Supplementary Information

Figure S1. Overexpression of PML recruits HDAC7 to PML NBs. PML was transiently transfected into HeLa cells. Immunostaining and confocal microscopy were performed according to Materials and Methods. The arrows represent a subset of cells that did not express transfected PML. Note that more HDAC7 was recruited to PML NBs in transfected cells.

Figure S2. Class IIa HDACs associate with PML in HeLa cells. *(A)* Expression plasmids encoding HDAC4, -5, OR -7 were singly or co-transfected with a PML expression plasmid followed by immunoprecipitation and immunoblotting with the indicated antibodies. FLAG-PML was co-precipitated with anti-HA antibodies only when HA-HDAC4, -5, or -7 were co-expressed (lanes 10, 12 & 14). *(B)* PML expression plasmid was transiently transfected into HeLa cells followed by immunostaining with anti-PML and anti-HDAC4 or anti-HDAC5 antibodies and confocal microscopy. Experiments were carried out as described in **Figure 1B**.

Figure S3. HDAC7 is the major Class IIa HDAC expressed in HUVECs. *(A)* Expression of class IIa HDAC mRNA levels in HUVECs. Equal amounts of RNA were used for RT-PCR (top panel) and real-time PCR (bottom panel) as described in "Materials and Methods". HDAC7 mRNA (lanes 3 & 7) was more abundant than HDAC4 (lanes 1 & or 5), or 5 (lanes 2 & 6). HDAC9 was undetectable under this condition. *(B)* Protein levels of class IIa HDACs in HUVECs, HeLa, and C2C12 cells. HA-HDAC4, HA-HDAC5, and HA-HDAC7 were transiently transfected into HeLa cells as a positive control (lanes 1-3 and 7). Whole cell extracts were prepared and probed with anti-HA antibodies. At equal

concentrations of HA-HDAC4, HDAC5, and HDAC7 (internal control for normalization) (lane 7), only endogenous HDAC7 was detected in HUVECs (lane 5).

Figure S4. CBP partially colocalizes with PML NBs. Confocal microscopic images of TNFα treated cells immunostained with anti-CBP (panels a-d), -HDAC1, -2, -3, -5, -6, and -N-CoR antibodies. Note: in untreated cells (data no shown), CBP partially colocalized with PML NBs and was largely colocalized when treated with TNFα. The distribution of the HDACs and N-CoR were not altered by TNFα treatment.

