

## Supplemental Procedures

### Antibodies and chemicals

The following reagents were obtained as indicated: PACS-2 (193), PACS-2 (834), PACS-1 (704) (Aslan et al., 2009; Simmen et al., 2005); Proteintech (PACS-2 19508-1-AP), PUMA (Yu et al., 2003), Chemicon (actin MAB1501), Novocastra (p53 CM5 #NCL-p53-CM5p), Calbiochem (p53 DO-1 #OP43; Leptomycin B #431050), Cell Signaling Technology (Acetyl-p53Lys<sub>382</sub> #2525; Acetyl-p53Lys<sub>379</sub> #2570; phospho-p53Ser<sub>15</sub> #9284; cleaved caspase-3 #9664; caspase-3 (3G2) #9668; PARP (46D11) #9532; SIRT1 #2028;  $\beta$ -tubulin #2148; DBC1 #5693), Santa Cruz Biotechnology (p53 FL-393 sc-6243; SIRT1 H-300 sc-15404; Bax N-20 sc-493), BD Pharmingen (p21 #556430; Topo II beta #611492; BrdU #550891), Genscript (THE-His antibody #A00186) Invitrogen (V5 R960-25) Sigma (Flag F7425; FLAG mAb M2-agarose #A2220; Dox #D1515; Trichostatin A #T8852), Covance (HA mAb clone HA.11 MMS-101R), Tocris (EX-527 #2780), and R&D Systems (TRAIL #375-TL). Tissue culture cells were irradiated as described above.

### Plasmids and siRNAs

p300HA, SIRT1-V5 and His<sub>6</sub>-SIRT1 $\Delta_{6-83}$  were provided by J. Denu and His<sub>6</sub>-SIRT1 $\Delta_{6-83}$  was prepared as described (Hallows et al., 2006); p53-FLAG (#10838; Addgene); His<sub>6</sub>-mSIRT1, GST-mSIRT1<sub>1-737</sub> (FL), SIRT1<sub>1-235</sub> (NT), SIRT1<sub>236-490</sub> (SD) and SIRT1<sub>491-737</sub> (CT), His<sub>6</sub>-SIRT1- $\Delta$ NT, which consists of His<sub>6</sub>-mSIRT1 residues 184-510 linked to mSIRT1 residues 631-655 (Hasegawa and Yoshikawa, 2008; Kang et al., 2011; Kim et al., 2007; Kim et al., 2008; Liu et al., 2011; Zhao et al., 2008), His<sub>6</sub>-PCAF<sub>352-832</sub>, GST-hp53<sub>373-385</sub>, GST-hPACS-2<sub>1-37</sub> (NTR), PACS-2<sub>38-202</sub> (FBR), PACS-2<sub>182-468</sub>(MR), PACS-2<sub>469-889</sub>(CTR) and His<sub>6</sub>-PACS-2<sub>38-217</sub> were prepared as previously described (Aslan et al., 2009; Kang et al., 2009; Simmen et al., 2005). PACS-1 HA, PACS-2 HA, and PACS-1-eGFP were described previously (Simmen et al., 2005). The mCherry coding sequence was ligated 5' to the hPACS-2 cDNA using standard methods. PACS-2-FLAG was generated by ligating the FLAG epitope 3' to the hPACS-2 coding sequence. Epitope- and mCherry-tagged hPACS-2 $\Delta$ NLS plasmids

containing an RR<sub>237</sub>→AA substitution or *C. elegans* PACS ((T18H9.7a cDNA provided by D. Sieburth) were generated by standard methods. siRNAs used in this study (Dharmacon; Control #D-001210, PACS-2 #M-022015, p21 M-003471, SIRT1 #M-003540; Qiagen; PACS-2 #SI04193112 and #SI04314730, p21 #SI00299810, SIRT1 #SI04954068) were nucleofected (Amaxa) twice at 48hr intervals and plasmids were transfected for 18 hr using Lipofectamine 2000 (#11668-027; Life Technologies) according to manufacturer's instructions.

