Supporting Information

Table S1. The dose-response of EpEX on MSC doubling time

EpEX (µg/mL)	0	0.3	1	3	6	9
Average doubling time	32.4	28.1	26.8	21.1	26.6	26.4
SEM	1.15	1.44	2.12	0.74	1.12	1.97

Figure S1

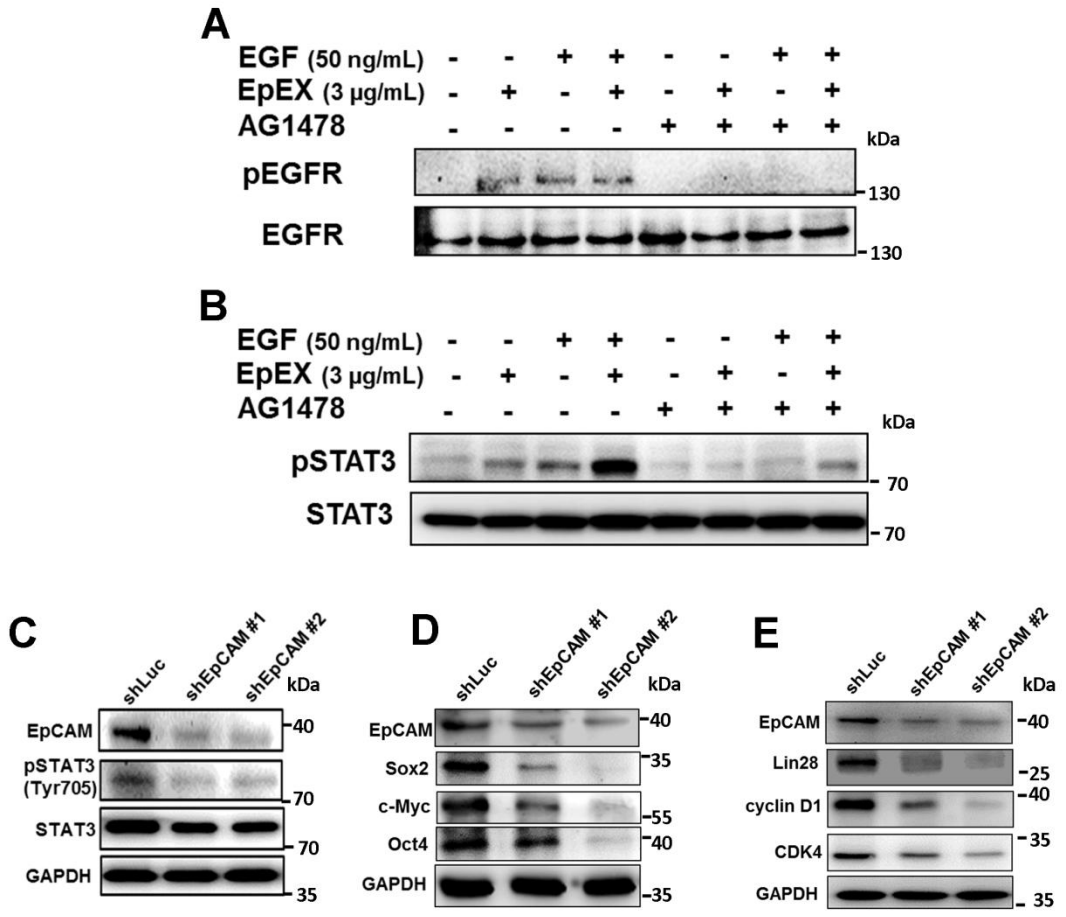

Figure S1. EpEX and EGF induce the phosphorylation of EGFR and STAT3, and EpCAM is crucial for maintaining expression of cell cycle regulators and stemness markers. MSCs were pretreated with or without an EGFR inhibitor, AG1478, and then treated with EGF, EpEX, or co-treated with EGF and EpEX at indicated time. The phosphorylation of EGFR (Tyr845) (A) and STAT3 (Tyr705) (B) were examined by Western blotting. The inhibition of EpCAM significantly decreases phospho-STAT3, cell cycle regulators, and stemness markers. MSCs were made to express EpCAM shRNA, and the protein levels of phosphorylation of STAT3, total STAT3 (C), the pluripotency factors (Sox2, Oct4, c-Myc and EpCAM) (D) and cell cycle regulators (cyclin D and CDK4) (E) were detected by Western blotting.

