

Molecular Pharmacology
Supplemental figures

Role of JunB in Adenosine A_{2B} Receptor-mediated VEGF Production

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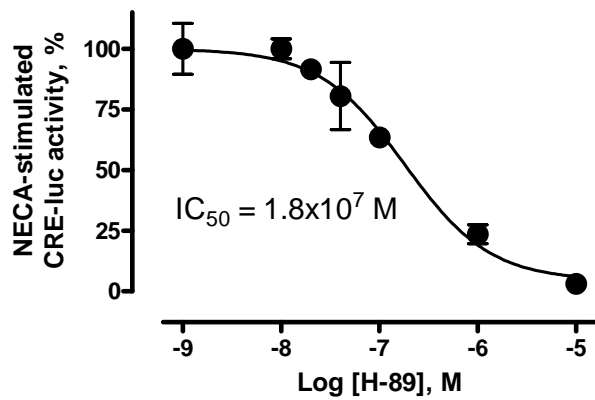
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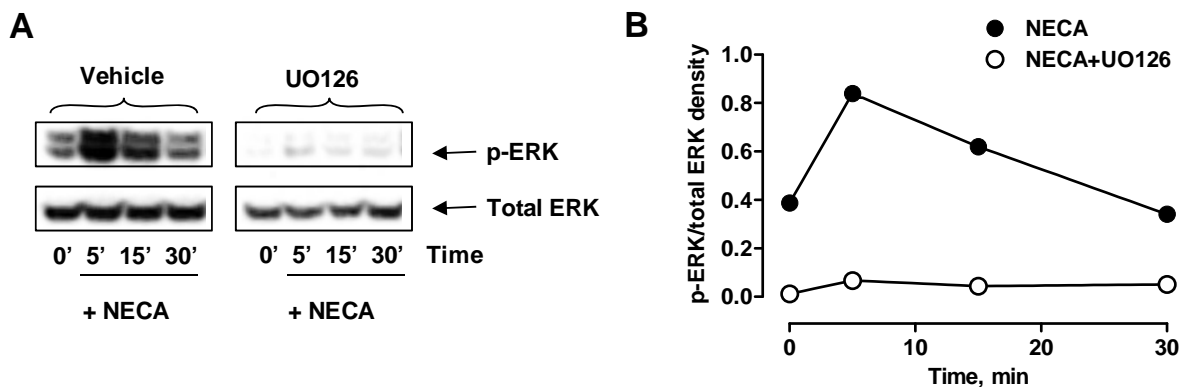
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Supplemental Figure 1. *Effect of the protein kinase A inhibitor H-89 on NECA-induced cAMP response element (CRE)-driven luciferase reporter activity in HMEC-1 cells.*

