

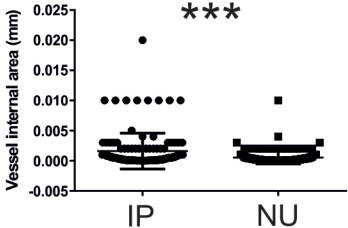
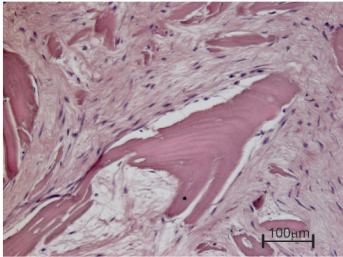
A**B**

Figure 2E

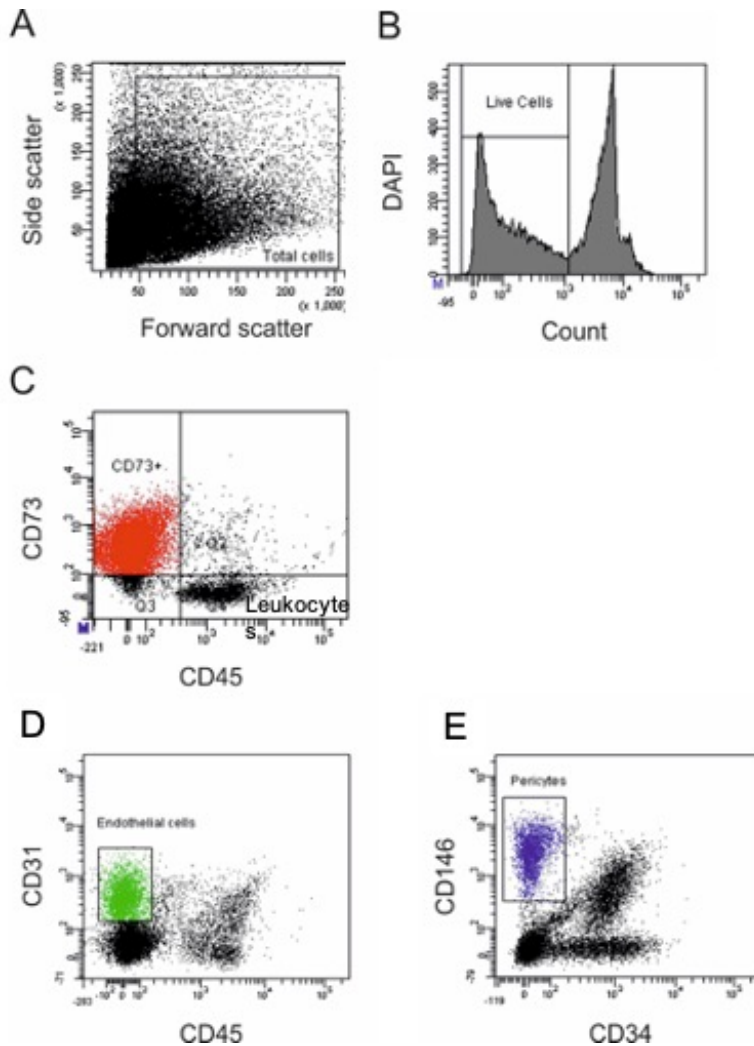


Figure 2E: Cell populations in tissue digests of non-union tissue.

Cells are isolated from debris based on forward and side scatter profile (A). Dead cells are removed based on uptake of DAPI (B). Leukocytes are identified by positive expression of CD45 and negative expression of CD73 (C). Endothelial cells are identified based on negative expression of CD45 and positive expression of CD31 (CD45-CD31+) (D). Pericytes are identified by pre-gating on negative expression of CD45 then by negative expression of CD34 and positive expression of CD146 (CD45-CD34-CD146+) (E).

Specificity	Application	Conjugate	Clone	Host	Dilution	Manufacturer
CD31	IHC	None	JC70A	Mouse	1:50	DakoCytomation
CD31	FC	BV605	WM59	Mouse	1:20	Biolegend
CD45	IHC	None	2B11+PD7/26	Mouse	1:50	DakoCytomation
CD45	FC	PE-Cy7	HI30	Mouse	1:20	BD Pharmingen
CD146	FC	FITC	P1H12	Mouse	1:20	BD Pharmingen
BMP-2	IHC	None	Polyclonal	Rabbit	1:100	AbD Serotech
SDF-1	IHC	None	79018	Mouse	1:20	R&D Systems
VEGF	IHC	None	Polyclonal	Rabbit	1:20	Santa Cruz Biotech