Figure S2

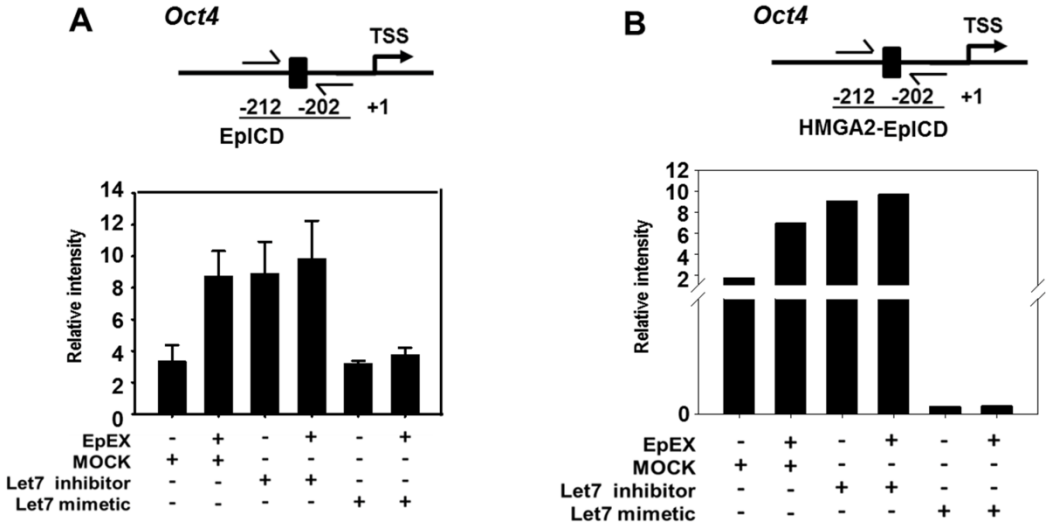

Figure S2. **EpEX increases the binding of EpICD to the promoter of Oct4 by inhibiting let7.** (A) MSCs were transfected with let7 inhibitor or mimetics then treated with EpEX (3 μg/mL). Binding of EpICD to the Oct4 promoter was examined by chromatin immunoprecipitation (ChIP). EpICD was pulled down by a specific anti-EpICD antibody. The cross-linked DNA was isolated and then probed by qPCR with specific primers for the Oct4 promoter. (B) MSCs were transfected with let7 inhibitor or mimetics then treated with EpEX (3 μg/mL). Binding of EpICD-HMGA2 was examined by sequential ChIP. EpICD was pulled down by a specific anti-EpICD antibody, followed by pull-down with a HMGA2 antibody. To detect bound Oct4 promoter, the cross-linked DNA was isolated and then amplified by qPCR with specific primers.

Figure S3

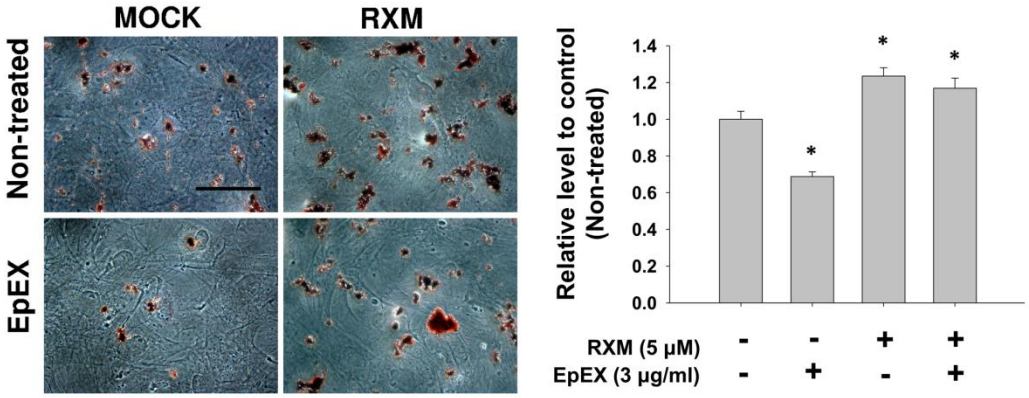

Figure S3. **EpEX inhibits MSC adipogensis.** To detect adipogenesis, MSCs were cultured in adipogenic induction medium for 21 days and then stained with Oil Red O. Scale bar: 100 μm (Left panel). The quantification results of the Oil Red O staining (Right panel). Data represent the mean ± SD. *P < 0.05, compared with control without RXM and EpEX group.

