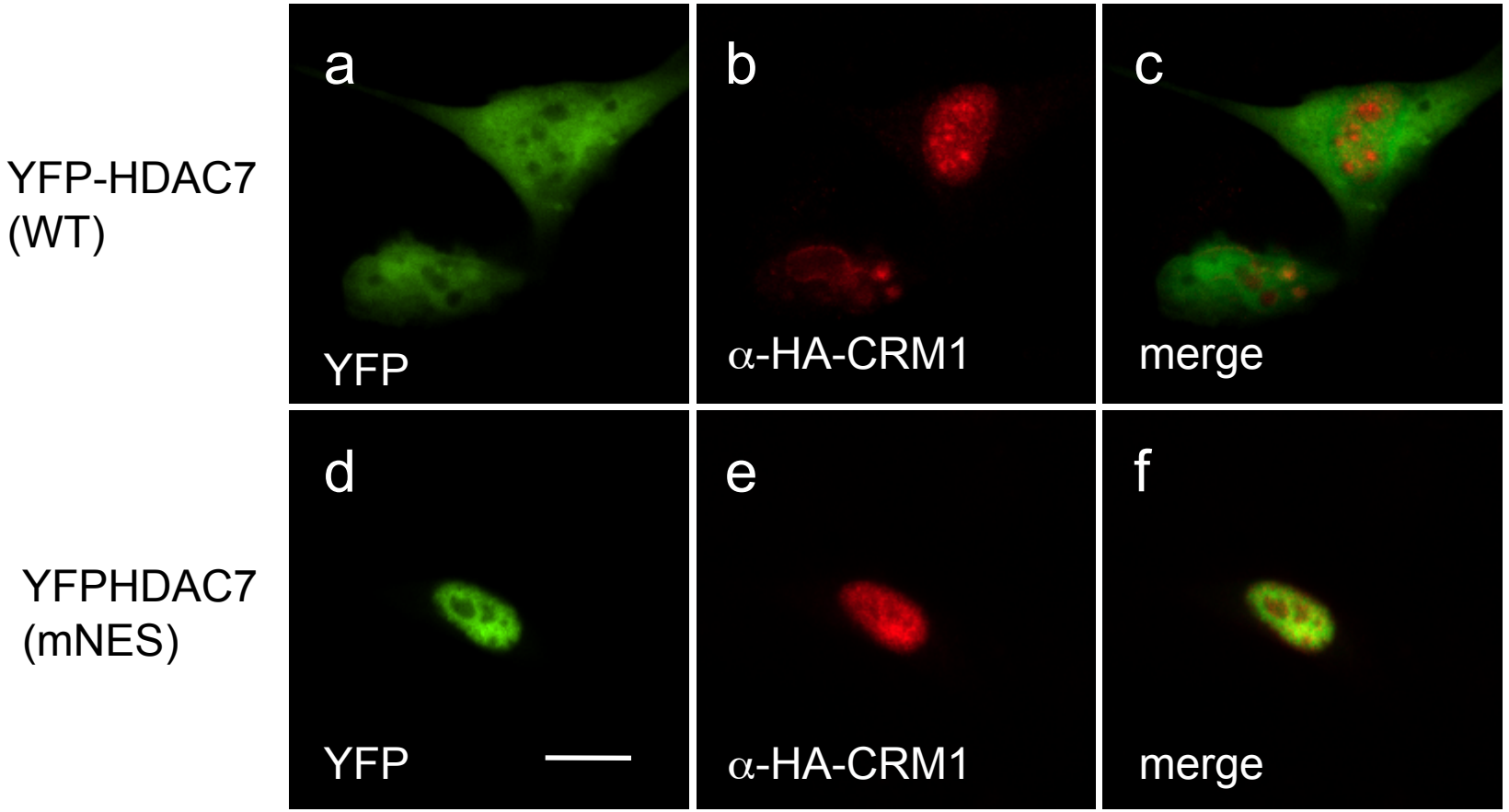