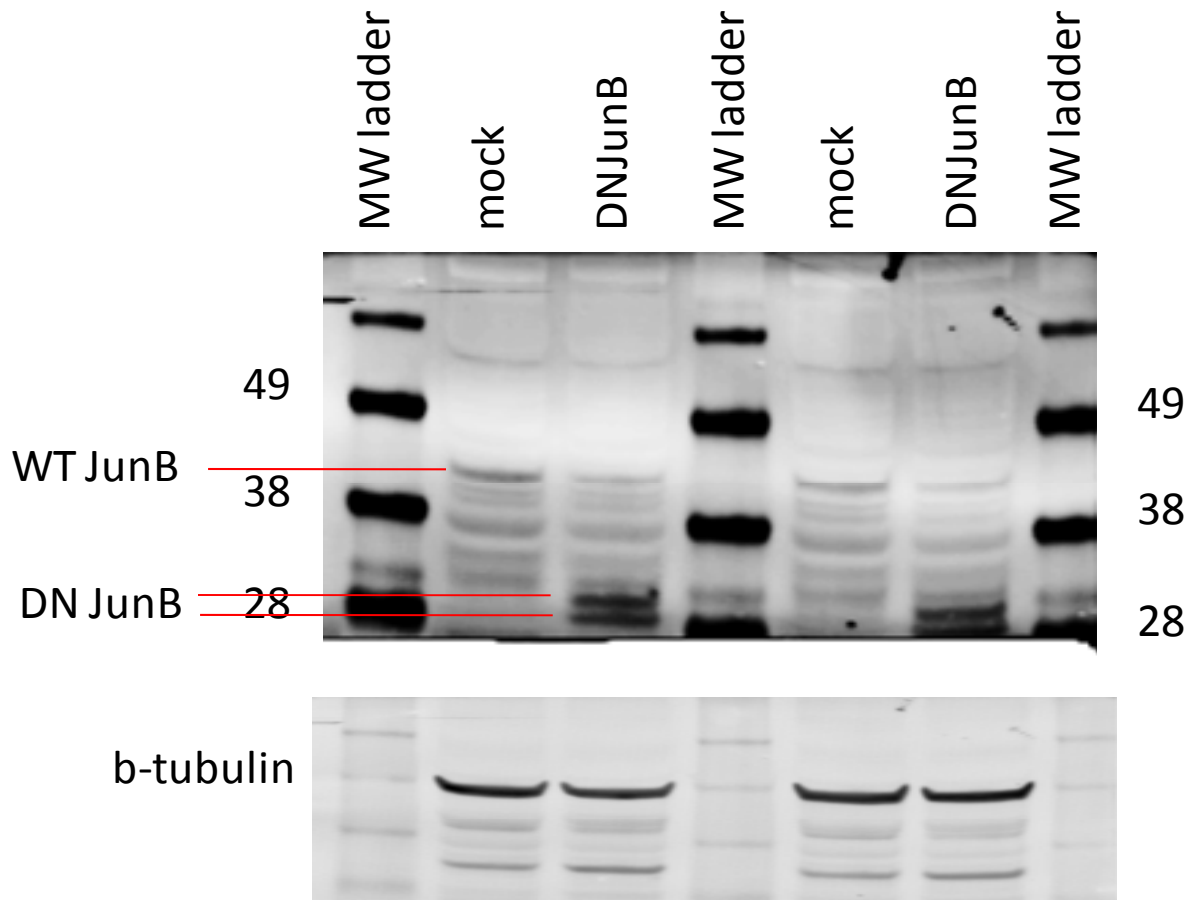
HMEC-1 cells transfected with a CRE reporter were incubated in the absence or presence of 10 μ M NECA in the absence or presence of increasing concentrations of H-89 for 6 hours. H-89 was added to cells 30 min before addition of NECA. Potency of H-89 on cAMP/PKA signaling transduction in HMEC-1 cells was determined as IC_{50} from concentration-response curve of H-89 effects on NECA-stimulated CRE luciferase reporter activity. Values are presented as mean \pm SEM (n=3).



Supplemental Figure 2. Effect of the MEK inhibitor UO126 on NECA-induced ERK activation in HMEC-1 cells.

A, Time course of ERK activation by NECA in HMEC-1 cells in the absence (Vehicle) or presence of 1 μ M UO126. Cells were stimulated with 10 μ M NECA for indicated periods of time. ERK activation was determined by immunoblotting with antibody specific for phosphorylated ERK. Total ERK was used as a loading control. A representative blot of 3 experiments is shown.

B, Levels of phosphorylated ERK quantified from Western blot data by densitometry and expressed as percentage of corresponding levels in resting cells in the absence of UO126 normalized to total ERK levels.



Supplemental Figure 3. Expression of JunB and DNJunB in LLC cells stably transfected with plasmid encoding DNJunB or with an empty vector (mock).

Western blot analysis was performed using antibodies against a common epitope in the sequences of JunB and DNJunB. Molecular weight range is shown on both sides of gel. Red lines indicate positions of JunB and DNJunB. Immunostaining of β -tubulin was used as a loading control.