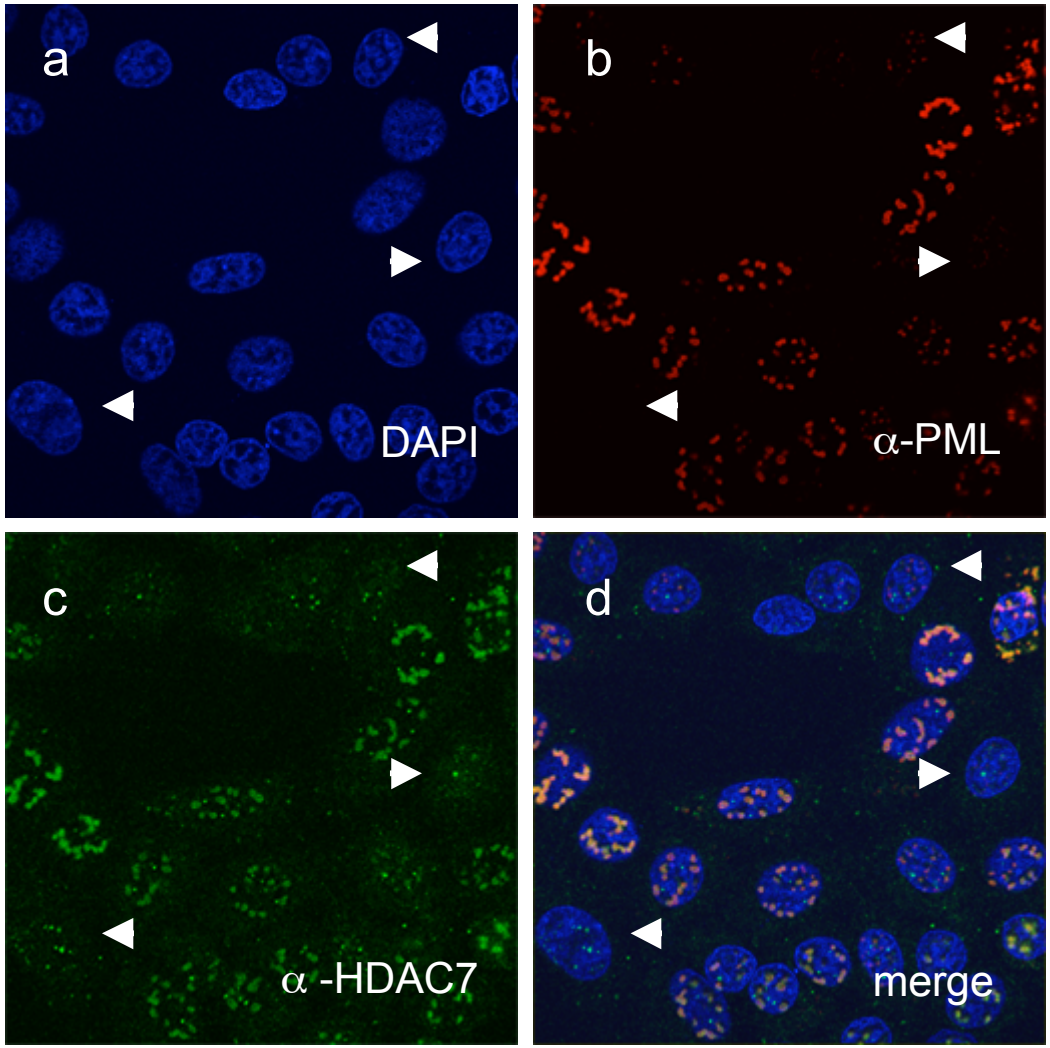
Figure S5. PML does not associate with the MMP-10 promoter following TNFα exposure of HUVECs. ChIP assays were performed as described in Figure 3. While changes in histone H4 acetylation were observed in response to TNFα stimulation, no PML association with MMP-10 promoter was detected.

Figure S6. PML overcomes Class IIa HDAC-mediated MEF2 repression. *(A)* Transient transfection reporter assays were performed as described in Fig. 4 except that HDAC7, PML, or MEF2C expression plasmids were included as indicated. In the presence of transfected HDAC7, overexpression of PML increased MEF2 reporter activity. (*B)* Expression plasmids of Class IIa HDACs were co-transfected with PML and a MEF2 reporter construct. Transfection assays were performed as described in Fig. 4. Note that in the presence of transfected PML, overexpression of HDAC4, -5, or -7 decreased MEF2 reporter activity.

Figure S7. Amino acids 552-633 are insufficient to bind HDAC7. HA-HDAC7 and FLAG-PML4 full-length or truncations were co-transfected into HeLa cells. Whole cell extracts were prepared and coimmunoprecipitations were performed followed by immunoblots with anti-HA and anti-FLAG antibodies. Note that full-length PML4 co-precipitated with anti-HA antibodies, but amino acids 332-552 or 552-633 did not.

