

1 **E08-04-0369 Jin**

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3 **Figure S1.** Activation of TORC1 transcriptional activity by MEKK1 in HEK293 cells.

4 HEK293 cells were transfected with the indicated plasmids as in Figure 1.

5

6 **Figure S2.** TORC1-mediated transcriptional activation is not mediated through ERK2,

7 JNK, p38, PKA or calcineurin. HeLa cells were treated with progressively increasing

8 amounts of the indicated inhibitors (U0126, SP600125, SB205380, H89 and CsA) as in

9 Figure 2.

10

11 **Figure S3.** MEKK1-induced phosphorylation of TORC1 in HEK293 cells. HEK293 cells

12 were transfected with the indicated plasmids and analyzed as in Figure 3.

13

14 **Figure S4.** MEKK1-induced and S167-independent phosphorylation of TORC1 in

15 HEK293 cells. HEK293 cells were transfected with the indicated plasmids and analyzed

16 as in Figure 4.

17

18 **Figure S5.** Mapping of an activation domain in TORC1. (A) A diagram of truncated

19 TORC1 mutants. (B) Influence of TORC1 and its mutants on CRE-dependent

20 transcription. HEK293T cells were transfected with reporter construct pCRE-Luc paired

21 with increasing amounts of empty pM vector (vec) or an expression vector for full-length

22 TORC1 (T1) or one of the TORC1 mutants.

23

1 **Figure S6.** Dominant interfering activity of GalTORC1M1 mutant. HeLa cells were
2 transfected with reporter plasmid pCRE-Luc, TORC1 expression vector and an
3 expression vector for V5-tagged TORC1 mutant M1 (V5M1; A) or Gal4 DNA-binding
4 domain-fused M1 (GalM1; B).

5

6 **Figure S7.** Repression of TORC1 mRNA expression by shRNAs. HeLa cells were
7 transfected with empty shRNA expression vector (vec) and expression plasmids for
8 shGFP, shTORC1a and shTORC1b, respectively, before treatment with 10 ng/ml IL-1 α
9 for 1 hr. TORC1 and GAPDH transcripts were analyzed by quantitative real-time RT-
10 PCR.

11

12 **Figure S8.** Influence of IL-1 α on the expression of TORC1 mRNA. HeLa cells were
13 transfected with expression plasmid for V5-tagged TORC1 (lanes 1-4) or TORC1 S167A
14 (lanes 5-8). Cells were treated with 10 ng/ml IL-1 α for the indicated duration of time.
15 TORC1 and GAPDH transcripts were analyzed by semi-quantitative RT-PCR (A) or
16 quantitative real-time RT-PCR (B).

17

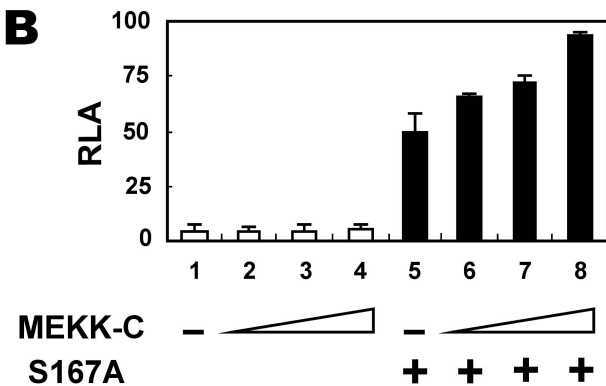
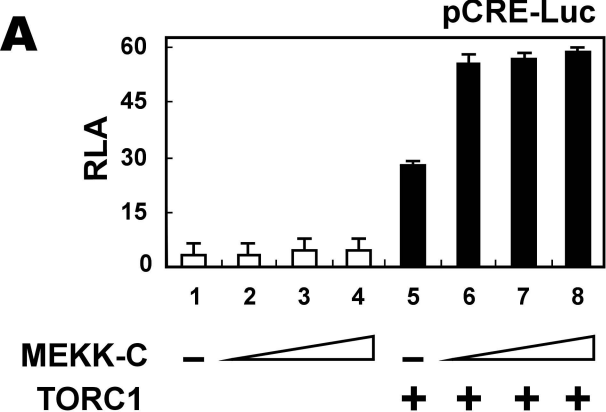


