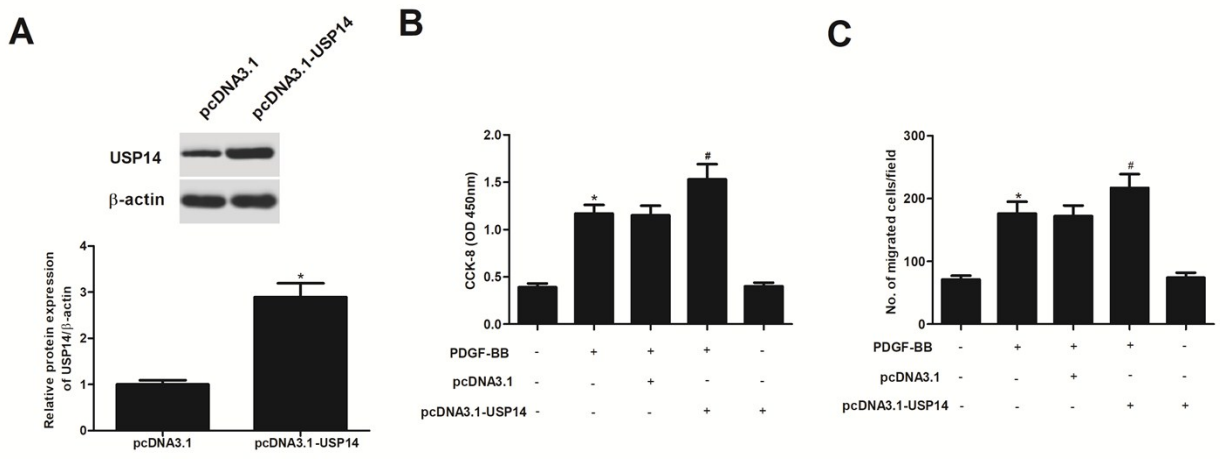
