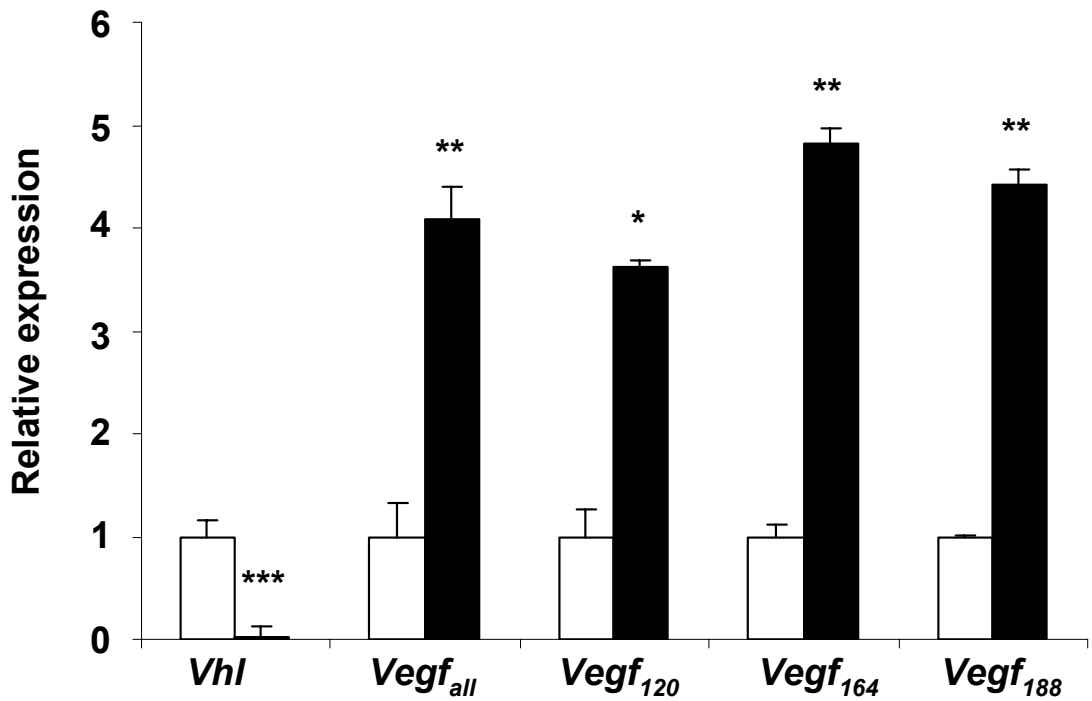
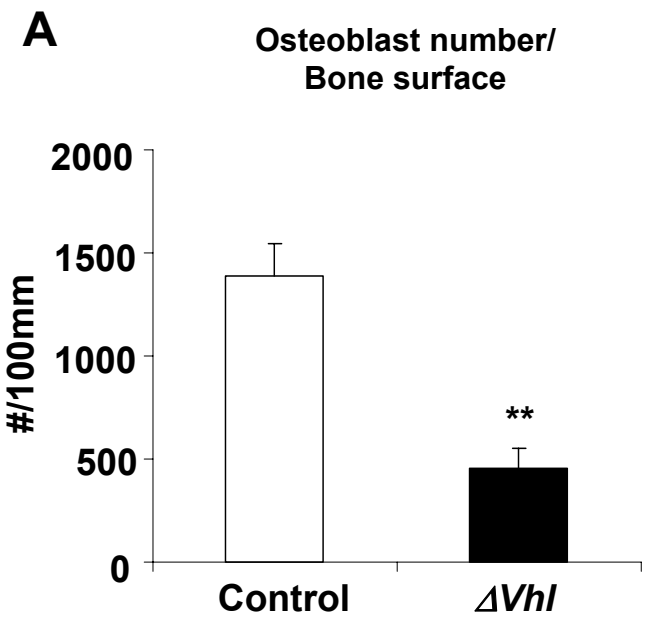
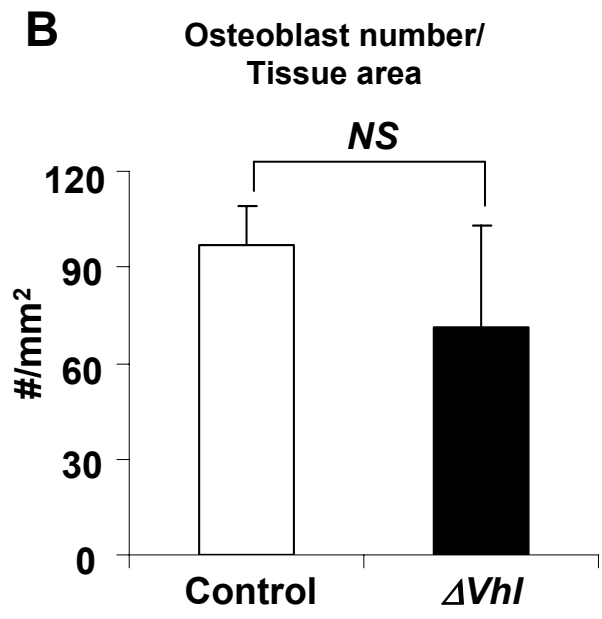