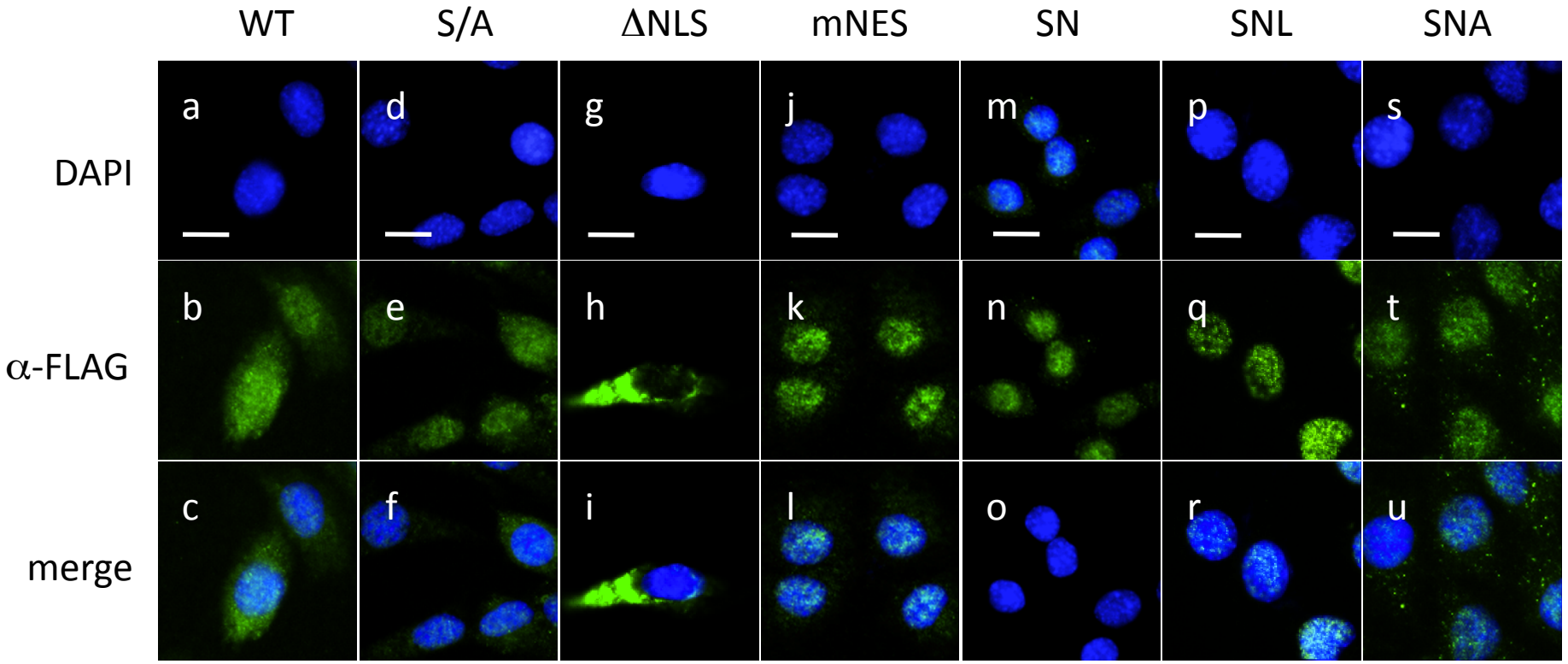