Figure S1

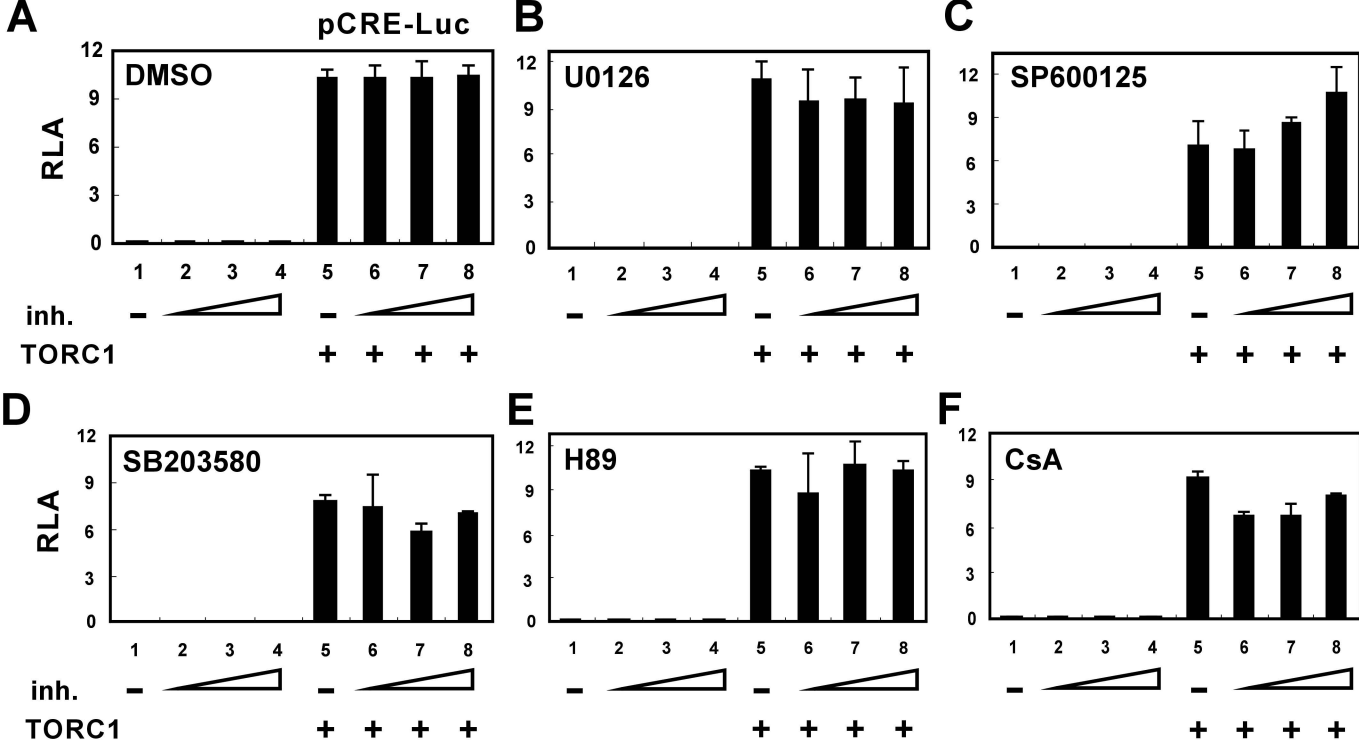