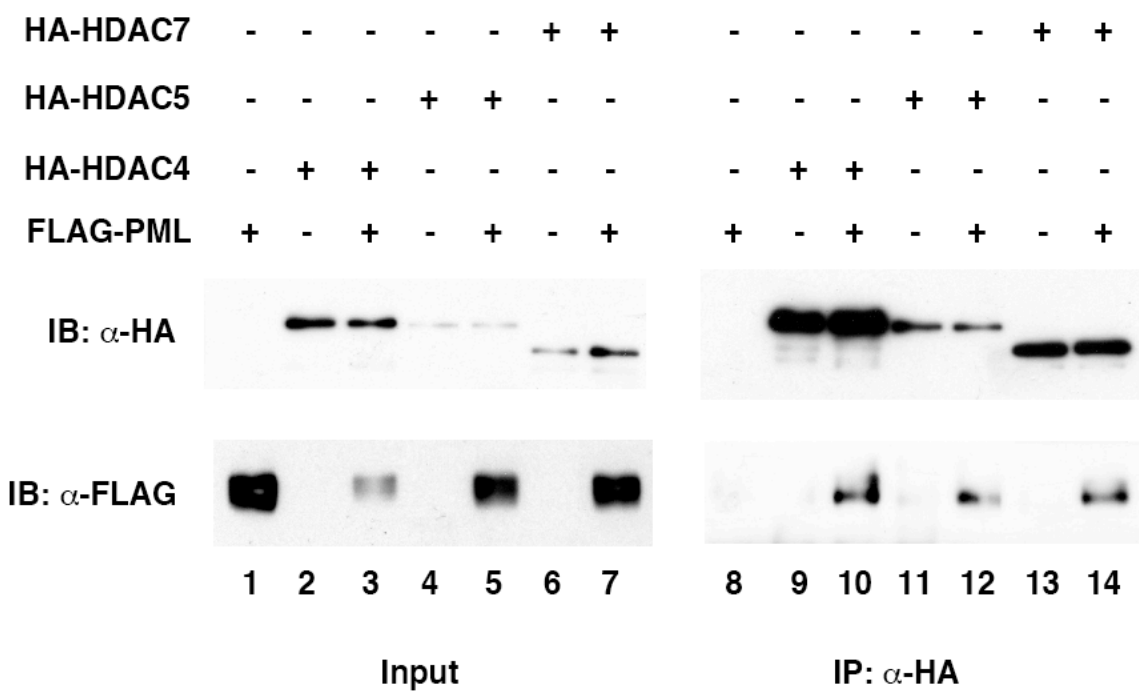
Figure S8. Overexpression of HDAC7 enhanced the association between HDAC7 and PML. HeLa cells were transfected with or without HA-HDAC7 expression plasmids. Whole cell extracts were prepared followed by immunoprecipitation with normal IgG or anti-PML antibodies. Immunoprecipitates were subjected to immunoblotting with anti-PML or anti-HDAC7 antibodies. In the presence of exogenous HA-HDAC7 (lane 1), more HDAC7 was co-precipitated by anti-PML antibodies (lanes 5).

Figure S9. Anti-HDAC7 antibodies recognize a single band whose intensity is decreased by HDAC7 siRNA in HUVECs. siRNA knockdown of HDAC7 was carried out using either DF1 (lanes 1 & 2) or LF2000 (lanes 3 & 4) transfection reagents in HUVECs with control (lanes 1 & 3) or HDAC7 siRNA (lanes 2 & 4). Whole cell extracts were prepared for immunoblotting with anti-HDAC7 antibodies. Note that anti-HDAC7 antibody only detects a single band. Furthermore, knockdown of HDAC7 resulted in a decrease in the single band detected by our antibody in HUVEC extracts. This further demonstrates the specificity of our antibody.



