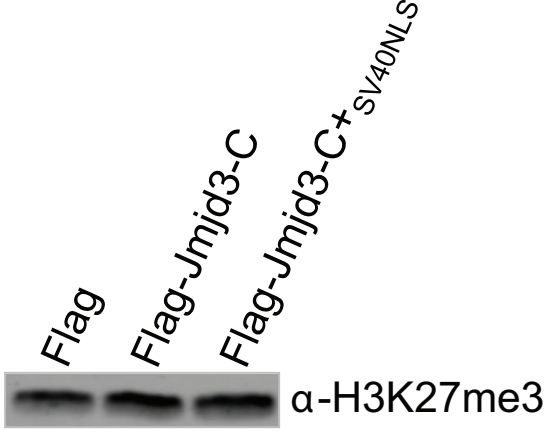
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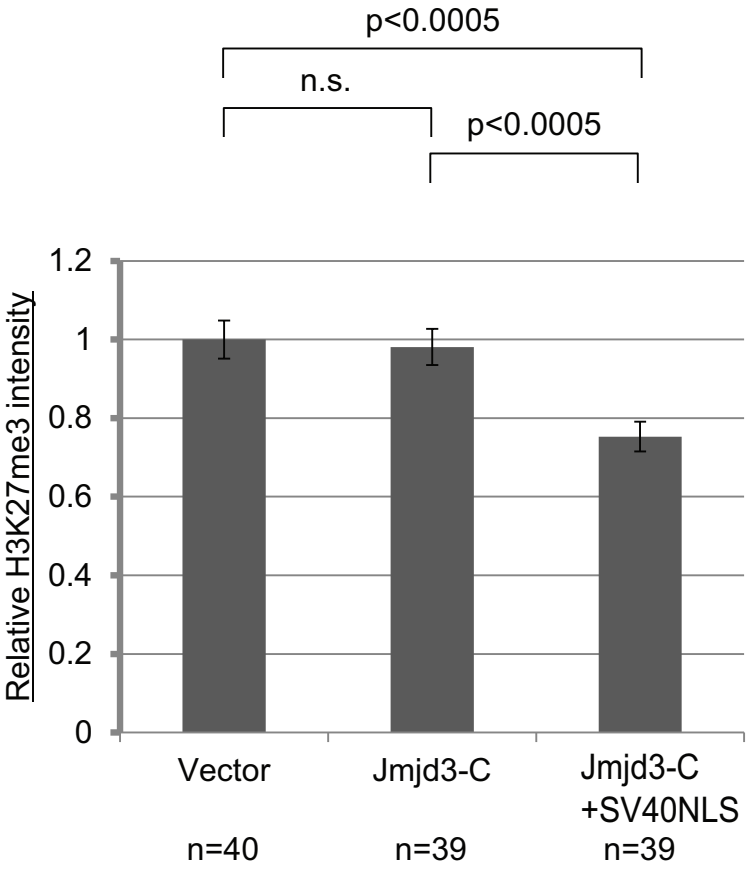
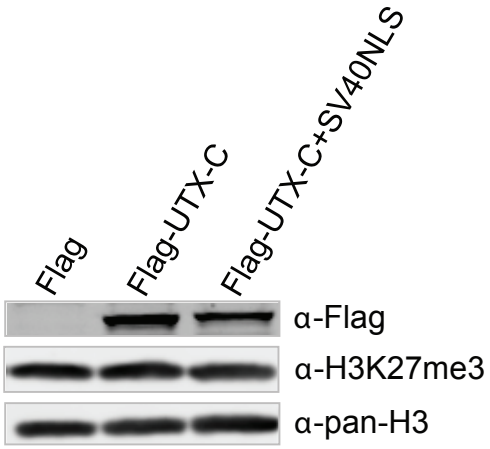
Flag-Jmjd3-C+SV40NLS
α-H3K27me3

B


p<0.0005
n.s.
p<0.0005
Relative H3K27me3 intensity
1.2
1
0.8
0.6
0.4
0.2
0
Vector
Jmjd3-C
Jmjd3-C
+SV40NLS
n=40
n=39
n=39