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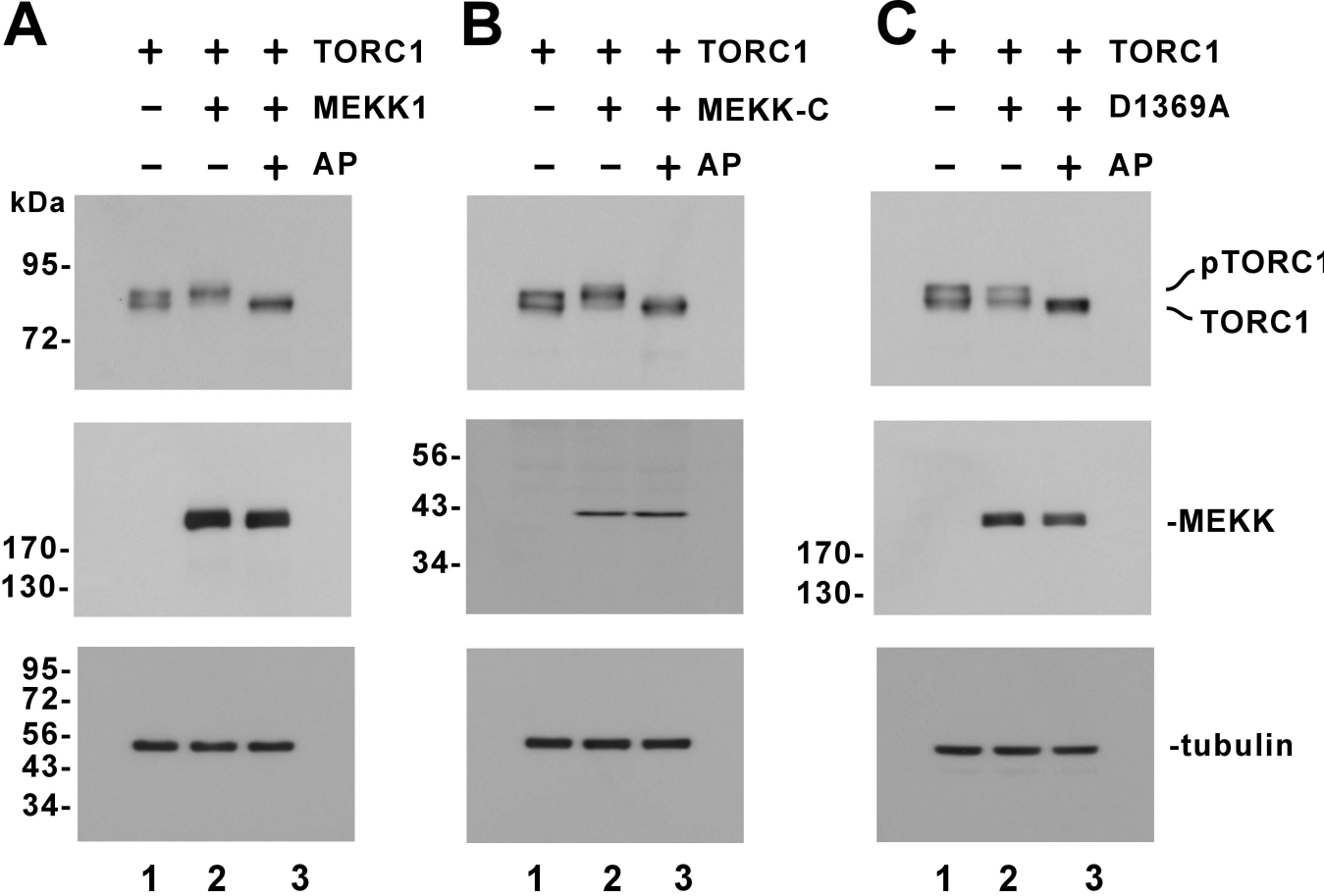