Gao_FigS1

A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HA-HDAC5 - - - + + - - - - - + + - -
HA-HDAC4 - + + - - - - - + + - - - -
FLAG-PML + - + - + - + + - + - + - +
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IB: α-FLAG
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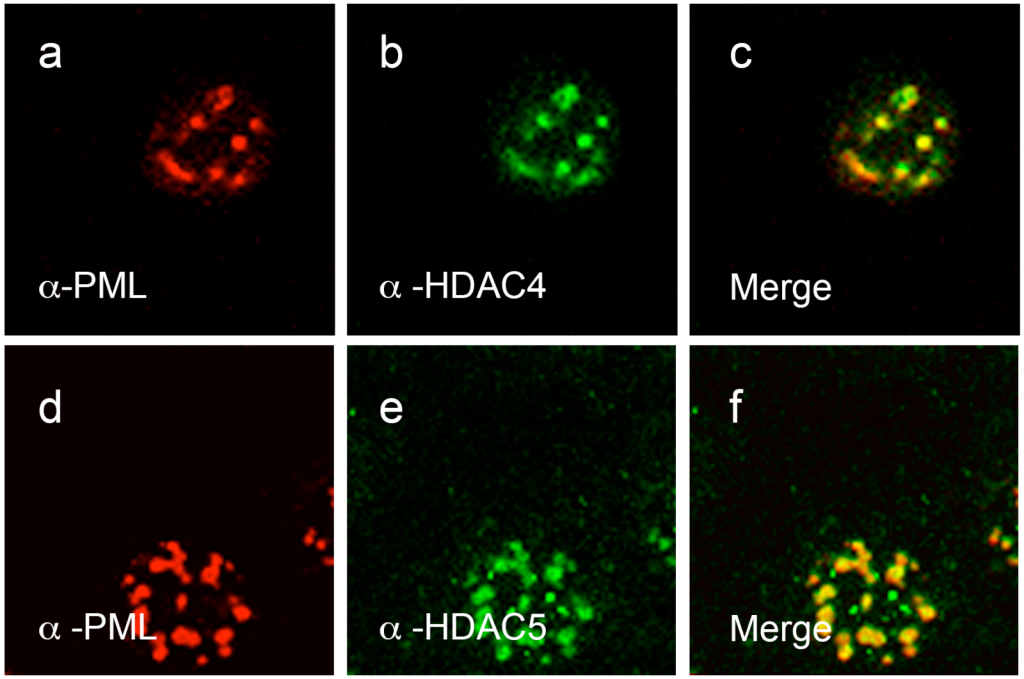
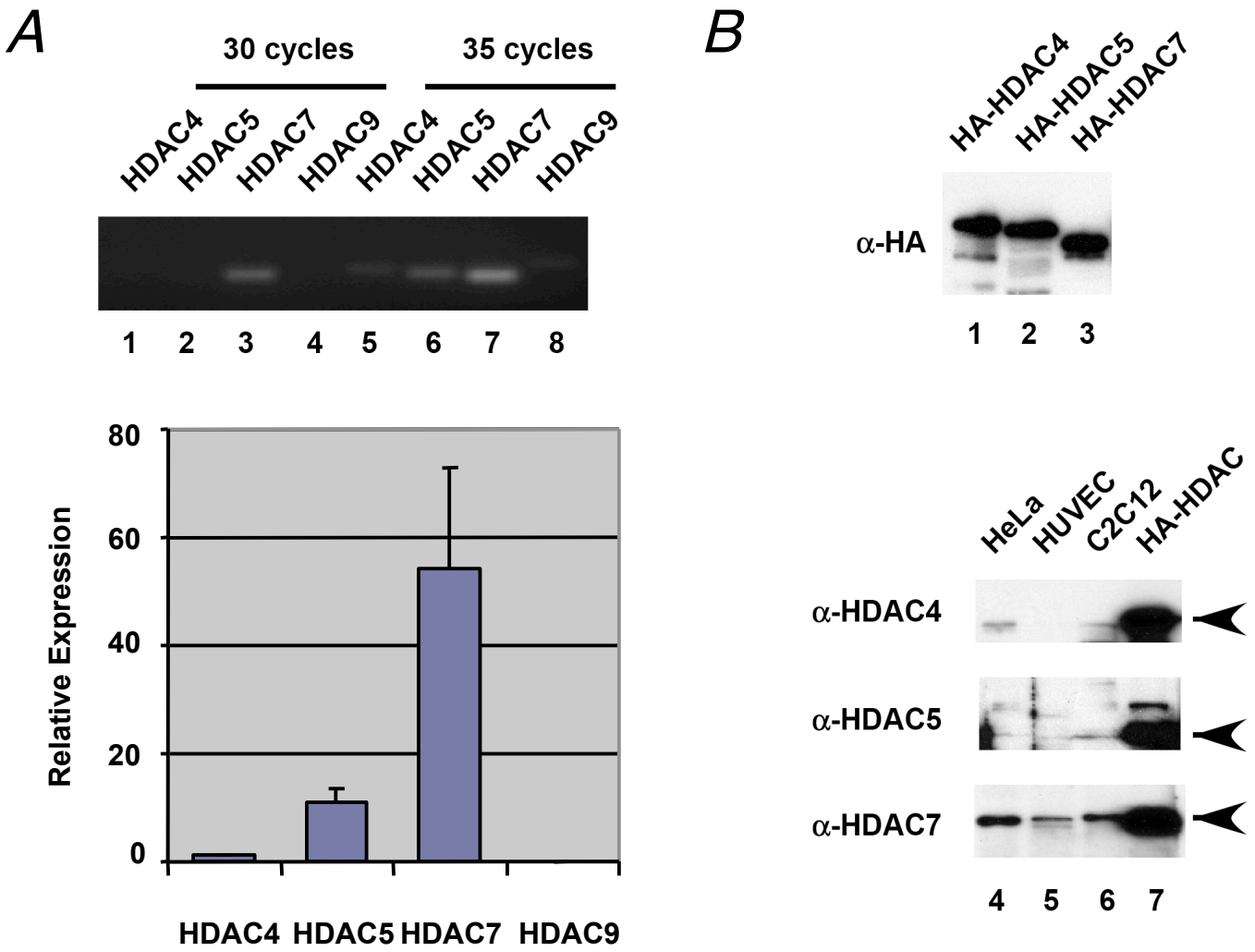
Input
IP: α-HA

