RNA-binding protein RBM3 prevents NO-induced apoptosis in human neuroblastoma cells by modulating p38 signaling and miR-143

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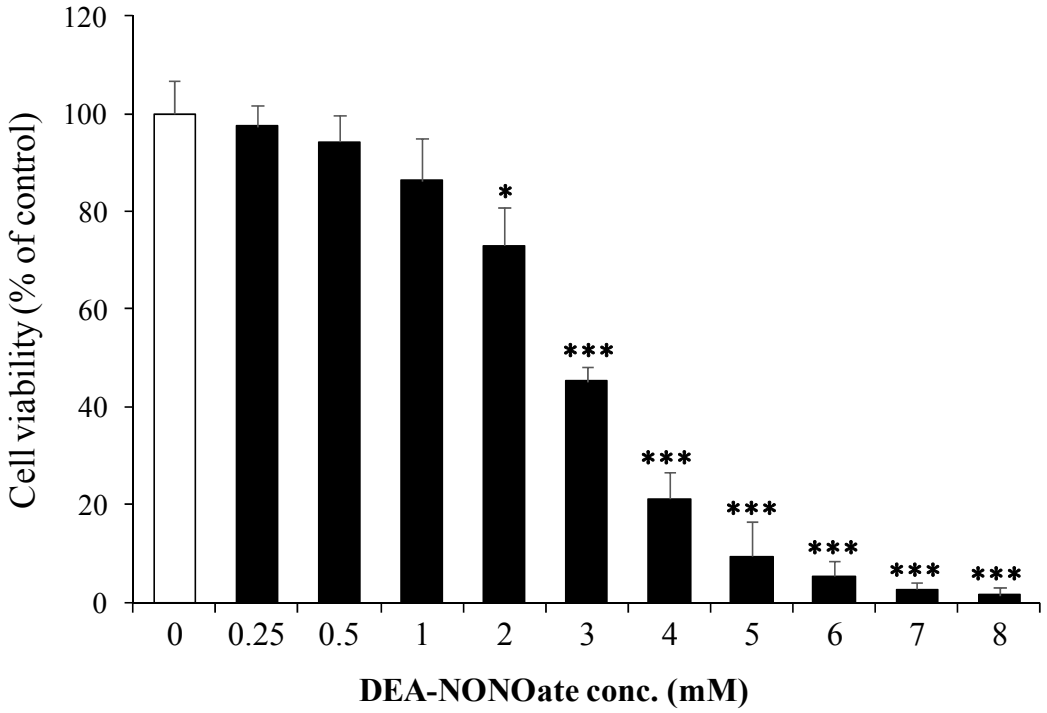

Figure S1. DEA NONOate induces a dose-dependent cytotoxicity in SH-SY5Y neuroblastoma cells. Cells were treated various concentrations of DEA NONOate for 16 h, and cell viability was assessed by MTT assay. *$P < 0.05$ and ***$P < 0.001$ versus control group. All results shown are representative of three independent experiments.