Fig. S1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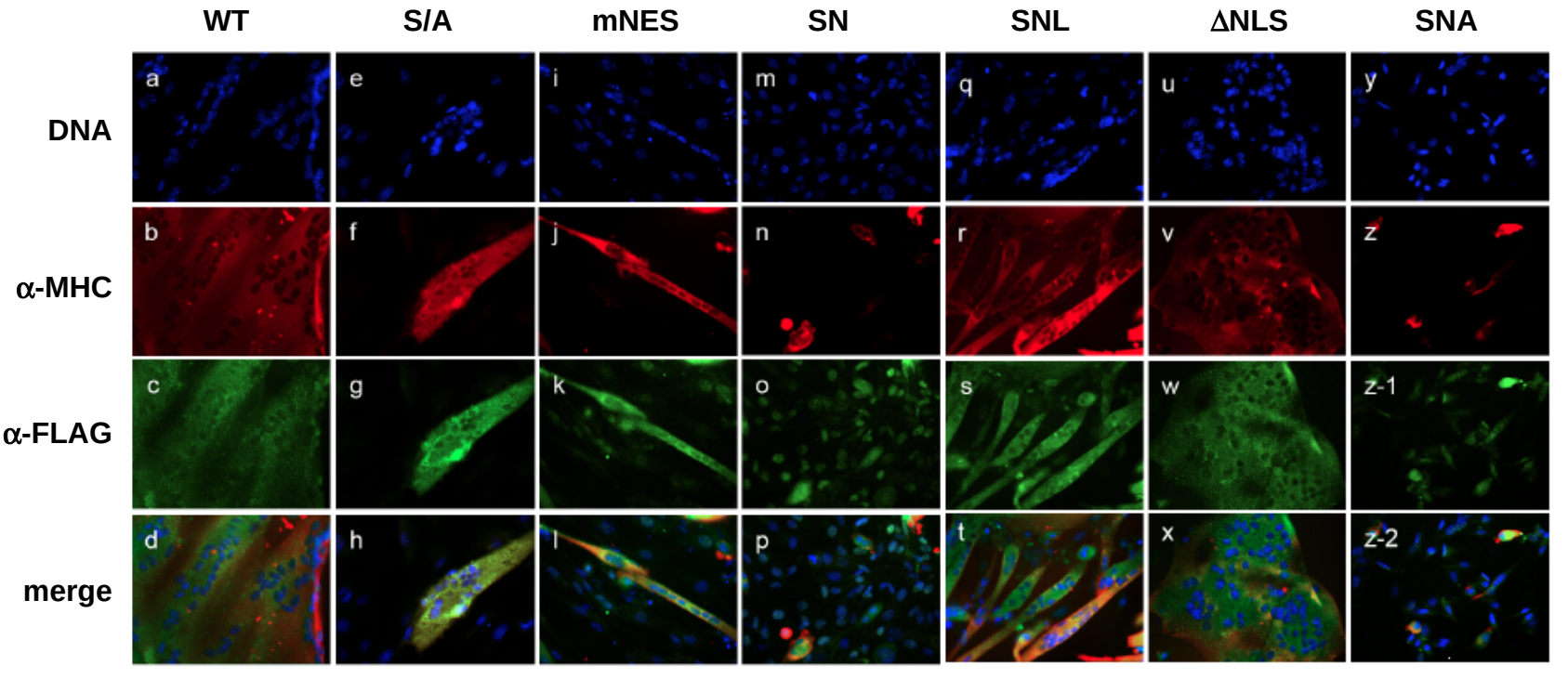
S/A
ΔNLS
mNES
SN
SNL
SNA
DAPI
α-FLAG
merge
a
d
g
j
m
p
s
b
e
h
k
n
q
t
c
f
i
l
o
r
u