B



Gao_FigS3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HDAC5
HDAC7
HDAC9
HDAC4
HDAC5
HDAC7
HDAC9
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Relative Expression
80
60
40
20
0
HDAC4 HDAC5 HDAC7 HDAC9
B
HA-HDAC4
HA-HDAC5
HA-HDAC7
α-HA
1 2 3
HeLa
HUVEC
C2C12
HA-HDAC
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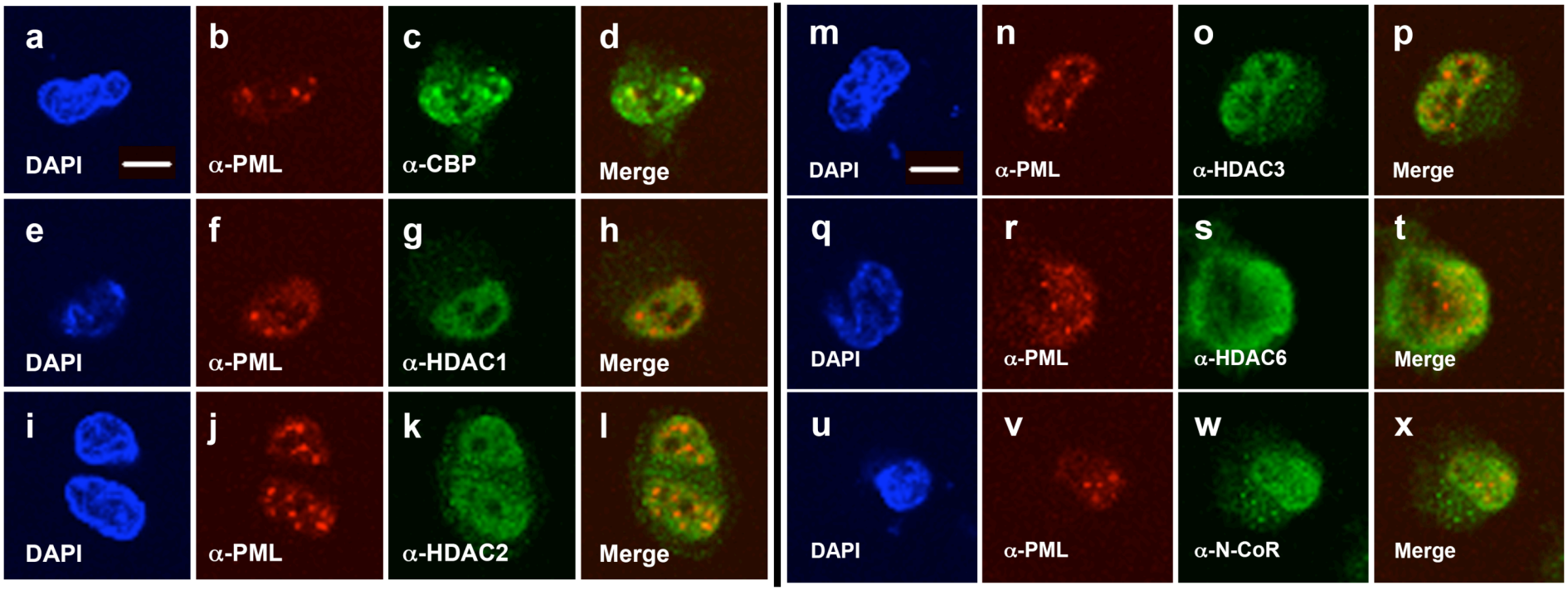
α-HDAC5
α-HDAC7
4 5 6 7