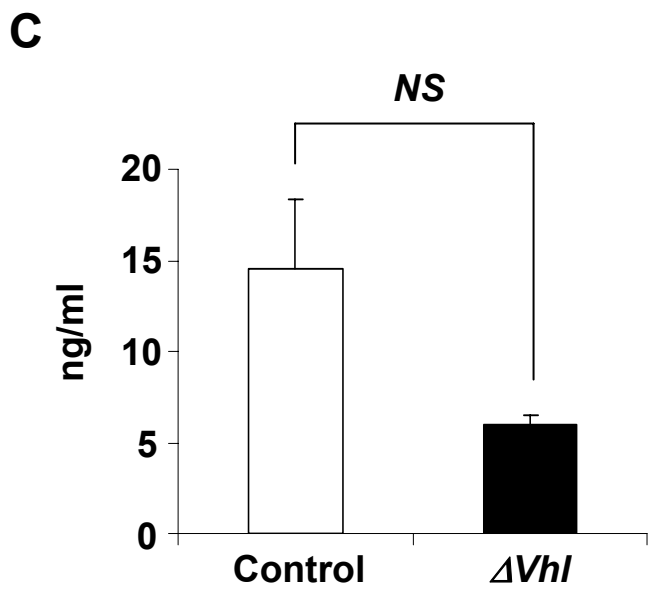
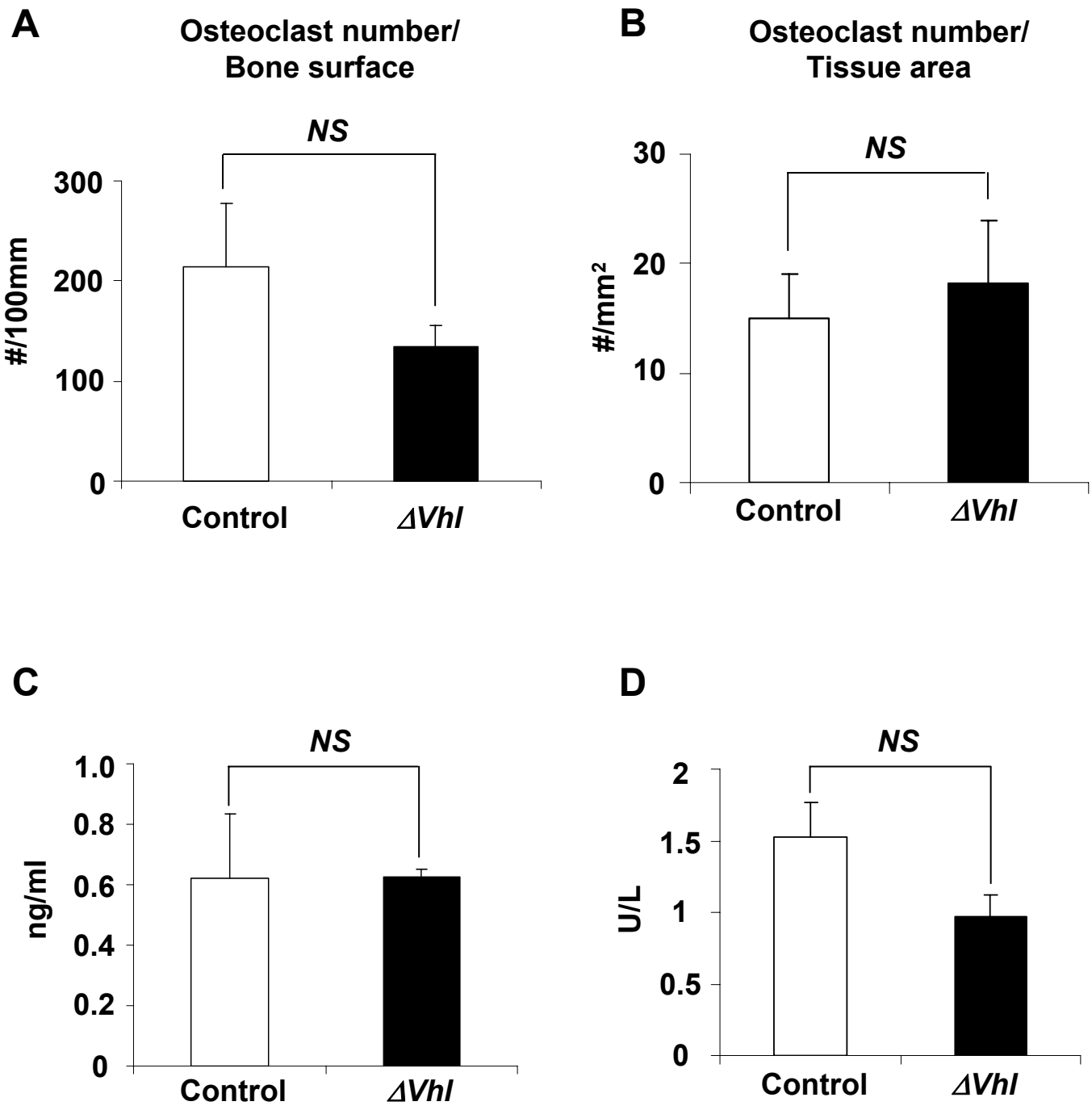