A



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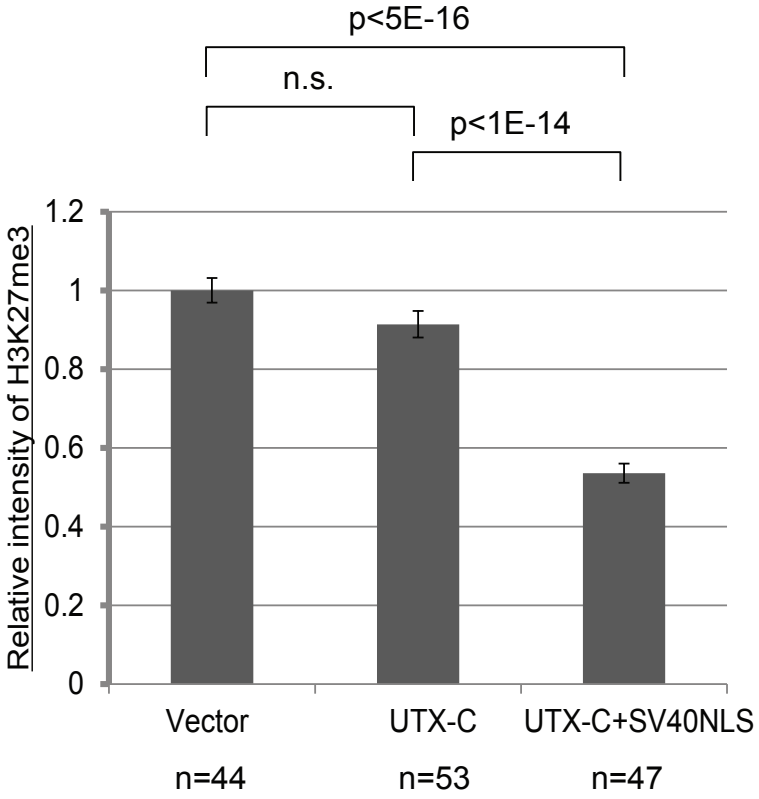
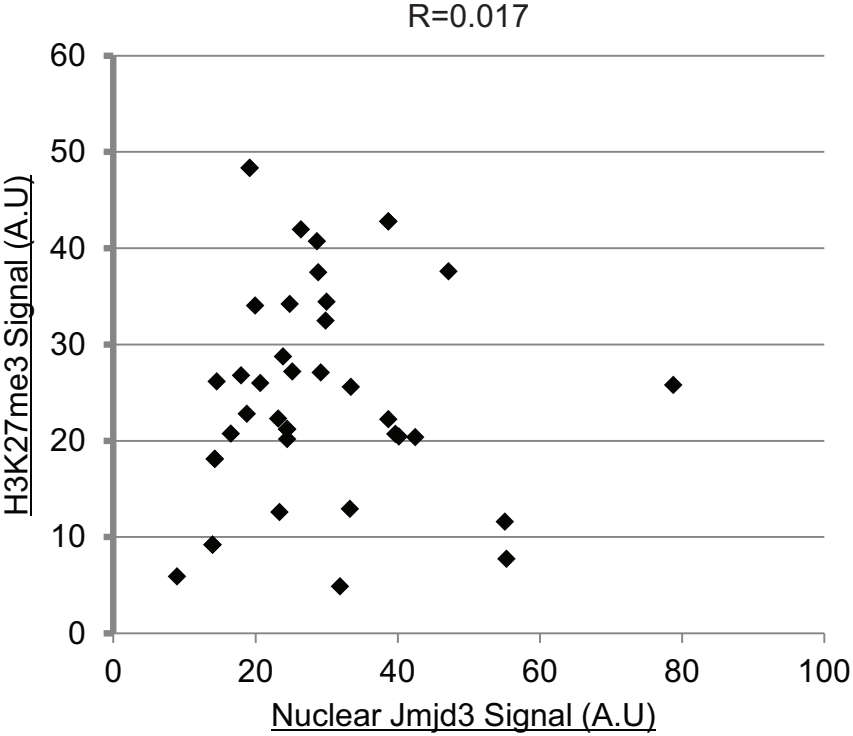


Kamikawa, Y. Figure S2



n=40

Kamikawa, Y. Figure S3

Supplemental Figure Legends

Figure S1.
(A) The total H3K27me3 levels were not altered by either the over expression of Jmjd3-C or Jmjd3-C+SV40NLS. Western blot of whole cell extracts prepared from Flag-alone, Flag-Jmjd3-C, and Flag-Jmjd3-C+SV40NLS shown in Figure 3A probed with anti-H3K27me3 antibodies. **(B)** An independent experiment and quantification of Figure 3B and C. The signal intensity of H3K27me3 in Jmjd3-C or Jmjd3-C+SV40NLS transfected cells was quantified. The values represent the mean of the individual transfectants. n= the total numbers of counted cells. The error bars represent one standard error. Statistics were performed using the Student's *t* test analysis.

Figure S2.
(A) The expression of Flag-UTX-C and UTX-C+SV40NLS was confirmed by western blot using anti-Flag antibodies. Pan-H3 was used as a protein loading control. The total H3K27me3 levels were not altered by either over expression of UTC-C or UTX-C+SV40NLS. **(B)** An independent experiment and quantification of Figure 3E and 3F. The signal intensity of H3K27me3 in UTX-C or UTX-C+SV40NLS transfected cells was quantified. The values represent the mean of the individual transfectants. n= the total number of counted cells. The error bars represent one standard error from the mean. Statistics were performed using the Student's *t* test analysis.

Figure S3.
An independent experiment and quantification of Figure 4C and D. The signals of the endogenous nuclear Jmjd3 and H3K27me3 in individual cells were plotted. n= the total numbers of counted cells. A.U.= arbitrary unit of fluorescence. R= correlation coefficiency.

Supplemental Table. 1 The sequences of oligonucleotide used in this study

Name	Sequence
Jmjd3 Full length Fwd BamHI	AAG**GGATCC** ATGCATCGGGCAGTGGAC
Jmjd3 Full length Rev XbaI	ATA**TCTAGA** TCATCGAGACGTGCTGGC
Jmjd3-C Fwd BamHI	AA**GGATCC** CTCTTTGATTTCCCACCCACTC
Jmjd3- N Rev XbaI	AAT**TCTAGA** TCA CCGAATGGATTCATCCAGC
Jmjd3 NLS1 K198A/R199A Mutant top	GGGGGTTCAGCAGATgcCgcCACTGGAGGGCCCCC
Jmjd3 NLS1 K198A/R199A Mutant bottom	GGGGGCCCTCCAGTGgcGgcATCTGCTGAACCCCC
Jmjd3 NLS2 K228A/R229A Mutant top	CCTTAGCCCTGGAGGCgcGgcCAGGAGAGGCTGCAGC
Jmjd3 NLS2 K228A/R229A Mutant bottom	GCTGCAGCCTCTCCTGgcCgcGCCTCCAGGGCTAAGG
UTX-C Fwd EcoRI	ATA**GAATTC** GTCCACTGGGCCTTCCCAGCATCTC
UTX-C Rev XbaI	ATT**TCTAGA** TCAAGATGAGGCGGATGGTAATGGAGGAG

The boldfaced letters and lowercase letters represent restriction enzyme sites and replaced nucleotides for mutagenesis, respectively.