Gao_FigS4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d
Merge
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Merge
i
DAPI
j
α-PML
k
α-HDAC2
l
Merge
m
DAPI
n
α-PML
o
α-HDAC3
p
Merge
q
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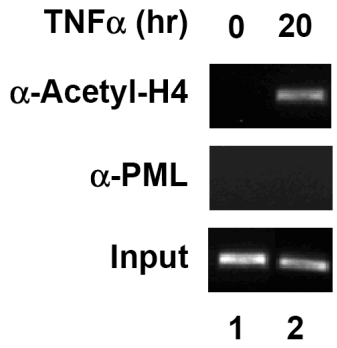
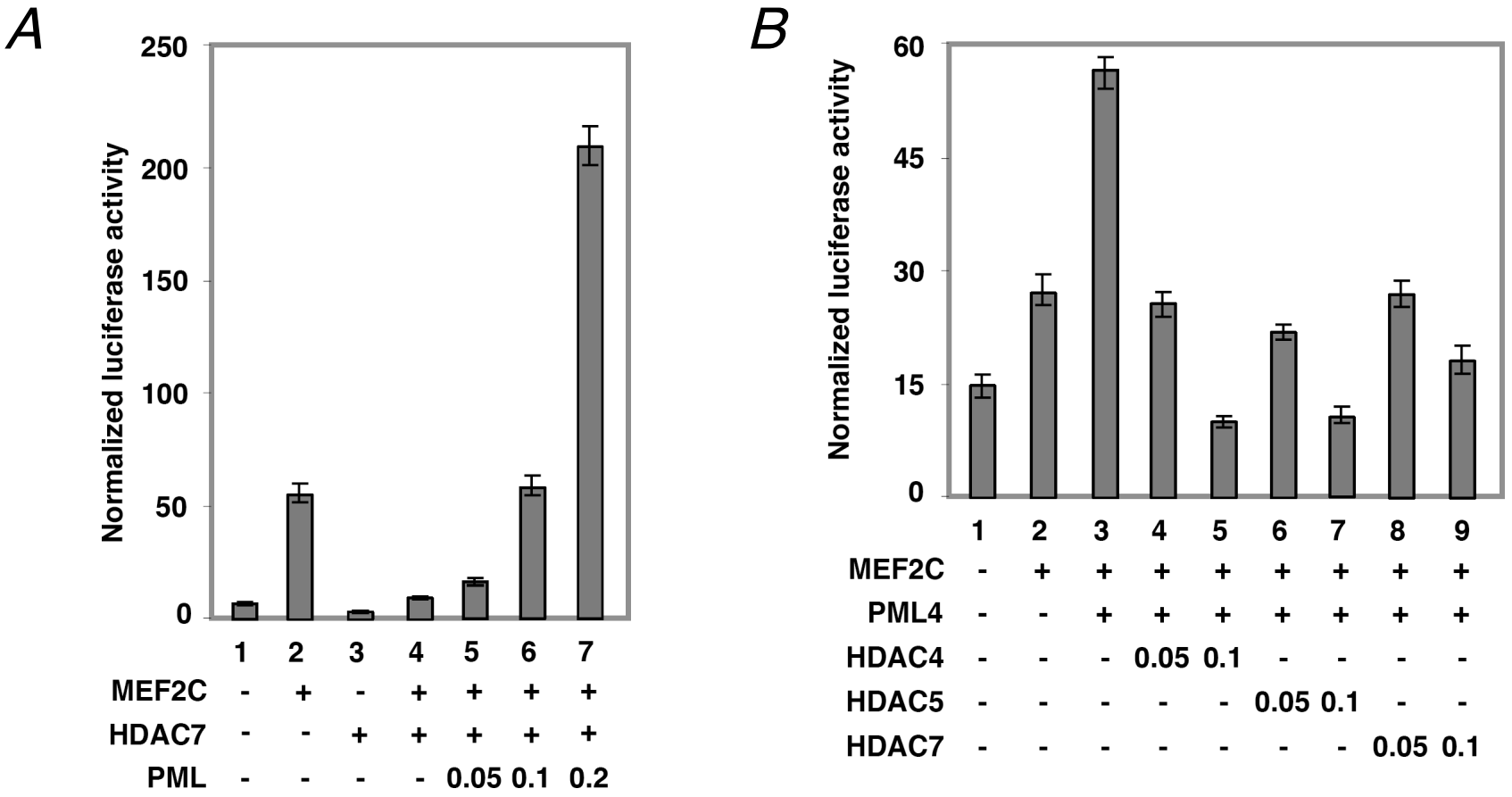