Relative expression
6
5
4
3
2
1
0
***
**
*
**
**
Vhl
Vegf all
Vegf 120
Vegf 164
Vegf 188

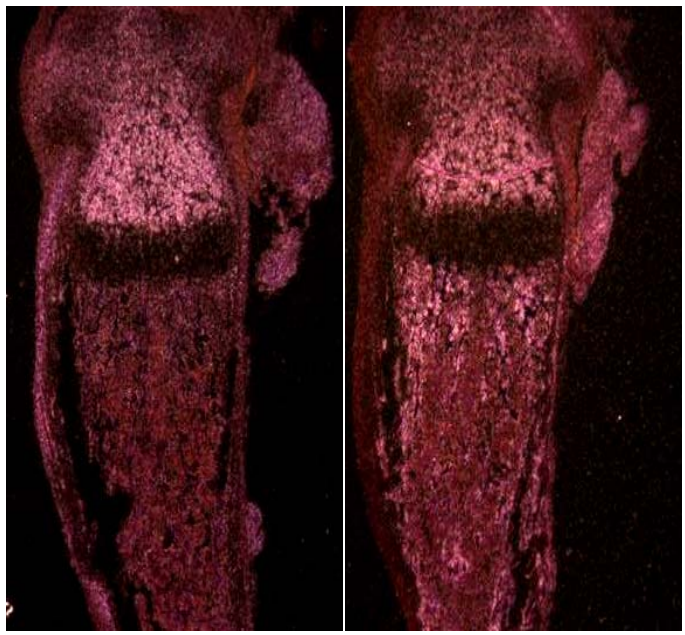

A
Osteoblast number/
Bone surface
2000
1500
1000
500
0
#/100mm
**
Control
ΔVhl


B
Osteoblast number/
Tissue area
NS
120
90
60
30
0
#/mm²
Control
ΔVhl


C
NS
20
15
10
5
0
ng/ml
Control
ΔVhl

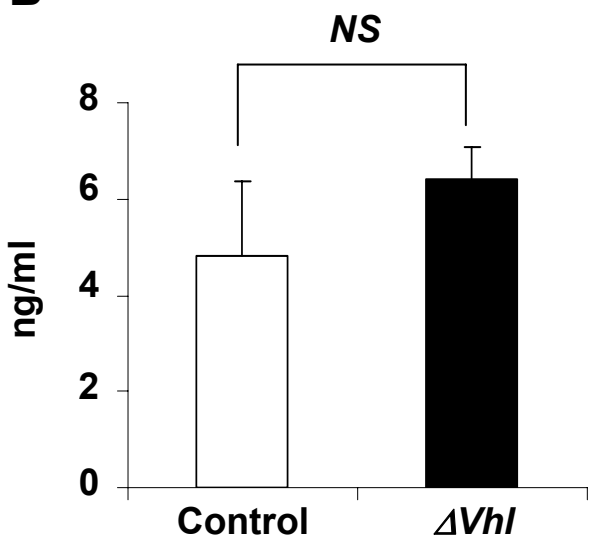

A
Osteoclast number/
Bone surface
NS
#/100mm
300
200
100
0
Control
ΔVhl
B
Osteoclast number/
Tissue area
NS
#/mm²
30
20
10
0
Control
ΔVhl
C
NS
ng/ml
1.0
0.8
0.6
0.4
0.2
0
Control
ΔVhl
D
NS
U/L
2
1.5
1
0.5
0
Control
ΔVhl