Figure S4

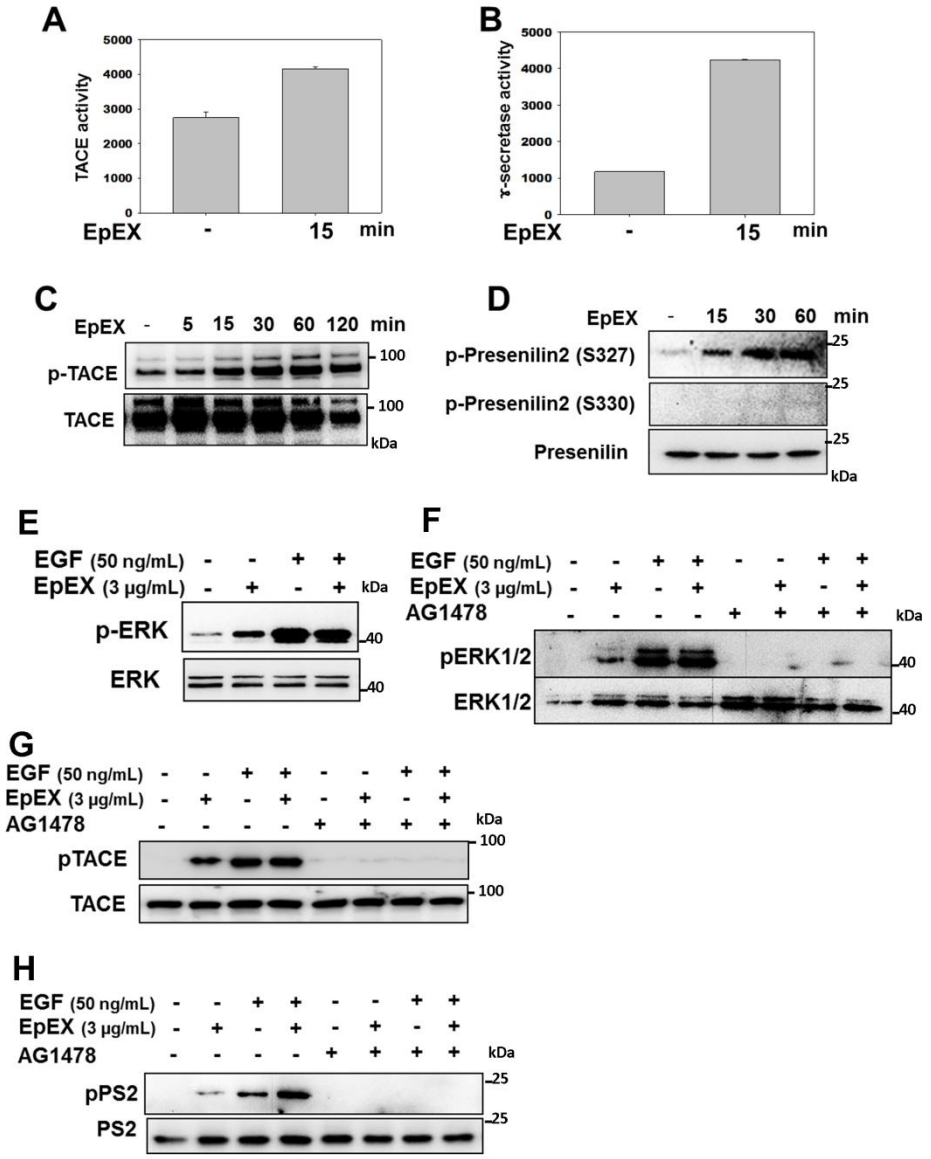

Figure S4. EpEX induces the phosphorylation and activity of TACE and γ-secretase. MSCs were stimulated by EpEX for the indicated times, and the activity of TACE (A) and γ-secretase (B) were detected. MSCs were stimulated by EpEX (3 μg/mL) for the indicated times. Western blot analysis was performed to detect the phosphorylation of TACE (C) and Presenilin 2 (D). MSCs were treated with EGF, EpEX, or co-treated with EGF and EpEX for 5 min. The phosphorylation of ERK1/2 (E) was examined by Western blotting with specific antibodies. MSCs were pretreated with or without an EGFR inhibitor, AG1478, and then treated with EGF, EpEX, or co-treated with EGF and EpEX for indicated times. The phosphorylation of ERK1/2 (F), TACE (Ser435)(G), PS2 (Ser327)(H) were examined by Western blotting with specific antibodies.