Gao_FigS5

Gao_FigS6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200
150
100
50
0
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MEF2C - + - + + + +
HDAC7 - - + + + + +
PML - - - - 0.05 0.1 0.2
B
Normalized luciferase activity
60
45
30
15
0
1 2 3 4 5 6 7 8 9
MEF2C - + + + + + + + +
PML4 - - + + + + + + +
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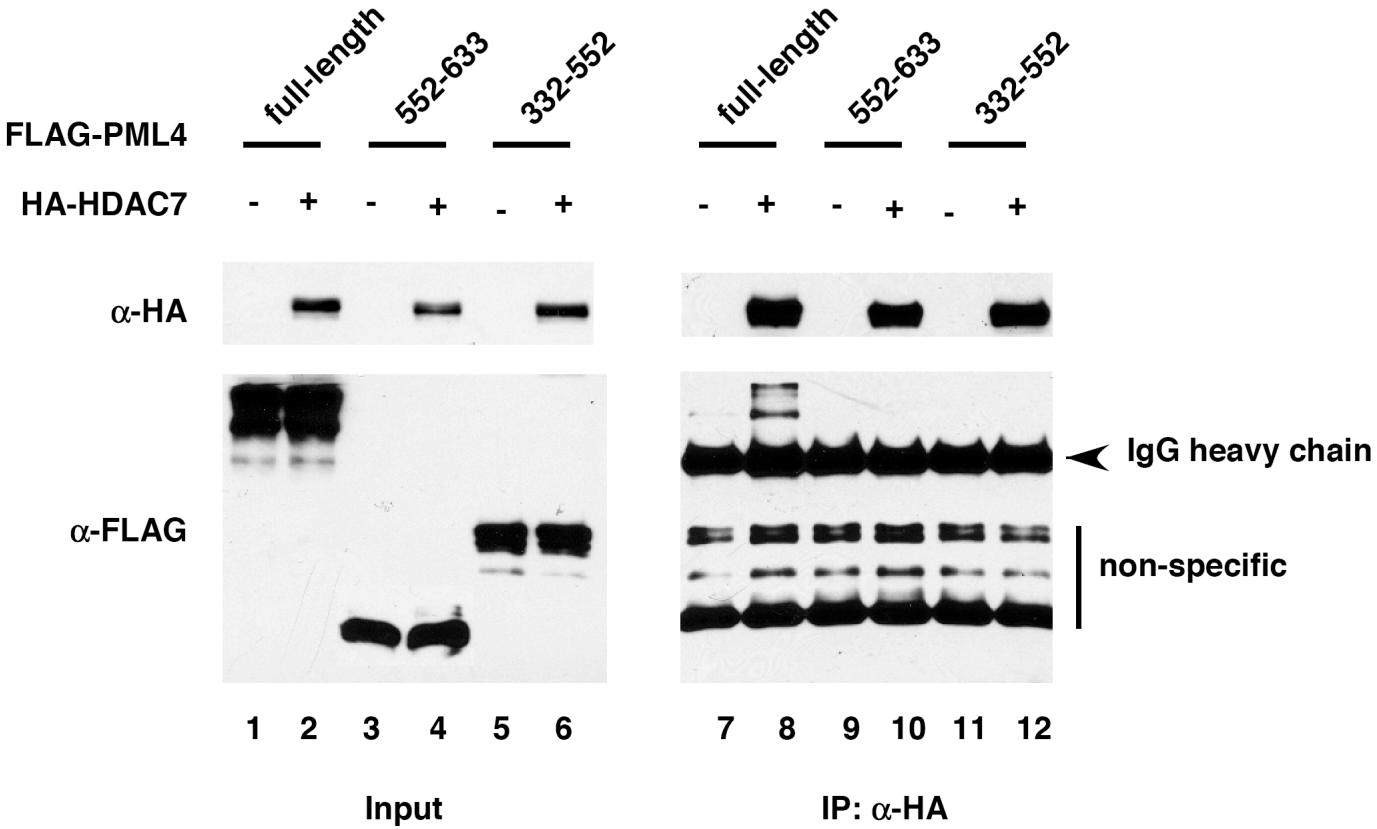
HDAC5 - - - - - 0.05 0.1 - -
HDAC7 - - - - - - - 0.05 0.1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Input
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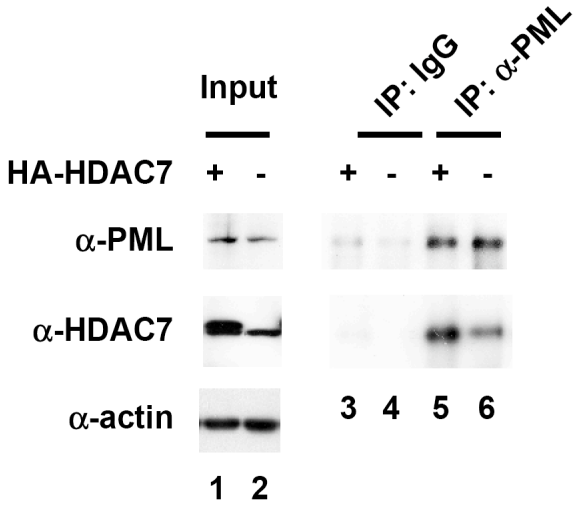
Gao_FigS7



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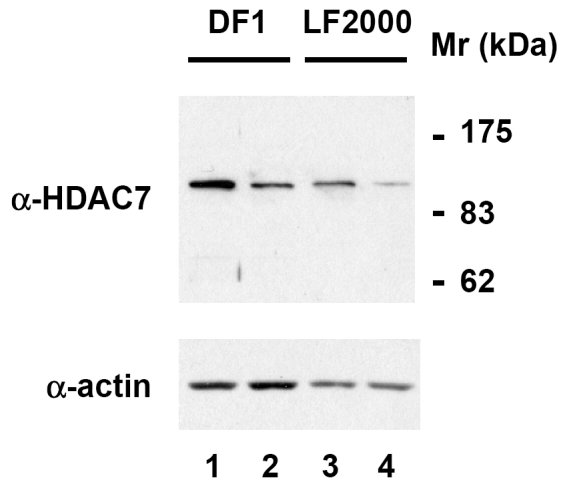

Gao_FigS9