Supplementary table 1: Antibodies

Gene	Taqman assay	Description
<i>18s</i>	Hs 99999901_s1	18S ribosomal RNA
<i>ACAN</i>	Hs 00153936_m1	aggrecan
<i>ANGPT1</i>	Hs 00181613_m1	angiopoietin 1
<i>ANGPTL4</i>	Hs 01101127_m1	angiopoietin-like 4
<i>ARNT</i>	Hs 01121918_m1	aryl hydrocarbon receptor nuclear translocator
<i>BMP2</i>	Hs 00154192_m1	bone morphogenetic protein 2
<i>BMP7</i>	Hs 00233477_m1	bone morphogenetic protein 7
<i>BMPER</i>	Hs 00403062_m1	BMP binding endothelial regulator
<i>CEBPA</i>	Hs 00269972_s1	CCAAT/enhancer binding protein (C/EBP)
<i>COL1A2</i>	Hs 01028971_m1	collagen, type I, α 2
<i>DDR2</i>	Hs 00178815_m1	discoidin domain receptor tyrosine kinase 2
<i>EGLN1</i>	Hs 00254392_m1	egl nine homolog 1 (<i>C. elegans</i>)
<i>FABP4</i>	Hs 00609791_m1	fatty acid binding protein 4, adipocyte
<i>FLT1</i>	Hs 01052937_m1	fms-related tyrosine kinase 1 (VEGFR1)
<i>FRZB</i>	Hs 00173503_m1	frizzled-related protein
<i>FZD4</i>	Hs 00201853_m1	frizzled family receptor 4
<i>FZD5</i>	Hs 00258278_s1	frizzled family receptor 5
<i>GAPDH</i>	Hs 99999905_m1	glyceraldehyde-3-phosphate dehydrogenase
<i>HIF1A</i>	Hs 00936371_m1	hypoxia inducible factor 1, α subunit
<i>HIF1AN</i>	Hs 00215495_m1	hypoxia inducible factor 1, α subunit inhibitor
<i>HPRT</i>	Hs 99999909_m1	hypoxanthine phosphoribosyltransferase 1
<i>IGF2</i>	Hs 01005963_m1	insulin-like growth factor 2
<i>KDR</i>	Hs 00911702_m1	kinase insert domain receptor (VEGFR2)
<i>LRP5</i>	Hs 00182031_m1	low density lipoprotein receptor related protein 5
<i>MCAM</i>	Hs 00174838_m1	melanoma cell adhesion molecule
<i>MYOD1</i>	Hs 00159528_m1	myogenic differentiation 1
<i>NANOG</i>	Hs 02387400_g1	Nanog homeobox
<i>NGFR</i>	Hs 00182120_m1	nerve growth factor receptor
<i>OMD</i>	Hs 00192325_m1	osteomodulin
<i>PDGFRA</i>	Hs 00998018_m1	platelet-derived growth factor receptor α
<i>PDGFRL</i>	Hs 00185122_m1	platelet-derived growth factor receptor-like
<i>PECAMI</i>	Hs 00169777_m1	platelet/endothelial cell adhesion molecule
<i>PGF</i>	Hs 01119262_m1	placental growth factor
<i>POU5F1</i>	Hs 00999632_g1	POU class 5 homeobox 1
<i>PPARG</i>	Hs 01115513_m1	peroxisome proliferator-activated receptor γ
<i>PTN*</i>	Hs 00383255_m1	pleiotrophin
<i>PTPRC</i>	Hs 00894732_m1	protein tyrosine phosphatase, receptor C
<i>PTPRZ1*</i>	Hs 00960146_m1	protein tyrosine phosphatase, receptor type Z1
<i>SFRP1</i>	Hs 00610060_m1	secreted frizzled-related protein 1
<i>SFRP4</i>	Hs 00180066_m1	secreted frizzled-related protein 4
<i>SOX9</i>	Hs 00165814_m1	SRY (sex determining region Y)-box 9
<i>SPARC</i>	Hs 00277762_m1	osteonectin
<i>SPP1</i>	Hs 00959010_m1	secreted phosphoprotein 1
<i>TNFRSF11B</i>	Hs 00900360_m1	tumor necrosis factor receptor superfamily 11b
<i>VEGFA</i>	Hs 00900058_m1	vascular endothelial growth factor A
<i>VEGFB</i>	Hs 00173634_m1	vascular endothelial growth factor B
<i>VEGFC</i>	Hs 01099206_m1	vascular endothelial growth factor C
<i>VHL</i>	Hs 01650959_m1	von Hippel-Lindau tumor suppressor
<i>WIF1</i>	Hs 00183662_m1	WNT inhibitory factor 1
<i>WNT2</i>	Hs00608224_m1	wingless-type MMTV integration site family 2

Supplementary table 2: Taqman assays used for gene expression study

Supplementary Table 3. Control tissues used throughout the study

Colony formation and differentiation capacity of NU MSCs	Bone marrow aspirate
Cellular composition of NU tissue	Induced periosteum tissue
Histology and flow cytometry	Induced periosteum tissue
Transcriptional profiling	Induced periosteum tissue, bone marrow aspirate, skin fibroblasts (negative control)
Assessment of angiogenesis	Induced periosteum tissue