Fig. 2A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S/A
mNES
SN
SNL
ΔNLS
SNA
DNA
α-MHC
α-FLAG
merge
a
e
i
m
q
u
y
b
f
j
n
r
v
z
c
g
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o
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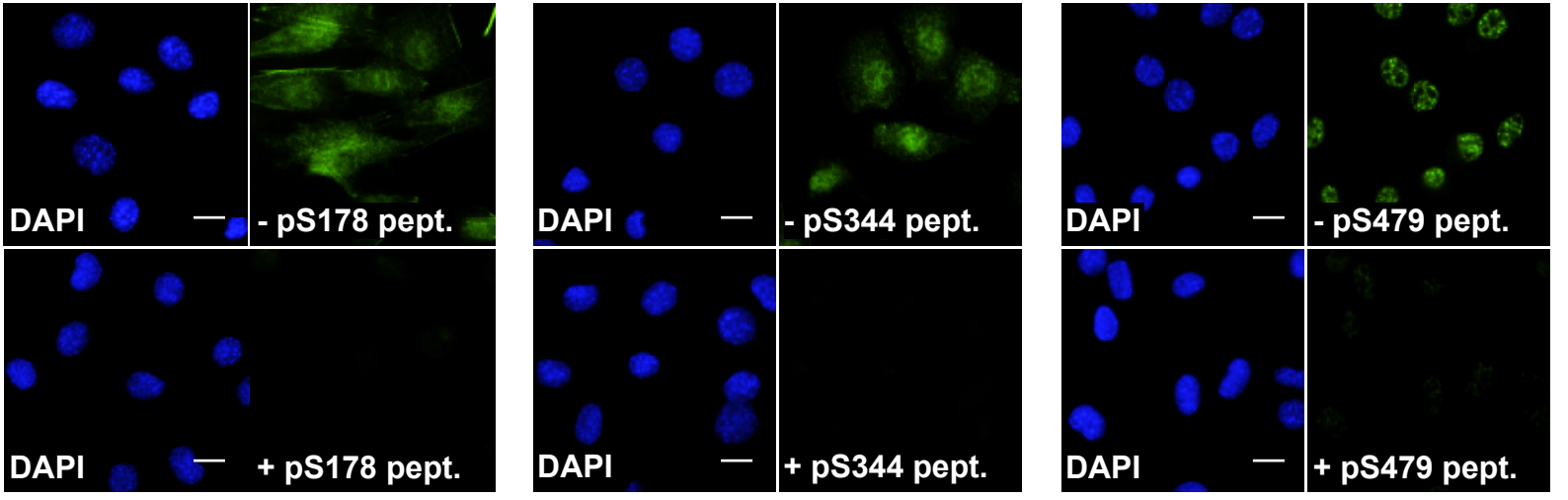
w
z-1
d
h
l
p
t
x
z-2

