

Figure S1

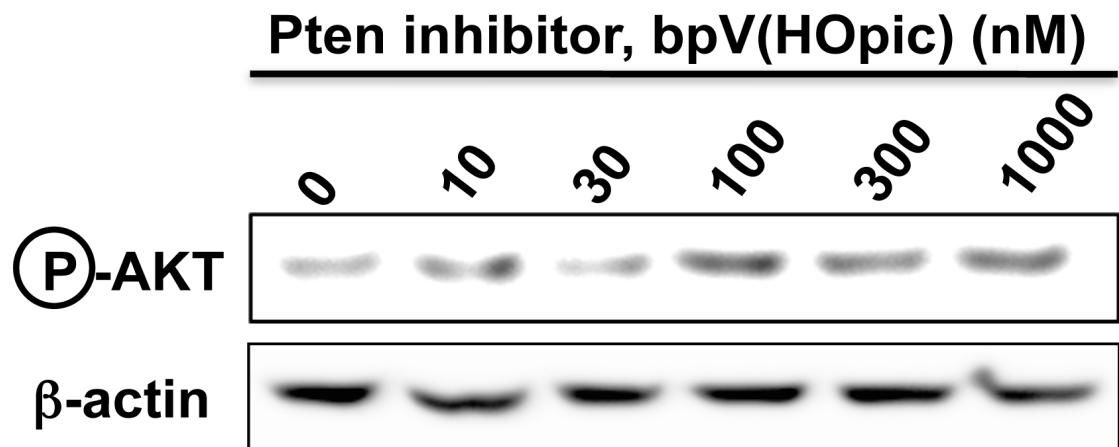


Figure S2

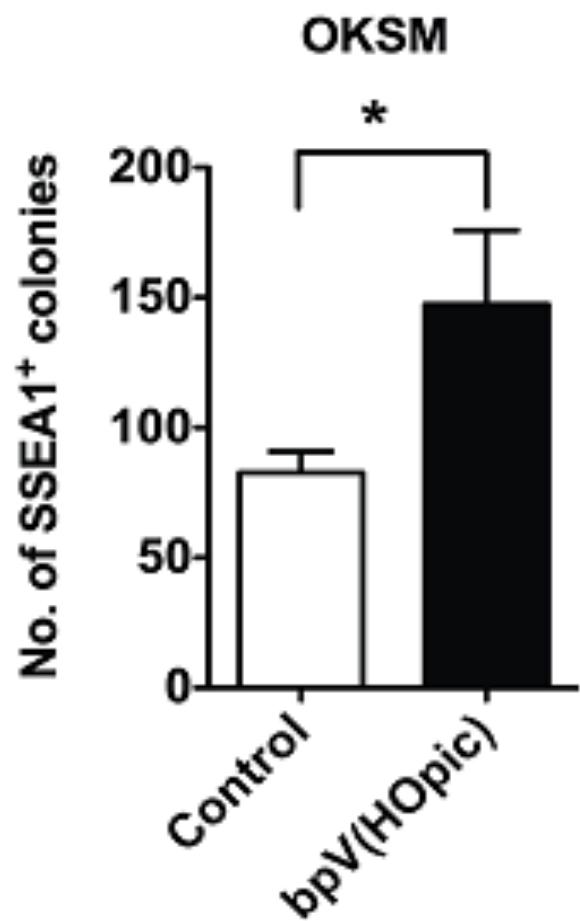


Figure S3

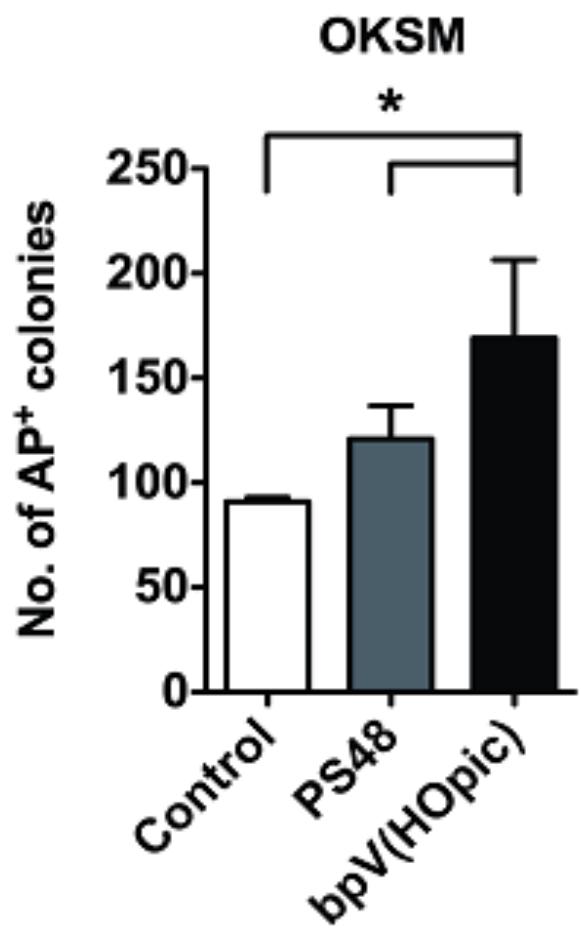


Figure S4

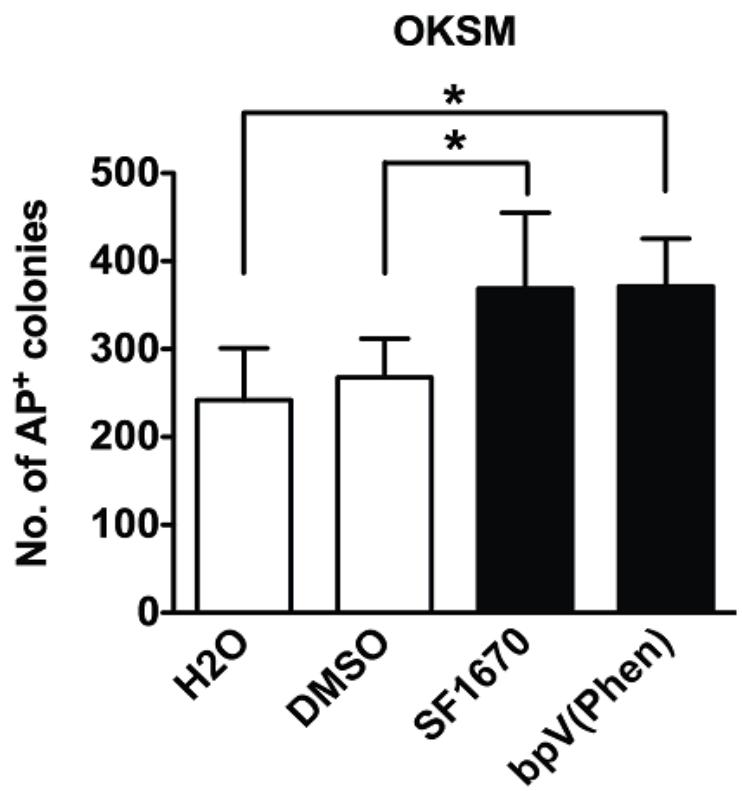