Figure S3

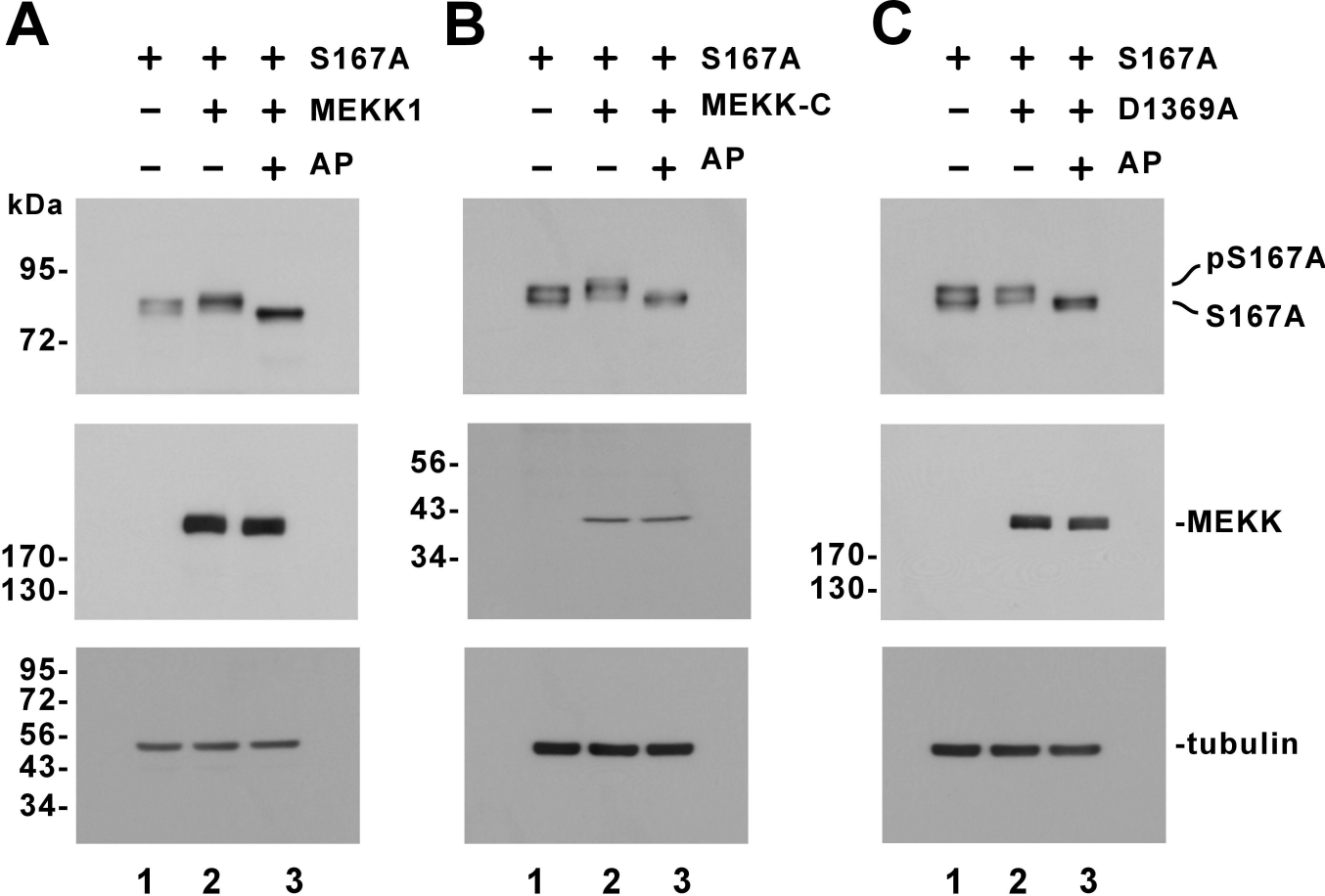


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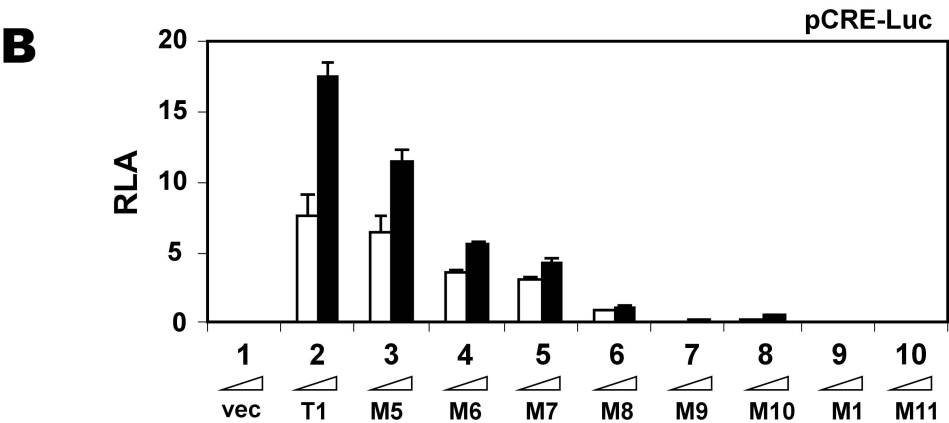
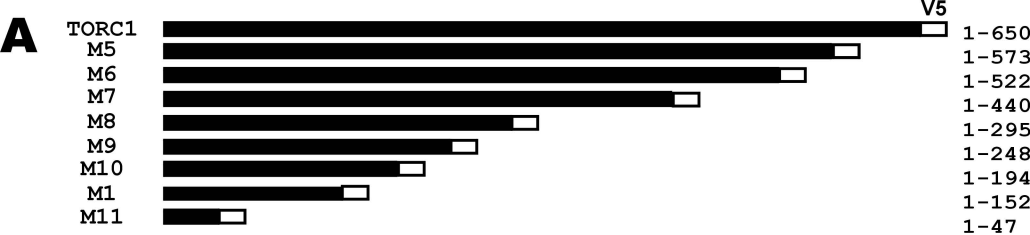