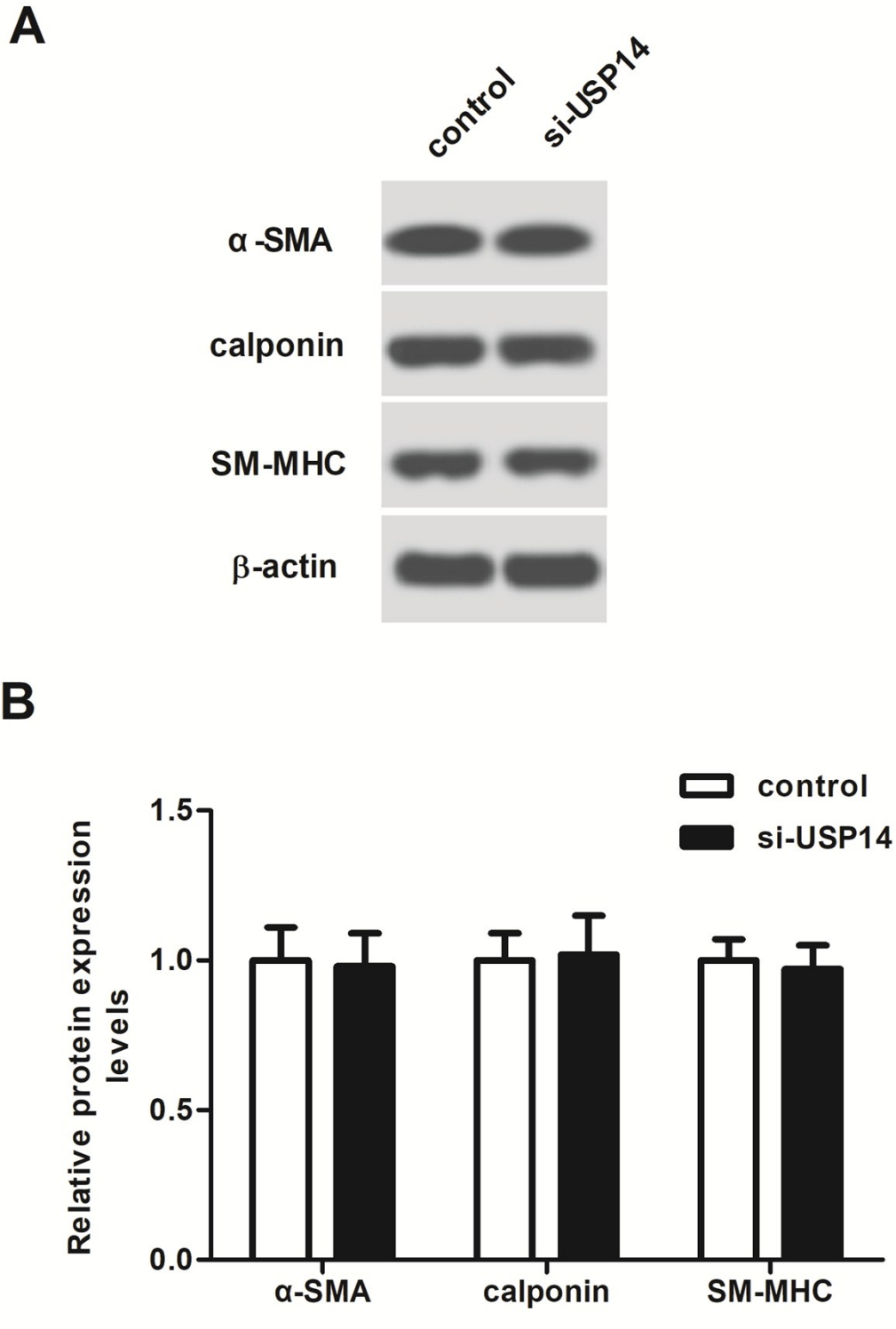