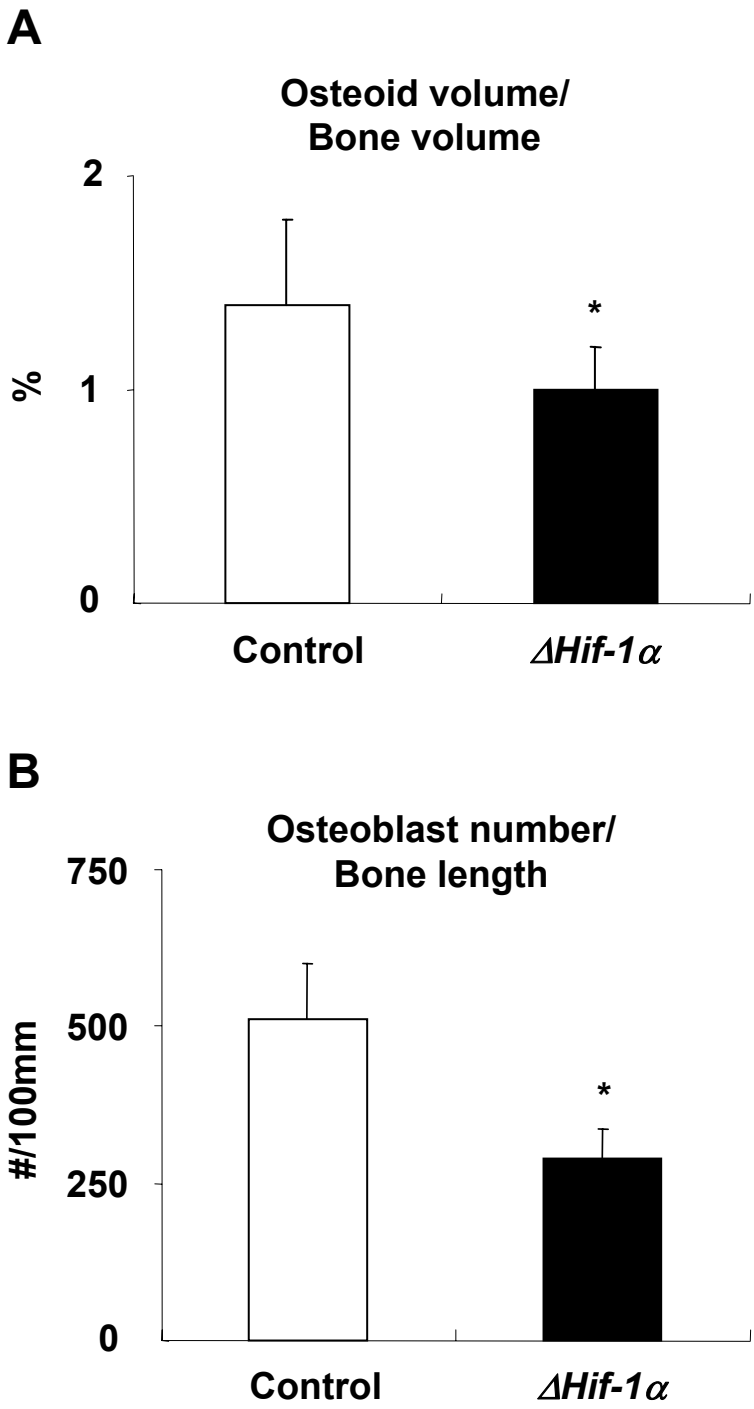
A



Control *ΔVhl*

B




A
Osteoid volume/
Bone volume
2
1
0
%
*
Control
ΔHif-1α
B
Osteoblast number/
Bone length
750
500
250
0
#/100mm
*
Control
ΔHif-1α

Supplemental figure 1

Vhl deletion in mouse osteoblasts is associated with increased *Vegf* mRNA. Confluent monolayers of *Vhl* floxed primary osteoblasts were infected with either Adeno GFP or Adeno CreM1 (100 MOI) for 48 h. Total mRNA was extracted from confluent monolayers and gene expression of $Vegf_{all}$, $Vegf_{120}$, $Vegf_{164}$, and $Vegf_{188}$ was determined by quantitative real-time PCR using sequence specific primers. Open bars represent Adeno GFP infection and solid bars represent Adeno CreM1 infection. $Vegf_{all}$, all *mVEGF* isoforms. *, $P < 0.05$; **, $P < 0.01$; ***, $P < 0.001$.

Supplemental figure 2

Osteoblast numbers and serum osteocalcin in *ΔVhl* and control mice. (**A** and **B**) Histomorphometric analyses for osteoblast numbers were performed on femoral sections from *ΔVhl* mice (solid bars, n = 7) and controls (open bars, n = 6) at 3 weeks of age as described under "Methods". (**A**) Osteoblast numbers per trabecular bone surface. (**B**) Osteoblast numbers per tissue area. Data represent mean ± SE. *, $P < 0.05$. (**C**) Serum levels of osteocalcin were measured in control and *ΔVhl* mice at 6 weeks of age by ELISA, as described under "Methods". Data represent mean ± SE. n = 3. *NS*, not significant.

Supplemental figure 3