Figure S5

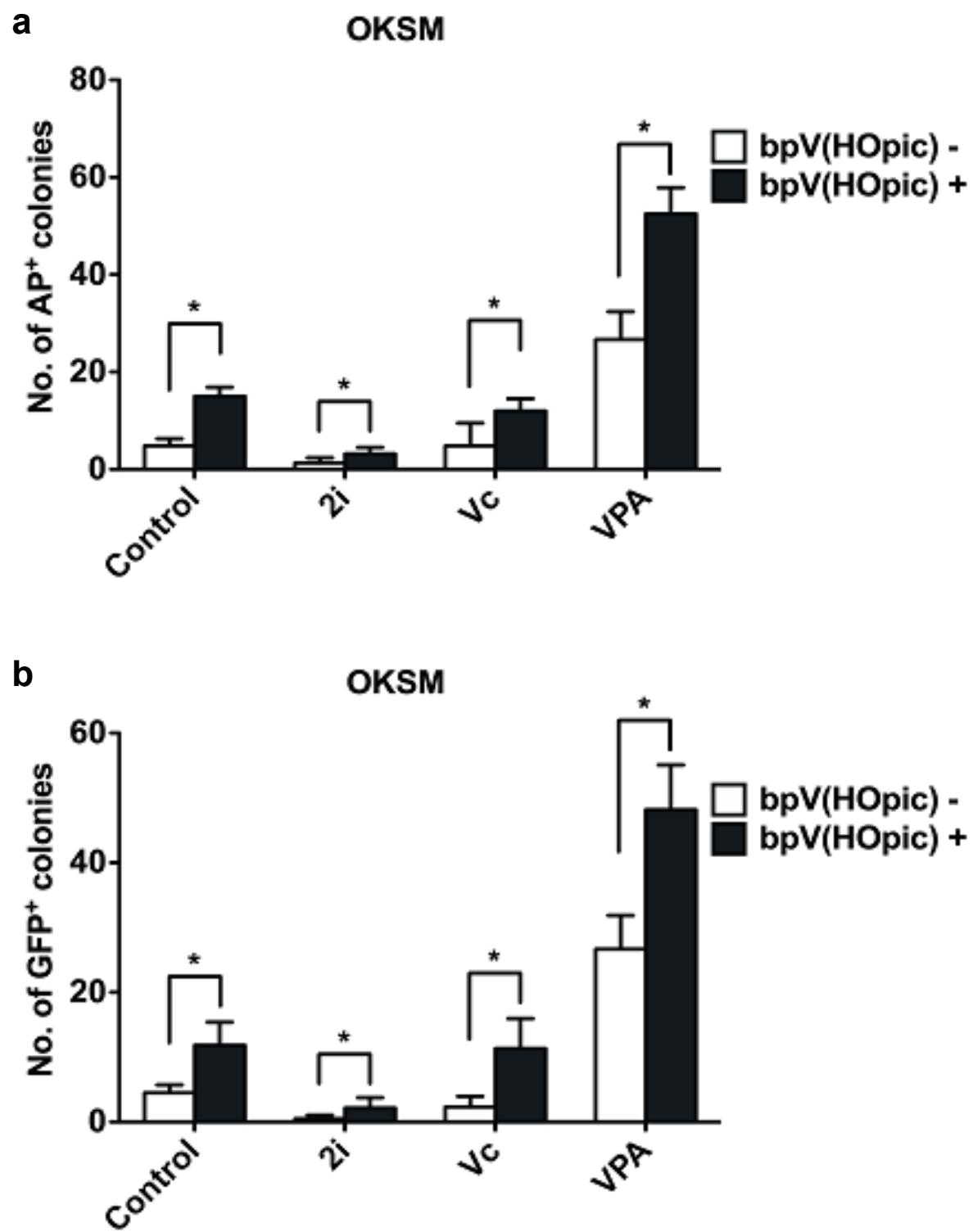


Figure S6

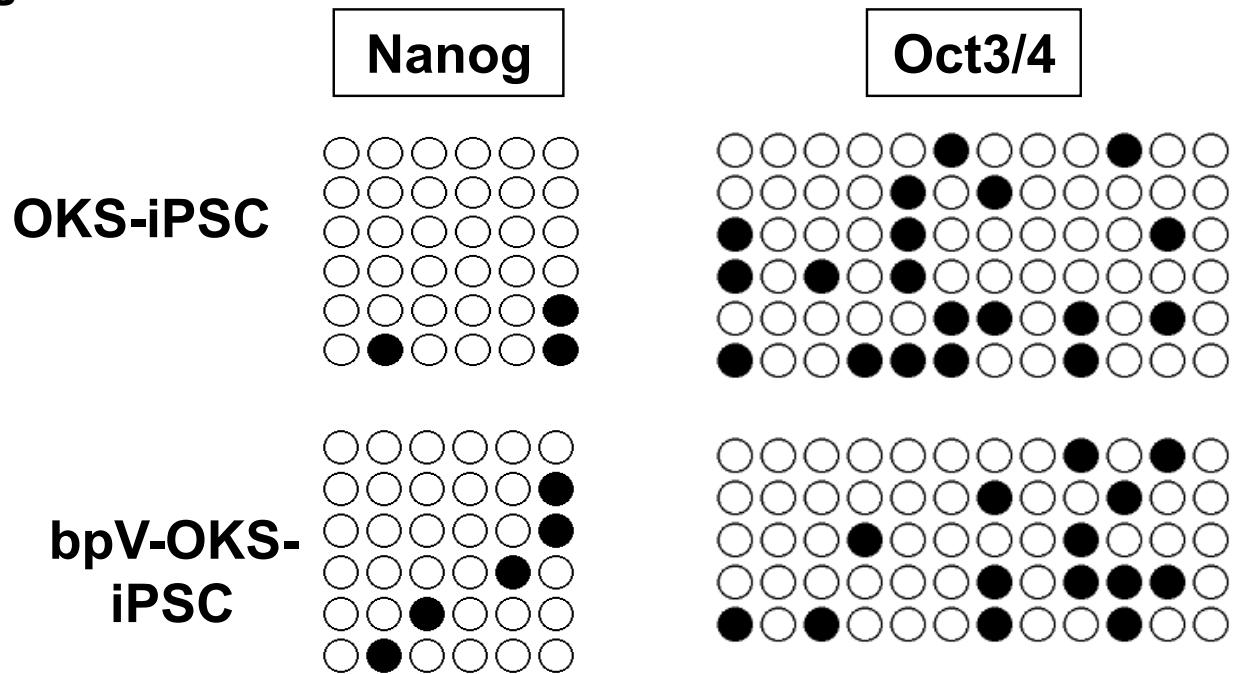


Figure S7

bpV-iPSC

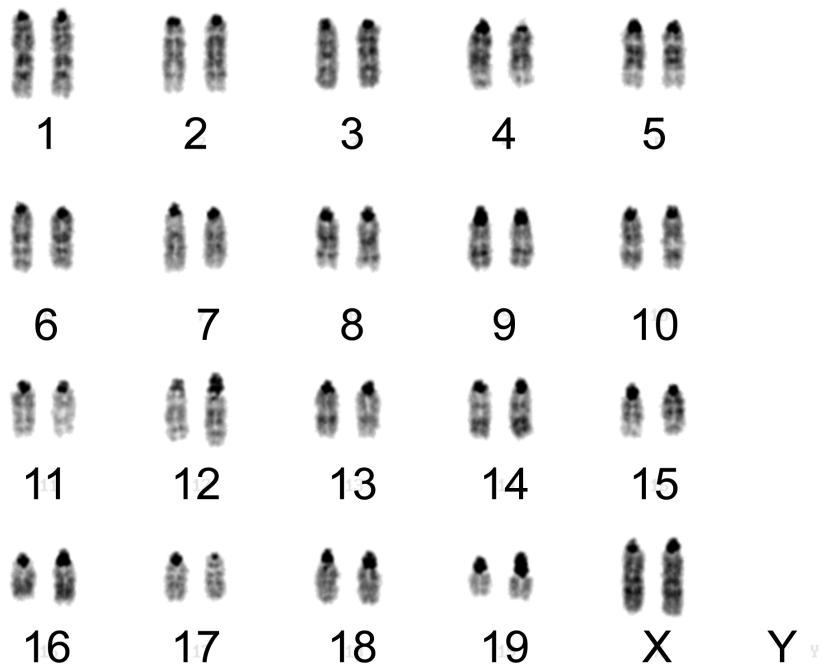


Figure S8

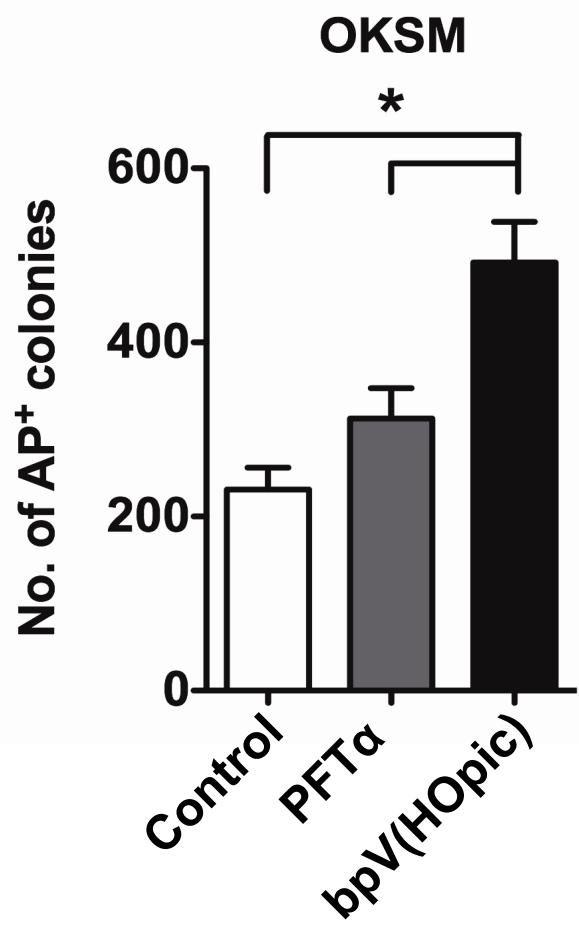