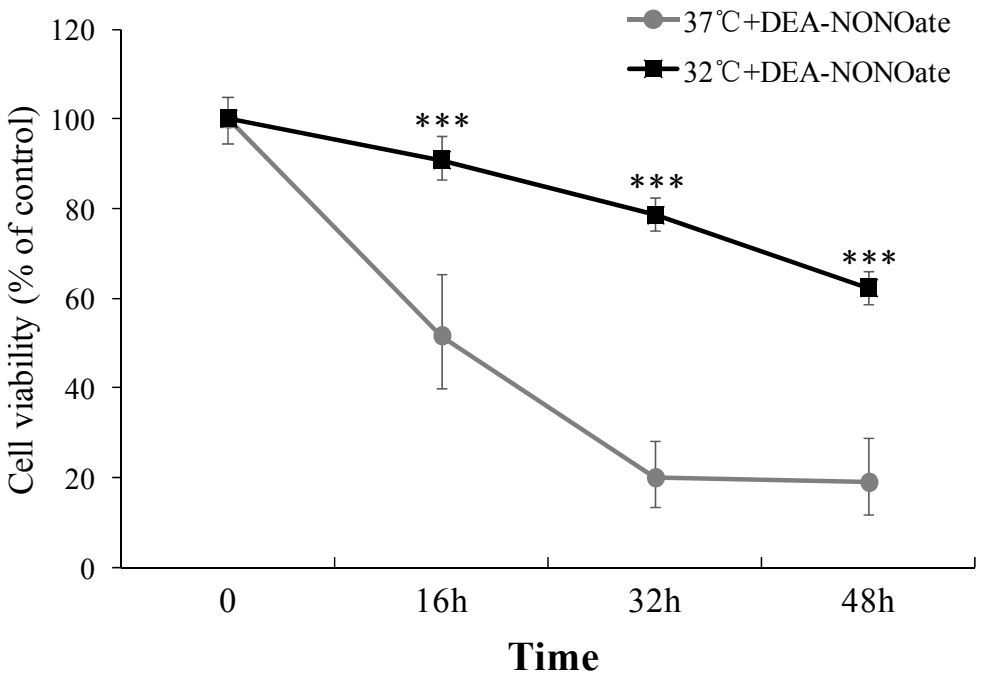

Figure S2. DEA NONOate induces a time-dependent cytotoxicity in SH-SY5Y cells and mild hypothermia (32ºC) reduces its cytotoxicity. SH-SY5Y cells were pre-cultured under normothermic (37ºC) or mild hypothermic (32ºC) conditions for 1 d and treated with DEA NONOate (3 mM) for 16−48 h, and then cell viability was assessed by MTT assay. ***$P < 0.001$ versus control group.

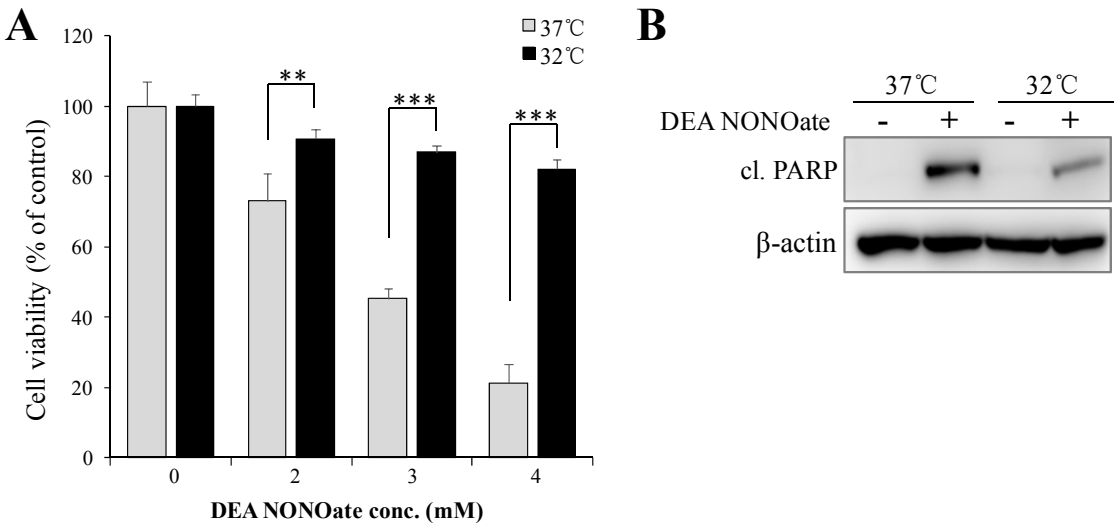

Figure S3. Mild hypothermia prevents SH-SY5Y neuroblastoma cells from DEA NONOate-induced apoptosis. SH-SY5Y cells were pre-cultured under normothermic (37ºC) or mild hypothermic (32ºC) conditions for 1 d and treated with DEA NONOate (3 mM) for 16 h. (**A**) The cell viability was assessed by MTT assay, and (**B**) cleaved (cl.) PARP was detected by Western blotting. β-actin served as a loading control. $^{**}P < 0.01$ and $^{***}P < 0.001$ versus control group.