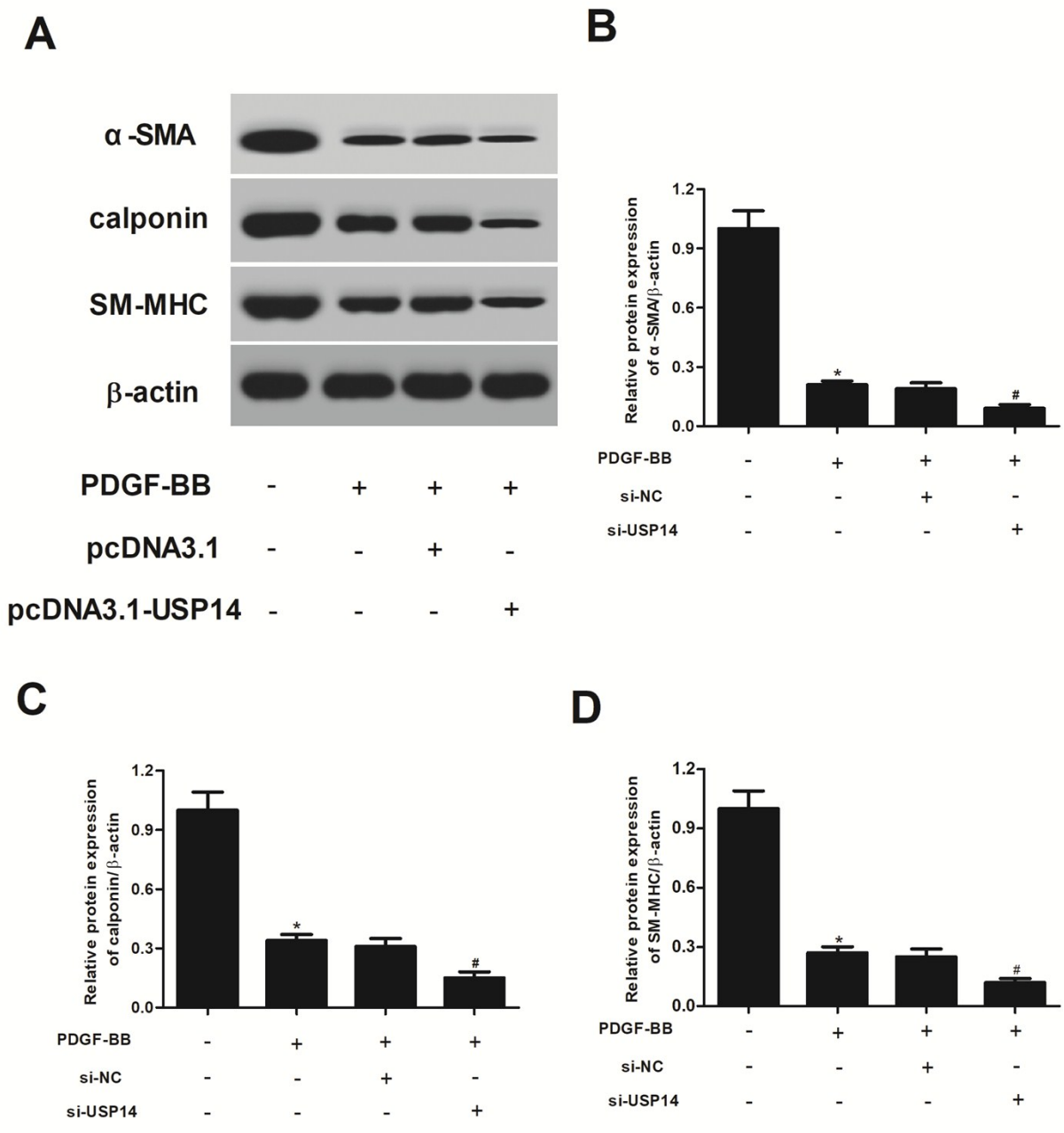