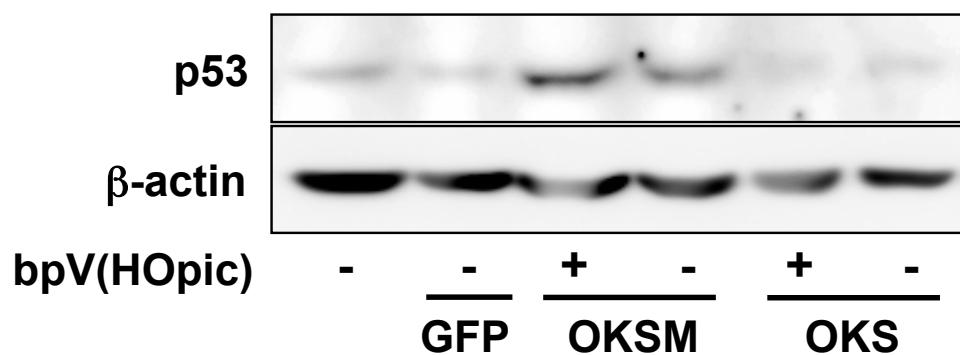


Figure S9



Supplementary Tables

Table S1 Summary of established bpV-iPSC cell lines

Mouse iPSC	Induction factors	Cell source	AP Stain	RT-PCR	Teratoma Formation	Karyotype analysis	Blastocyst Injectionion	Number of Chimeras
bpV-iPSC-1	OKSM	ICR MEF	✓	✓	ND	ND	ND	ND
bpV-iPSC-2	OKSM	ICR MEF	✓	✓	✓	ND	✓	0
bpV-iPSC-3	OKSM	ICR MEF	✓	✓	ND	ND	ND	ND
bpV-iPSC-4	OKSM	ICR MEF	✓	✓	ND	ND	ND	ND
bpV-iPSC-5	OKSM	ICR MEF	✓	✓	ND	ND	ND	ND
bpV-iPSC-6	OKSM	ICR MEF	✓	✓	✓	40, XY	✓	8
bpV-iPSC-7	OKSM	ICR MEF	✓	✓	✓	40, XX	✓	0
bpV-iPSC-8	OKSM	ICR MEF	✓	✓	✓	40, XY	✓	1

ND means not determined.