Osteoclast numbers and serum TRAP5b in *ΔVhl* and control mice. (**A** and **B**) Histomorphometric analyses for osteoclast numbers on femoral sections from *ΔVhl* mice (solid bars, n = 7) and controls (open bars, n = 6) at 3 weeks of age as described under "Methods". (**A**) Osteoclast numbers per trabecular bone surface. (**B**) Osteoclast numbers per tissue area. Data represent mean ± SE. *NS*, not significant. (**C** and **D**)

Serum levels of OPG and TRAP 5b were measured in control and *ΔVhl* mice at 6 weeks of age by ELISA, as described under "Methods". Data represent mean ± SE. n = 3. *NS*, not significant.

Supplemental figure 4

Elevated *Pgk* mRNA expression and un-changed circulating VEGF level in *ΔVhl* mice. (**A**) *In situ* hybridization analysis with *Pgk* mRNA on histological sections from 3-day-old control and *Vhl* mutant femurs was performed as described under "Methods". Magnification, × 40. (**B**) Serum levels of VEGF were measured in control and *ΔVhl* mice at 12 weeks of age by ELISA, as described under "Methods". Data represent mean ± SE. n = 5. *NS*, not significant.

Supplemental figure 5

Decreased bone volume and osteoblast numbers in mice lacking *Hif-1α* in osteoblasts. Histomorphometric analyses were performed on femoral sections from *ΔHif-1α* mice and controls at 3 weeks of age as described under "Methods". Comparison of osteoid volume (**A**) and osteoblast numbers (**B**) between control (open bars, n = 6) and *ΔHif-1α* mice (solid bars, n = 6) are shown. Data represent mean ± SE. *, $P < 0.05$.