Figure S5

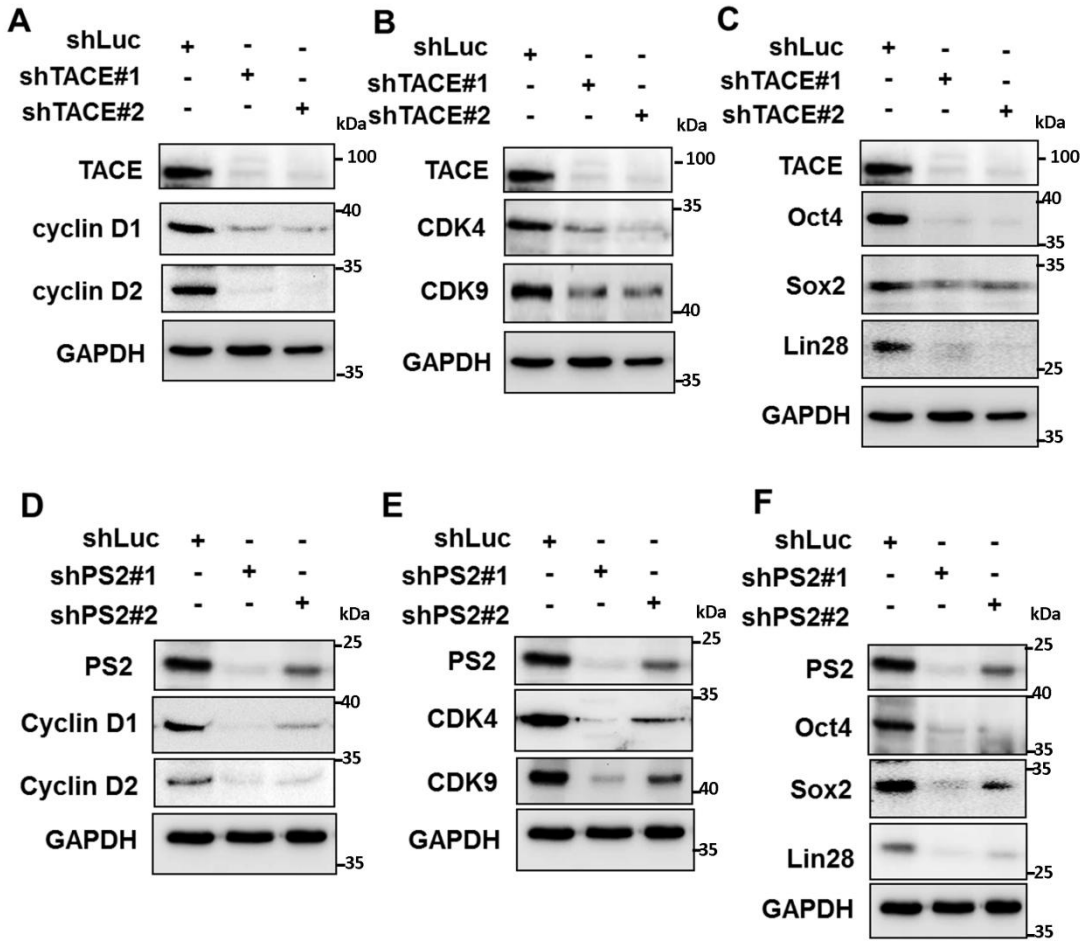

Figure S5. **TACE and presenilin 2 are crucial for the expression of cell cycle regulators and pluripotent markers.** (A, B, C) In MSCs, TACE was knocked down and the levels of cell cycle regulators and pluripotency markers were examined by Western blotting with specific antibodies. The results of figure S5A, S5B and S5C were from one and the same experiment and showed separately because of the size of the image; therefore S5A, S5B and S5C were with the same TACE and GAPDH. (D, E, F) In MSCs, presenilin 2 was knocked down and the levels of cell cycle regulators and pluripotency markers were examined by Western blotting with specific antibodies. The results of figure S5C, S5D and S5E were from one and the same experiment and showed separately because of the size of the image; therefore S5D, S5E and S5F were with the same PS2 and GAPDH.