Figure S5

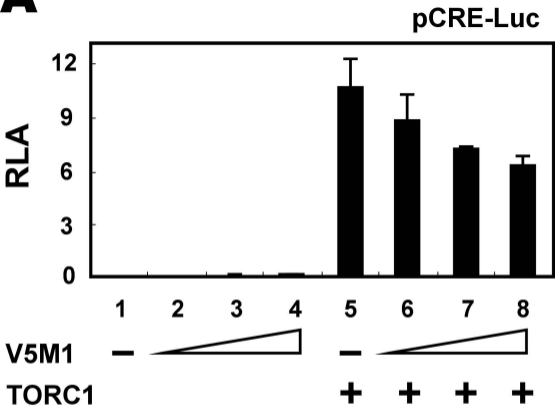
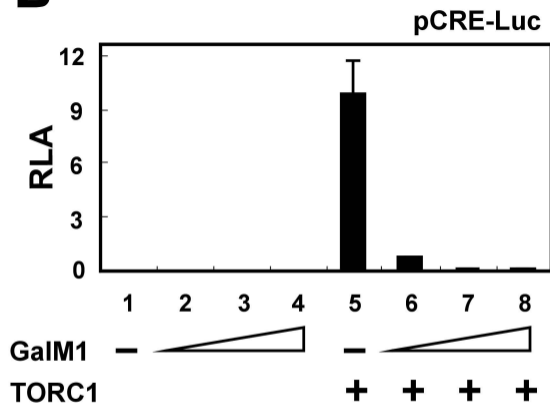
A**B**

Figure S6

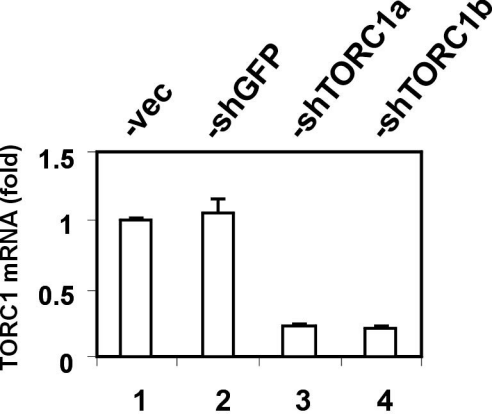
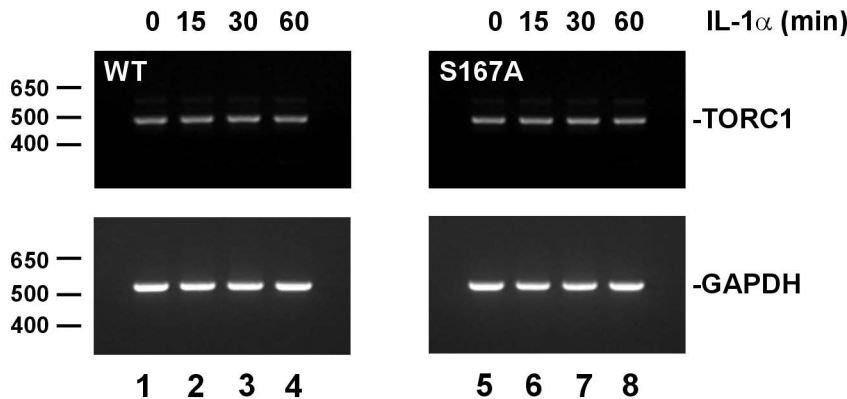
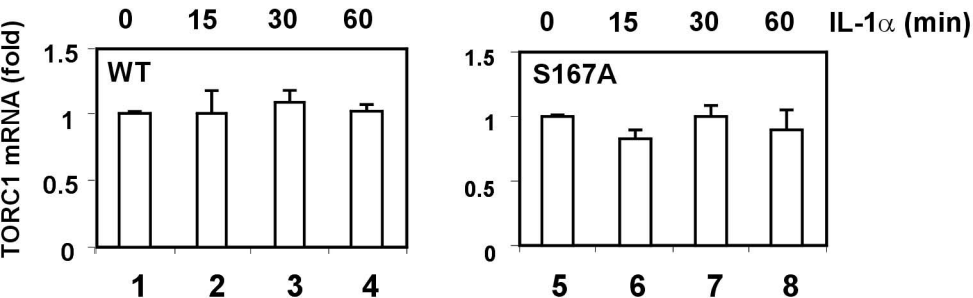


Figure S7

A**B****Figure S8**