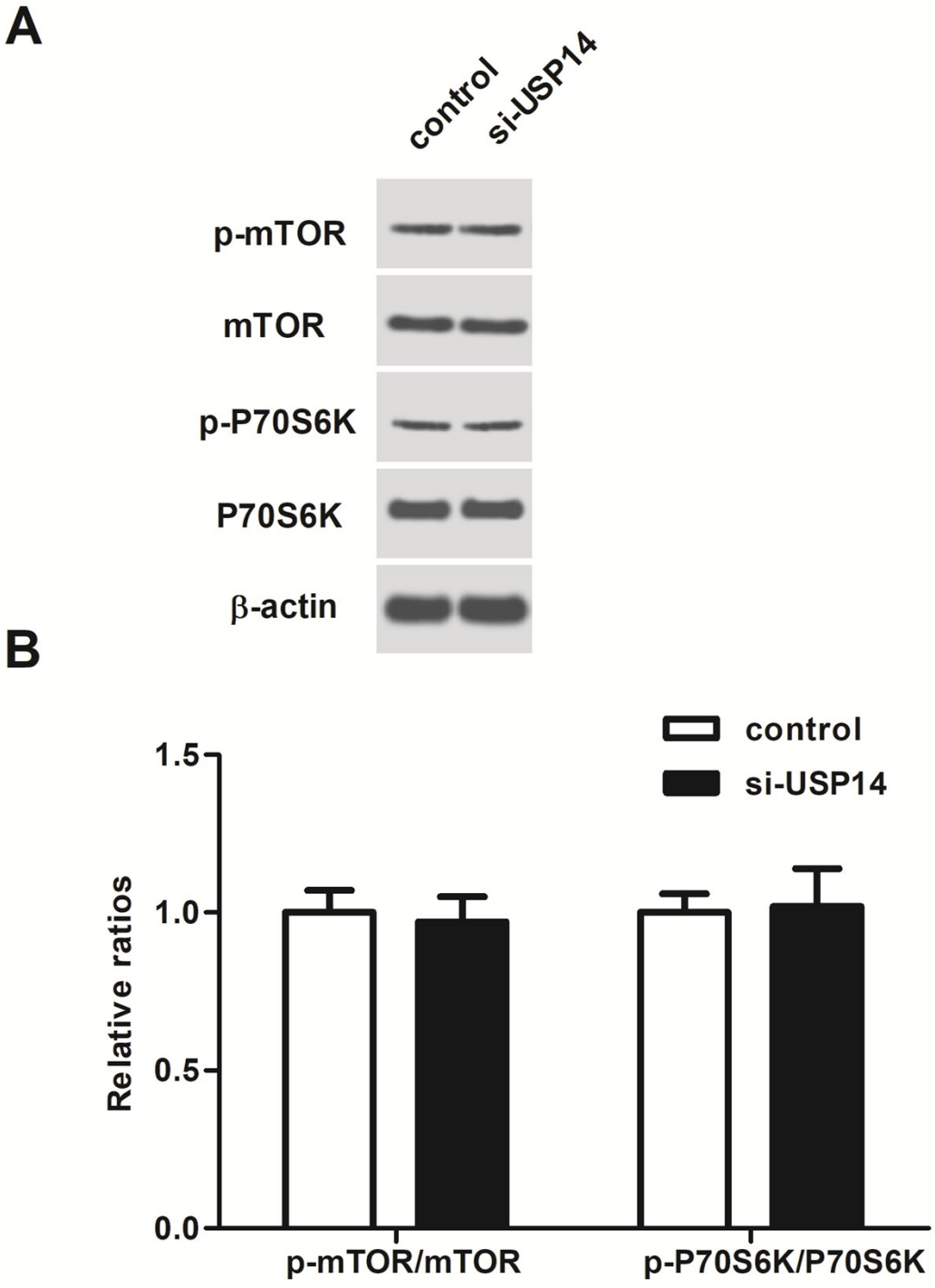
Supplementary Figure 1. Overexpression of USP14 promotes the proliferation and migration in PDGF-BB-induced HASMCs. (A) Western blot analysis was performed to detect the USP14 expression after transfection with pcDNA3.1 or pcDNA3.1-USP14. *$p < 0.05$ vs. pcDNA3.1 group. HASMCs were transfected with pcDNA3.1 or pcDNA3.1-USP14 in the presence of PDGF-BB (40 ng/ml) for 24 h; or only transfected with pcDNA3.1-USP14 for 24 h. (B) CCK-8 was performed to evaluate cell proliferation of HASMCs. (C) Transwell assays were carried out to evaluate cell migration of HASMCs. *$p < 0.05$ vs. control group; #$p < 0.05$ vs. pcDNA3.1+PDGF-BB group.