Figure S6

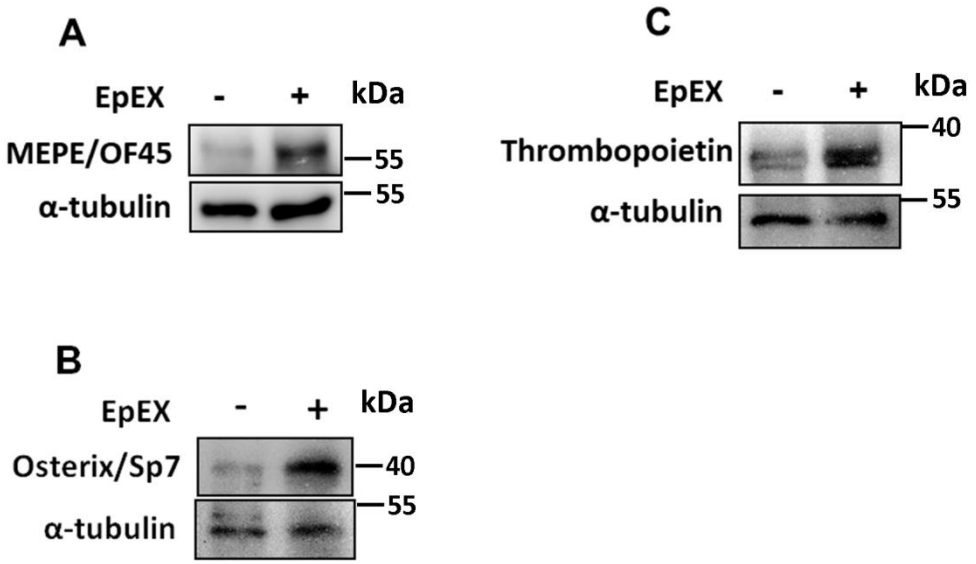

Figure S6. EpEX induces the expression of osteoprogenitor markers. MSCs were treated with or without EpEX and incubated in induction medium for osteogenesis. The osteoprogenitor markers, (A) MEPE/OF45, (B) Osterix/Sp7 and (C) thrombopoietin, were detected by Western blotting.

Figure S7

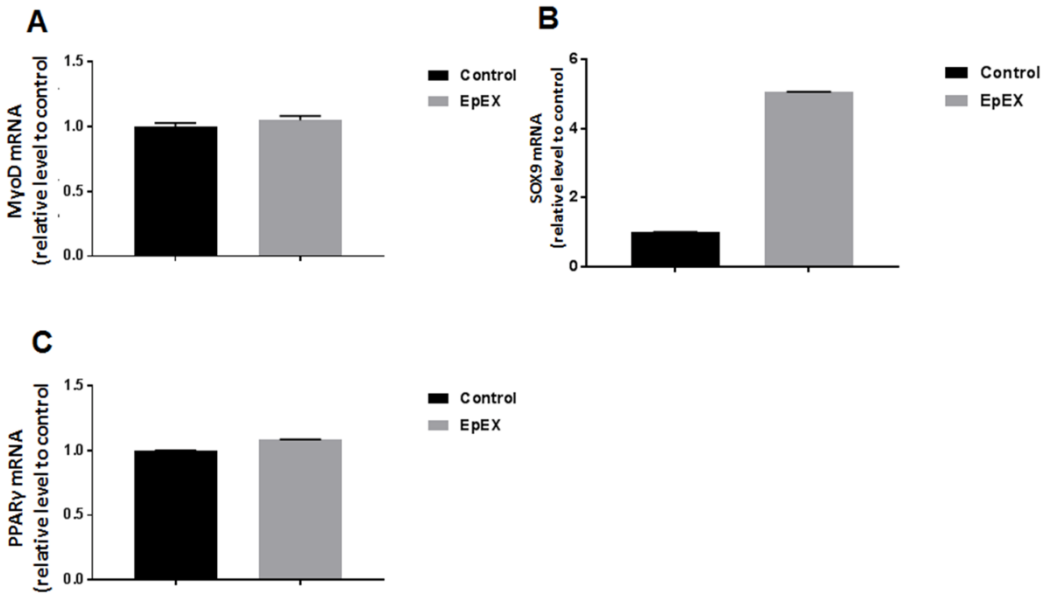

Figure S7. EpEX increases the mRNA expression of chondrogenesis marker, SOX9, but has no significant effect on MyoD and PPARγ. MSCs were treated with (gray bar) or without EpEX (black bar) and incubated in induction medium for myogenesis (A), chondrogenesis (B), or adipogenesis (C). At day 14, the mRNA expression levels of MyoD (A), SOX9 (B), PPARγ (C) was detected by specific primers and qPCR.