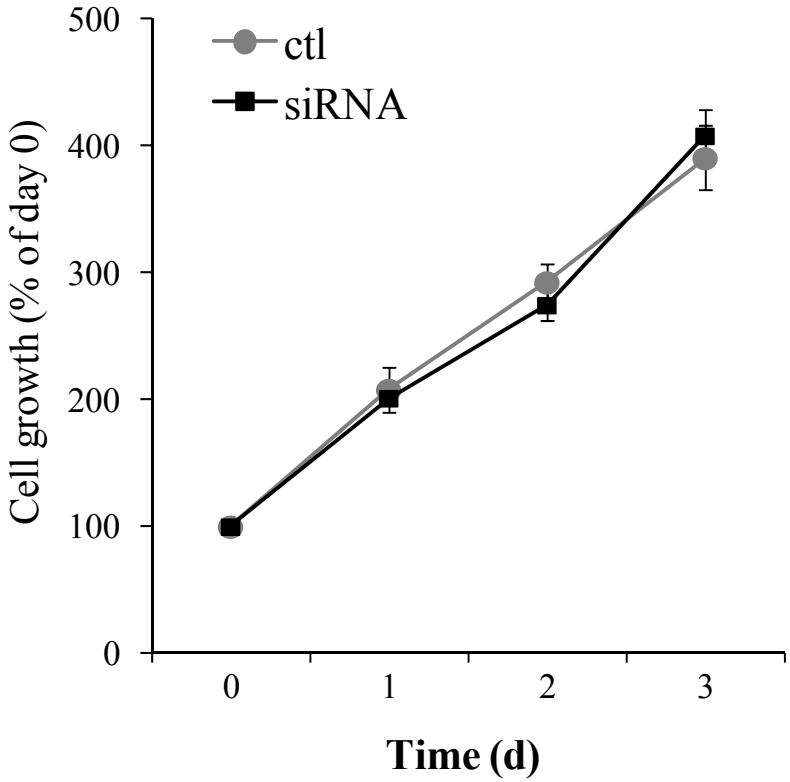

Figure S4. Effect of siRNAs on cell growth of SH-SY5Y under hypothermic conditions. 2 d post transfection of RBM3 siRNA or scramble siRNA (ctl), SH-SY5Y cells were continued to culture at 32ºC for 1−3 d, and MTT assay was performed to evaluate cytotoxicity of siRNAs under hypothermic conditions.

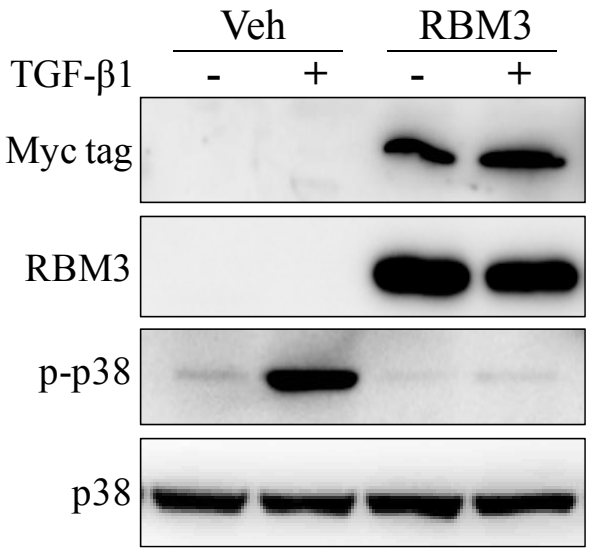

Figure S5. RBM3 inhibits TGF-β1-induced p38 activation. SH-SY5Y cells were transfected with plasmid pXJ40-myc (Veh) or pXJ40-myc-RBM3 (RBM3) for 2 d, and then treated with TGF-β1 (5 ng/mL) for 30 min. Western blotting was performed to detect the levels of phosphorylated p38 (p-p38). The total p38 served as a loading control.