A
control
si-USP14
α -SMA
calponin
SM-MHC
β-actin
B
control
si-USP14
Relative protein expression levels
1.5
1.0
0.5
0.0
α-SMA
calponin
SM-MHC

Supplementary Figure 2. Effect of si-USP14 on the expression levels of VSMCs markers in HASMCs. HASMCs were transfected with si-USP14 for 24 h. (A) The expression levels of VSMCs markers including α-SMA, calponin and SM-MHC were measured using western blot analysis. (B) Quantification analysis of α-SMA, calponin and SM-MHC.

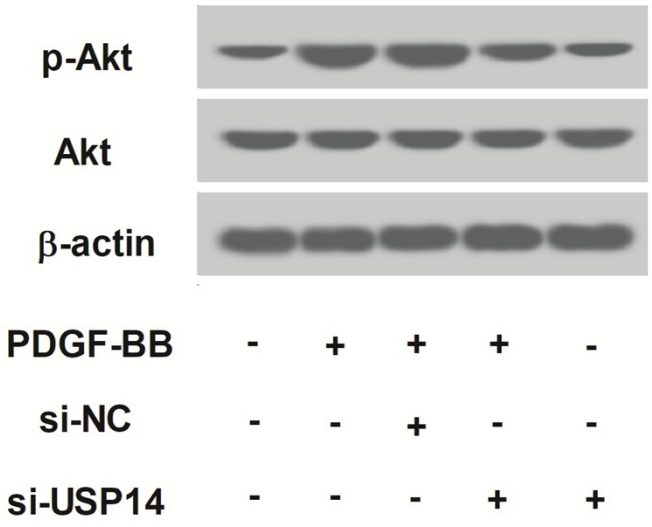


Supplementary Figure 3. Overexpression of USP14 inhibits the expression levels of VSMCs markers in PDGF-BB-stimulated HASMCs. HASMCs were transfected with pcDNA3.1 or pcDNA3.1-USP14 in the presence of PDGF-BB (40 ng/ml) for 24 h. (A) The expression levels of VSMCs markers including α-SMA, calponin and SM-MHC were measured using western blot analysis. (B-D) Quantification analysis of α-SMA, calponin and SM-MHC. *$p < 0.05$ vs. control group; #$p < 0.05$ vs. pcDNA3.1+PDGF-BB group.

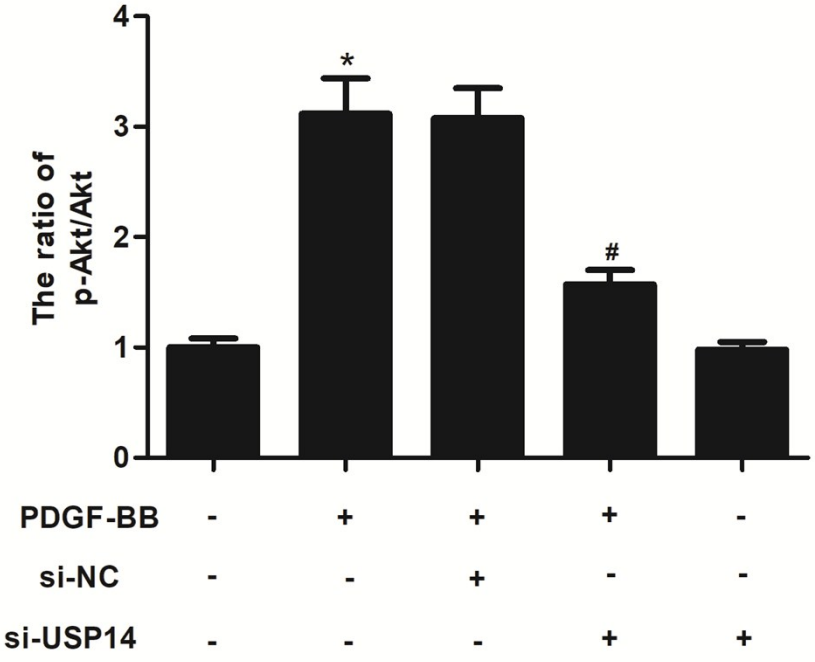

A
control
si-USP14
p-mTOR
mTOR
p-P70S6K
P70S6K
β-actin
B
control
si-USP14
Relative ratios
1.5
1.0
0.5
0.0
p-mTOR/mTOR
p-P70S6K/P70S6K

Supplemetary Figure 4. Effect of si-USP14 on mTOR/P70S6K signaling pathway in HASMCs. HASMCs were transfected with si-USP14 for 24 h. (A) The expression levels of mTOR, P70S6K, p-mTOR and p-P70S6K were detected using western blot analysis. (B) Quantification analysis of p-mTOR/mTOR and p-P70S6K/P70S6K.