Fig. S2B



α-pS178 antibody

α-pS344 antibody

α-pS479 antibody

Fig. S3

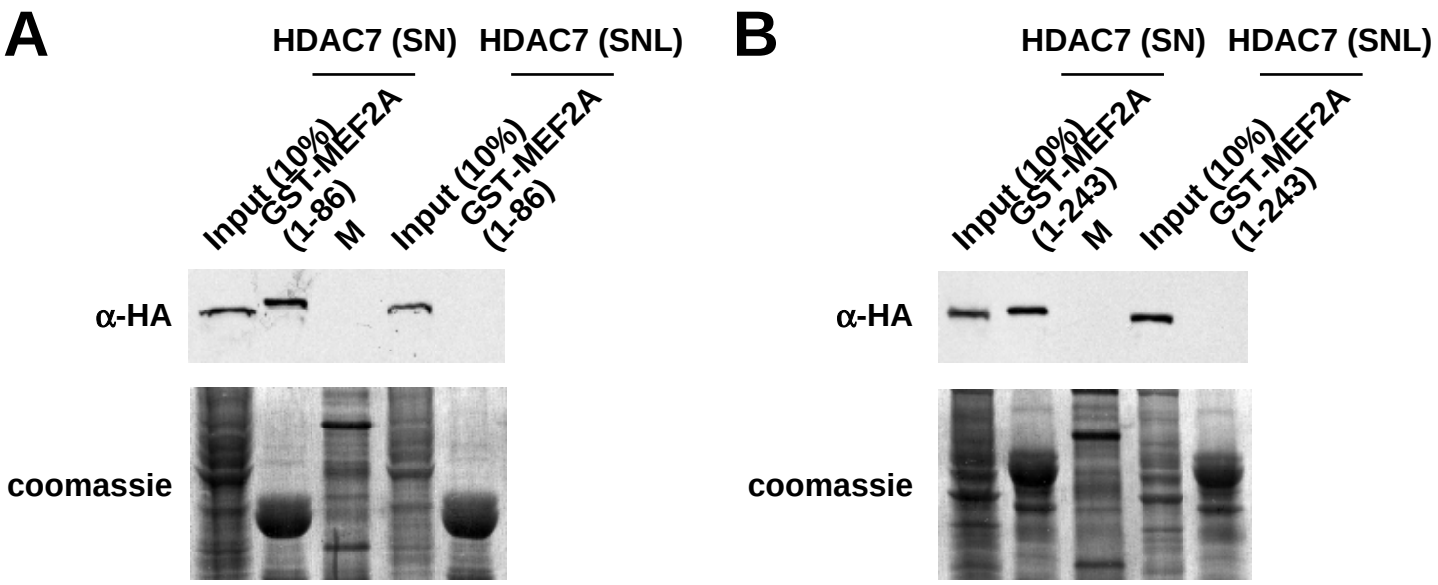

A
HDAC7 (SN)
HDAC7 (SNL)
Input (10%)
GST-MEF2A (1-86)
M
Input (10%)
GST-MEF2A (1-86)
α-HA
coomassie
B
HDAC7 (SN)
HDAC7 (SNL)
Input (10%)
GST-MEF2A (1-243)
M
Input (10%)
GST-MEF2A (1-243)
α-HA
coomassie

Fig. S4

Legends to Supplementary Figures

Fig. S1. The effect of overexpression of CRM1 on subcellular localization of HDAC7. An HA-CRM1 expression plasmid was co-transfected with YFP-HDAC7 (WT) (panels a-c) or YFP-HDAC7 (mNES) (panels d-f) expression plasmid followed by green fluorescence and immunofluorescence microscopy using anti-HA antibodies. Note that overexpression of CRM1 was capable of promoting cytoplasmic accumulation of wild-type HDAC7, but not the NES mutant (mNES). Scale bars: 20 μm.

Fig. S2. Subcellular distribution of exogenous FLAG-HDAC7 in C2C12 myocytes and myotubes. C2C12 cells were grown in GM or DM followed by immunofluorescence microscopy using anti-FLAG and anti-MHC antibodies as described in "Materials and Methods". (A) Subcellular distribution of stably transfected HDAC7 in undifferentiated myocytes. Note that subcellular distribution of the stably transfected wild-type HDAC7 (panels a-c) is more cytoplasmic than that of the transiently transfected wild-type protein. Scale bars: 20 μm. (B) Subcellular localization of stably transfected HDAC7 in C2C12 cells 4 days after cultured in differentiation medium.

Fig. S3. Specificity of phospho-specific antibodies against class IIa HDACs. The specificity of anti-pS178, pS344, and pS479 antibodies was examined according to our published protocol [1]. Phospho-specific antibodies were pre-incubated without (-) or with (+) phosphopeptides (pS178, pS344, or pS479 pept.) used to immunize the rabbits followed by incubation with C2C12 myocytes and fluorescence microscopy. DNA was stained by DAPI. Note that the reason that the staining patterns of pS344 and S479 differ from that in Figure 7 is likely due to differences in cell confluence. This figure shows staining of HDAC7 in cells about 50% confluence comparing to that in Figure 7, which is more than 90% confluence.

Figure S4. *In vitro* assays for the interaction between GST-MEF2A (1-86) and GST-MEF2A (1-243) and HDAC7 (SN) and HDAC7 (SNL). Immobilized, purified GST-MEF2A (1-86) (A) or GST-MEF2A (1-243) (B) fusion proteins were incubated with cell lysates expressing HA-HDAC7 (SN) or HA-HDAC7 (SNL). The pulldown fractions were subjected to Western blotting with anti-HA antibodies. M: marker.

REFERENCE

1. Gao, C., Li, X., Lam, M., Liu, Y., Chakraborty, S. and Kao, H.Y. (2006). CRM1 mediates nuclear export of HDAC7 independently of HDAC7 phosphorylation and association with 14-3-3s. FEBS Lett 580, 5096-104.