Figure S8

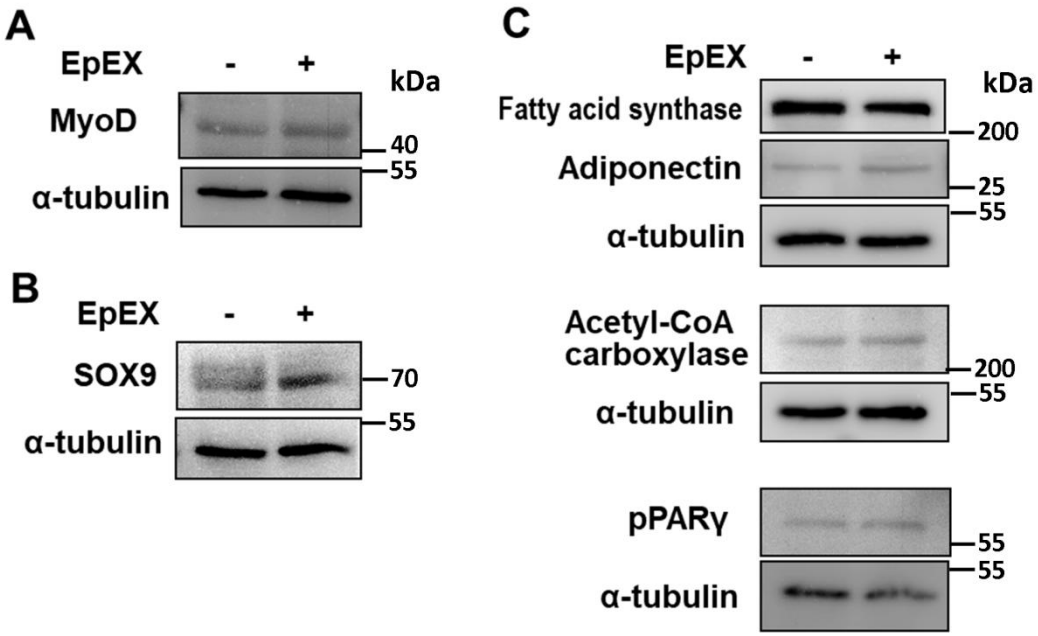

Figure S8. The impact of EpEX on expression of MSC lineage differentiation markers. MSCs were treated with or without EpEX and incubated in induction medium for (A) myogenesis, (B) chondrogenesis and (C) adipogenesis. After 14 days of induction, differentiation was analyzed. All experiments were carried out in triplicate. (A) Myogenesis marker: MyoD, (B) chondrogenesis marker: SOX9, and (C) adipogenic markers: Fatty acid synthase, Adiponectin, Acetyl-CoA carboxylase, and PPARγ, were detected by Western blotting.

Figure S9

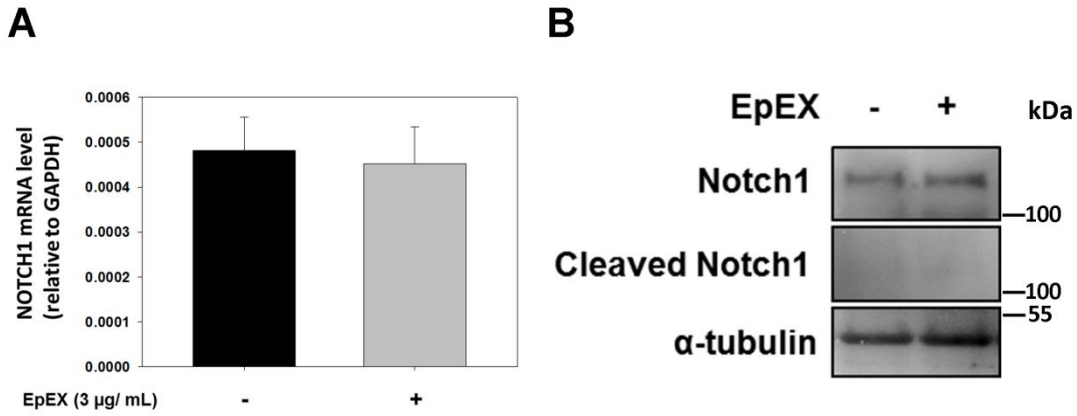

Figure S9. EpEX has no significant impact on the gene and protein expression of Notch1 and cleaved Notch1. MSCs were pretreated with or without EpEX (3 μg/mL) for 6 h, after which gene expression were examined by qPCR, and the protein expression of Notch1 and cleaved Notch1 were examined by Western blotting.