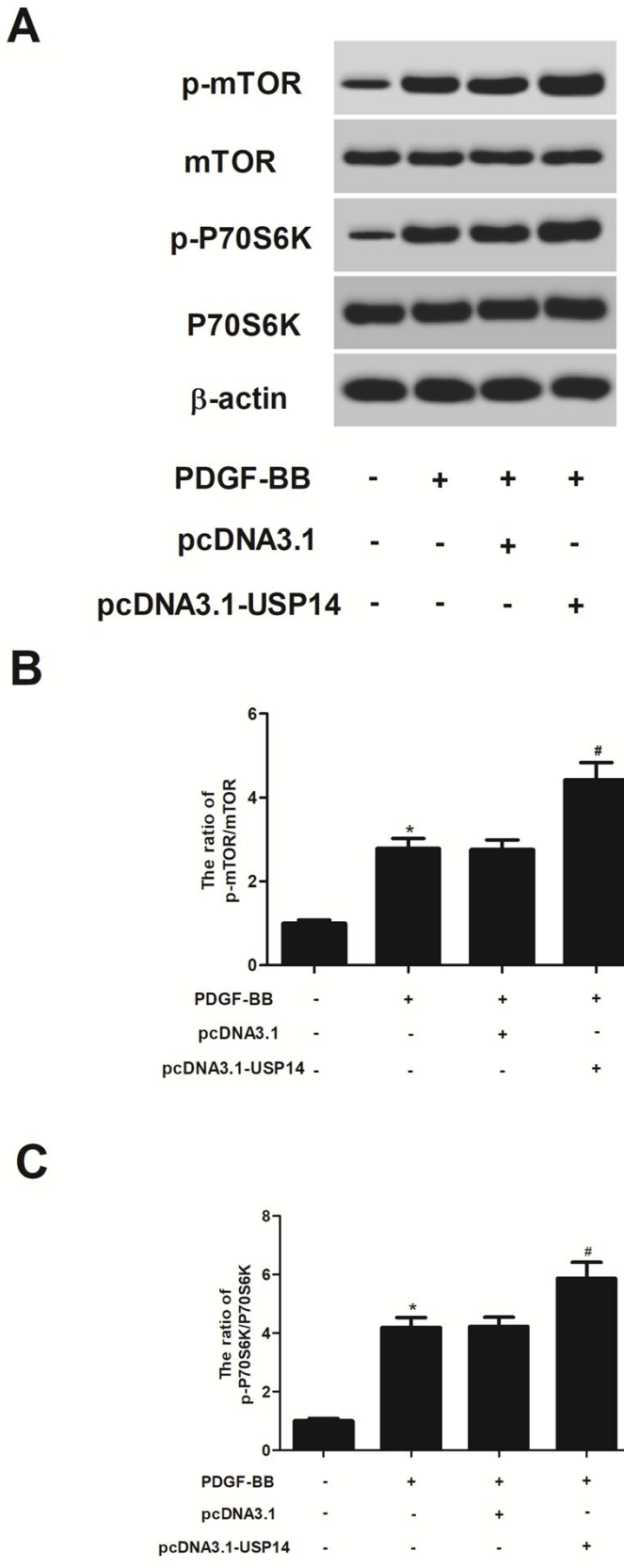
A


p-Akt
Akt
β-actin
PDGF-BB - + + + -
si-NC - - + - -
si-USP14 - - - + +

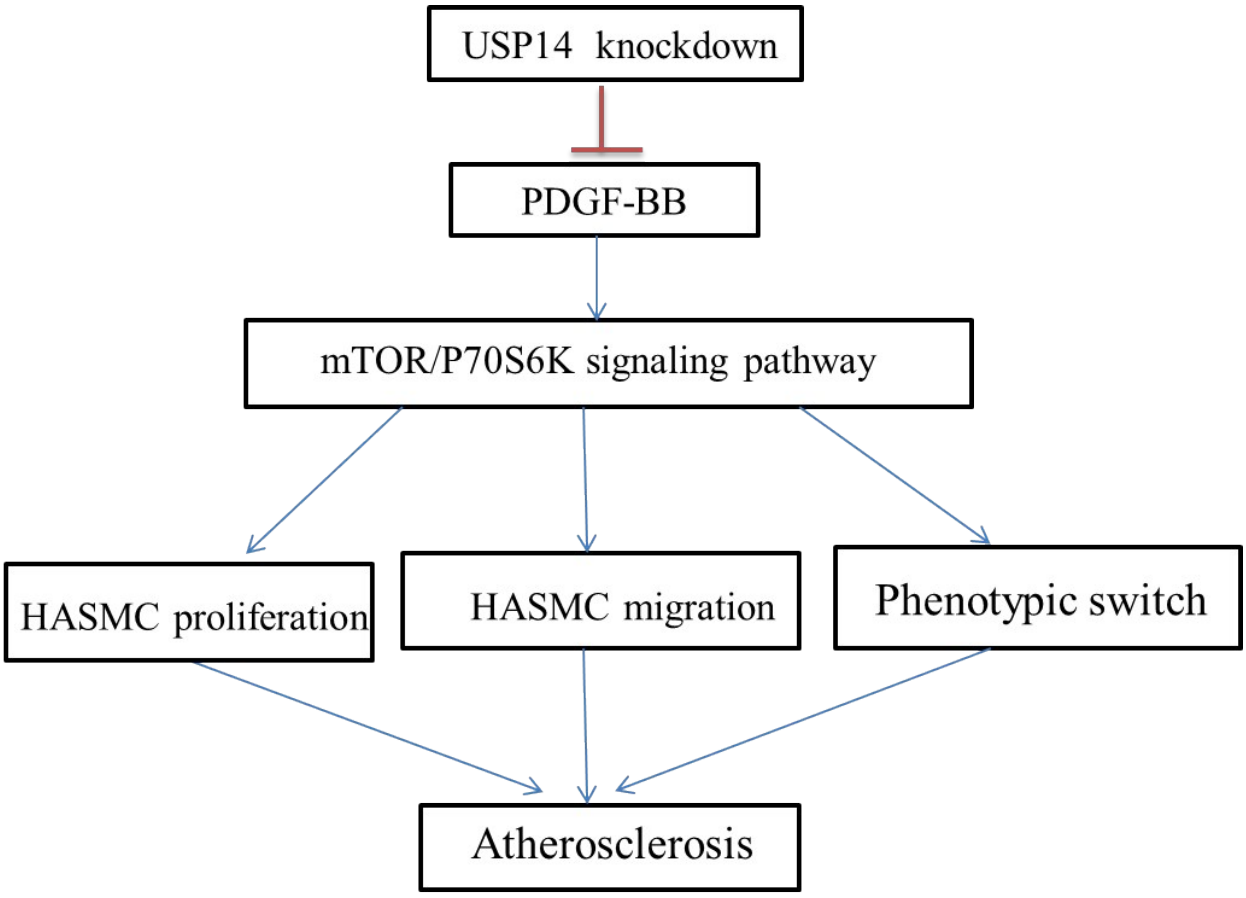
B


The ratio of p-Akt/Akt
0
1
2
3
4
*
#
PDGF-BB - + + + -
si-NC - - + - -
si-USP14 - - - + +

Supplemetary Figure 5. Knockdown of USP14 prevents the PDGF-BB-induced the phosphorylation of Akt in HASMCs. (A) The phosphorylation of Akt and total Akt were detected using western blot analysis. (B) Quantification analysis of p-Akt/Akt. *$p < 0.05$ vs. control group; #$p < 0.05$ vs. si-NC+PDGF-BB group.


A
p-mTOR
mTOR
p-P70S6K
P70S6K
β-actin
PDGF-BB - + + +
pcDNA3.1 - - + -
pcDNA3.1-USP14 - - - +
B
The ratio of p-mTOR/mTOR
0
2
4
6
*
#
PDGF-BB - + + +
pcDNA3.1 - - + -
pcDNA3.1-USP14 - - - +
C
The ratio of p-P70S6K/P70S6K
0
2
4
6
8
*
#
PDGF-BB - + + +
pcDNA3.1 - - + -
pcDNA3.1-USP14 - - - +

Supplementary Figure 6. Overexpression of USP14 promotes the PDGF-BB-induced activation of mTOR/P70S6K signaling in HASMCs. (A) The effect of USP14 overexpression on mTOR/P70S6K signaling pathway was examined by detecting the expression levels of mTOR, P70S6K, p-mTOR and p-P70S6K using western blot analysis. (B and C) Quantification analysis of p-mTOR/mTOR and p-P70S6K/P70S6K. *$p < 0.05$ vs. control group; #$p < 0.05$ vs. pcDNA3.1+PDGF-BB group.



Supplementary Figure 7. A proposed model showing USP14 regulates PDGF-BB-induced vascular smooth muscle cell dedifferentiation via the mTOR/P70S6K signaling pathway.