### ChIP analysis

10 cm plates of HCT116 cells were washed with PBS, fixed in 1% formaldehyde (10 min, RT), quenched with 125 mM glycine (5 min, RT) and then collected in Harvest Buffer [PBS + 10 mM DTT, and protease inhibitors [1 mM PMSF, 0.1 μM each of aprotinin, E-64, and leupeptin]]. Cells were washed in Buffer I [10 mM Hepes pH 6.5 + 0.25% Triton-X100, 10 mM EDTA, 0.5 mM EGTA, and protease inhibitors], Buffer II [10 mM Hepes pH 6.5 + 200 mM NaCl, 1 mM EDTA, 0.5 mM EGTA and protease inhibitors], then lysed in 300 μl Nuclear Lysis Buffer [50 mM Tris pH 8.0 + 1% SDS, 10 mM EDTA, and protease inhibitors] and incubated on ice for 10 min. Nuclear lysate was diluted 1:2 with ChIP Dilution Buffer [20 mM Tris pH 8.0 + 0.01% SDS, 1% Triton X100, 2 mM EDTA, 150 mM NaCl and protease inhibitors], and sonicated to generate ~400 bp fragments. Sheared chromatin was centrifuged (13.2K RPM, 10 min, 4°C) and supernatant diluted 1:5 in ChIP Dilution Buffer. Samples were pre-cleared with 10 μg IgG with 50 μl pre-blocked Protein A Sepharose (50% slurry) containing 3% BSA and 2 μg sonicated salmon sperm DNA for 2hr at 4°C. Samples were immunoprecipitated with 3 μg of control IgG, 2.5 μg p53 DO-1, or 10 μl of ac-p53 (K<sub>382</sub>) antibodies for 16hr at 4°C. Immune complexes were captured using 50 μl pre-blocked Protein A Sepharose for 2hr at 4°C, washed x2 with TSE-I (20 mM Tris pH 8.0 + 0.1% SDS, 1% Triton X100, 2 mM EDTA, 150 mM NaCl), x3 with TSE-II (TSE-I + 500 mM NaCl), x2 with Buffer III (10 mM Tris pH 8.0 + 0.25 M LiCl, 1% NP-40, 1% deoxycholate, 1 mM EDTA), and x2 with Tris-EDTA (10 mM Tris, 1 mM EDTA, pH 8.0). Samples were eluted (50 mM Tris pH 8.0 + 1% SDS, 1 mM EDTA, 0.1 M NaHCO<sub>3</sub>), incubated at 65°C for 16hr, DNA purified (QIAquick PCR DNA purification kit #28104) and analyzed for qPCR using Taqman Universal

PCR mastermix (#4304438; Life Technologies) and the following p21 primers and probe (5' to 3'): p21 promoter for-GTGGCTCTGATTGGCTTTCTG; p21 promoter rev-CTGAAAACAGGCAGCCC; and the p21 Taqman probe 6FAM-TGGCATAGAAGAGGCTGGTGGCTATTTTG.

qRT-PCR primers

Gene Name	Forward primer (5'-3')	Reverse primer (5'-3')	Accession number
m_GAPDH	CTGGAGAAACCTGCCAAGTA	TGTTGCTGTAGCCGTATTCA	NM_008084
m_p21	CCATGTCCAATCCTGGTGATG	CGAAGAGACAACGGCACACTT	NM_007669
h_GAPDH	CGGGGCTCTCCAGAACATC	ATGACCTTGCCACAGCCT	NM_002046
h_p21	GACTCTCAGGGTCGAAAACGG	GCGGATTAGGGCTTCCTCT	NM_000389
h_Puma	TGTGACCACTGGCATTCAAT	TCCTCCCTCTTCCGAGATTT	NM_001127240
h_Bax	GGGACGAACTGGACAGTAA	CAGTTGAAGTTGCCGTCAGA	NM_004324
h_14.3.3σ	CAGGCTACTTCTCCCTCCT	CTGCCACTGTCCAGTTCTCA	NM_006142
h_Gadd45a	AAGGGGCTGAGTGAGTTCAA	TTTTCTTCCTGCATGGTTC	NM_001199741
h_Mdm2	CCGAATAAGGTTTGCCTGAA	CAAATTGCAAAGGCACTGA	NM_002392
h_DR5	CCCACCTCAGCCATCAAAT	TCACGCCTCTAATTCCACCG	NM_003842