Table S2 Primers used for RT-PCR

Gene	Forward primer (5' to 3')	Reverse primer (5' to 3')
pMXs	GCCGGATCTAGCTAGTTAA	/
ERas	ACTGCCCTCATCAGACTGCTACT	CACTGCCTTGTACTCGGGTAGCTG
Fgf4	CGTGGTGAGCATCTCGGAGTGG	CCTTCTTGGTCCGCCGTTCTTA
Cript	TCACTCCAGACACATGGAAAGA	CCATATGGATCAAACCTTGCTT
Dax1	TGCTGCGGTCCAGGCCATCAAGAG	GGGCACTGTTCAGTCAGCGGATC
Zfp296	CCATTAGGGGCCATCATCGCTTC	CACTGCTCACTGGAGGGGGCTTGC
β-actin	ATGGATGACGATATCGCTGC	TGCGCTCAGGAGGAGCAATG
Nanog	AAGGCAGCCCTGATTCTTCT	GTGCTGAGCCCTTCTGAATC
c-Myc	GACTCCGTACAGCCATT	TTGGCAGCTGGATAGTCCTCCTT
Sox2	ATGATGGAGACGGAGCTGA	GTGGGAGGAAGAGGTAACCA
Oct3/4	GATCCTCGAACCTGGCTAAG	CGCCGGTTACAGAACCATAC
Klf4	GCTCTGCTCCCGTCCTCCTC	GGCATGAGCTTTGATAATGG

Table S3 Endogenous primers used for RT-PCR

Endo-Gene	Forward primer (5' to 3')	Reverse primer (5' to 3')
c-Myc	TGAAGAAGAGCAAGAAGATGAGG	CTTGAGCATGCATTAAATTCC
Sox2	GTGGTTACCTCTCCTCCCCT	CTTGAAAATCTCTCCCTTCTC
Oct3/4	TCTACTCAGCCCTTCTGAG	AACAGCATCACTGAGCTTCTTC
Klf4	CCTTACACATGAAGAGGCAC	CTGATTATCCATTACAAGCTGA

Table S4 Primers used for bisulfite sequencing

Gene	Forward primer (5' to 3')	Reverse primer (5' to 3')
Oct3/4 promoter	F1: GTTGTGGTTGGGATAT F2: ATGGGTTGAAATATTGGGTTATTAA	CCACCCTCTAACCTAACCTCTAAC
Nanog promoter 1	GTTATTAAAGGTAATAGAGAAAAATTGTT	ACAAAAAAACTATAAAATAACCCAAAC TA
Nanog promoter 2	F1: GAGGATGTTTTAAGTTTTTT F2: AATGTTATGGTGGATTTGTAGGT	CCCACACTCATATCAATATAAAC

Supplemental Materials and Methods

Chemical compounds

Compounds used for reprogramming (day 5–14 post-transduction of OKSM) were PFTα (20 μM; MBL, Woburn, MA), PD0325901 (1 μM; Wako), CHIR99021 (3 μM; Cayman Chemical Company, Ann Arbor, MI), Vc (50 μg/ml; Sigma) and VPA (1 mM; Sigma) with or without bpV(HOpic) (100 nM; Calbiochem).

Western blot analysis

MEFs transduced with OKSM or OKS on days 4 post-transduction were lysed in NP-40 lysis buffer (50 mM Tris-HCl, pH 7.4, 150 mM NaCl, 1 mM EDTA and 0.5% NP-40) with a protease inhibitor (Nacalai Tesque) and a Phosphatase inhibitor cocktail (Roche Diagnostics). The cell lysates were resolved by SDS-PAGE, and then transferred to a PVDF membrane. The membrane was blocked in PBS containing 0.1% Tween20 and 5% skim milk powder for 1 hr, and then incubated with antibodies against p53 (Novocastra, Newcastle, UK), β-actin (Santa Cruz Biotechnology) or phospho-Akt (Ser473) (Cell Signaling Technology, Boston, MA). After incubation with secondary antibodies for 1 hr, specific protein bands were detected by chemiluminescence on a LAS-3000 luminescent image analyzer (FUJIFILM, Tokyo, Japan).