SUPPLEMENTARY TABLES

Table S1, related to Figure 2. Major gross pathologies identified at necropsy.

		SD (87)	SRT1720 (87)
Mean age of death (weeks)		112	121
Liver	Steatosis	9.2%	3.4%
	Hepatocarcinoma	26.4%	38.6%
Kidneys	Enlarged	4.6%	3.4%
	Glomerulonephritis	17.2%	18.2%
Spleen	Enlarged	33.3%	46.6%[#]
Heart	Enlarged	9.2%	3.4%
	Ischemic foci	6.9%	6.8%

The percentage of mice with various pathologies is represented. SD, standard diet.

[#] $p = 0.09$ comparing SD- to SRT1720-treated mice using Fisher's Exact Test.

Table S2, related to Figure 2. Blinded histopathological scoring of mice at sacrifice.

		SD	SRT1720
Liver	Lymphocyte infiltration	1.0 ± 0.0 (4/6)	1.0 ± 0.0 (2/8)
	Steatosis	2.3 ± 0.3 (4/6)	1.0 ± 0.0 (2/8)*
Kidney	Lymphocyte infiltration	1.6 ± 0.3 (5/5)	1.9 ± 0.4 (8/8)*
	Glomerulonephritis	1.2 ± 0.5 (3/5)	1.0 ± 0 (8/8)
Heart	Lesion	0 (0/6)	0 (0/8)
Spleen	Congestion	6 (6/6)	7 (7/8)

Lymphocyte infiltration and fatty change (steatosis) are scored on a scale of 1-4 (most severe); data is mean ± SEM (%). Heart and spleen data is represented as number of cases. n=6 SD, n=8 SRT1720 (76 weeks age, 38 weeks on diet). * $p < 0.05$ comparing SD- to SRT1720-treated mice using Fisher's Exact Test.

Table S3, related to Figure 3. List of the ten most highly up-regulated and down-regulated genes in liver and muscle in response to SRT1720 treatment in SD-fed mice.

Liver		Muscle	
Gene	**(zratio) SD1720:SD**	**Gene**	**(zratio) SD1720:SD**
Socs2	18.05	Cish	19.45
Cish	13.38	Chac1	18.54
Slc25a30	13.03	Clec2d	6.69
Serpina4-ps1	12.76	Ddp	6.10
Hhex	11.21	Pik3ip1	6.07
Txnip	11.17	Car3	5.71
Rgs16	10.36	Cntnap2	5.67
Ctgf	10.14	Bhlhb2	5.60
Chrna4	8.95	Hist1h4j	5.25
Cyp1a2	8.94	Bcl6b	5.09
Rcan1	-10.07	1190002H23Rik	-8.86
Ddit4	-10.60	**Cdkn1a**	**-9.17**
Orm2	-11.50	Cmya1	-9.56
Gadd45a	-11.68	Ankrd1	-9.67
*S100a9	-11.98	Mustn1	-10.23
Cdkn1a	**-12.20**	Ankrd1	-10.40
*S100a8	-14.10	**Gadd45b**	**-10.97**
Saa2	**-15.06**	Axud1	-11.11
Saa1	**-15.18**	Srxn1	-11.44
Lcn2	**-27.10**	Mt1	-16.84

Boldface, NF-κB target genes; *, up-regulates transcription of genes that are under the control of NF-κB.

Table S4, related to Figure 3. The most affected pathways in the liver of SD-fed mice with and without SRT1720 supplementation

Pathway	Z-Score SD1720:SD
RIBOSOMAL_PROTEINS	5.66
CHOLESTEROL_BIOSYNTHESIS	4.95
TERPENOID_BIOSYNTHESIS	4.50
BIOSYNTHESIS_OF_STEROIDS	4.10
MALATEXPATHWAY	3.19
APPEL_IMATINIB_UP	3.07
GUO_HEX_UP	2.61
DNA_REPLICATION_REACTOME	2.45
ST_GA12_PATHWAY	2.15
SARCOMAS_LIPOSARCOMA_DN	2.09
CIS_RESIST_LUNG_DN	1.99
SETPATHWAY	1.76
TGFBETA_C4_UP	1.73
TCYTOTOXICPATHWAY	-1.13
LIZUKA_G2_SM_G3	-1.31
TCRMOLECULE	-1.36
ADIPOGENESIS_HMSC_CLASS5_UP	-1.39
ST_IL_13_PATHWAY	-1.44
ST_INTERLEUKIN_13_PATHWAY	-1.44
STRIATED_MUSCLE_CONTRACTION	-1.55
HIVNEFPATHWAY	-1.57
UBIQUITIN_MEDIATED_PROTEOLYSIS	-1.76
FSH_GRANULOSA_DN	-2.00
LH_GRANULOSA_DN	-2.00
PYRIMIDINE_METABOLISM	-2.14
REN_E2F1_TARGETS	-2.22

FLECHNER_KIDNEY_TRANSPLANT_REJECTION_DN	-2.42
SCHUMACHER_MYC_UP	-2.68
GLUTAMATE_METABOLISM	-2.82
TAKEDA_NUP8_HOXA9_3D_UP	-3.00
IFNALPHA_HCC_UP	-3.19
ET743_RESIST_UP	-3.21
MYOD_NIH3T3_UP	-3.46
HEATSHOCK_YOUNG_UP	-3.55
BYSTRYKH_HSC_BRAIN_TRANS_GLOCUS	-4.02
RADAEVA_IFNA_UP	-4.37
ICHIBA_GVHD	-4.39
IL22BPPATHWAY	-4.65
IFNALPHA_NL_UP	-5.05

Table S5, related to Figure 3. The most affected pathways in the muscle of SD-fed mice with and without SRT1720 supplementation.

See excel file.

Table S6, related to Figure 4. List of the most highly expressed genes that were significantly up- and down-regulated by SRT1720 treatment in wild-type (WT) MEF cells. This list of genes was sorted using *Sirt1*-KO MEFs for comparison. The last column indicates whether the SRT1720-mediated change observed for a given gene is significantly different between WT and *SIRT1*-KO MEFs.

		MEF WT	MEF *Sirt1*-KO	KO:WT
Accession	Symbol	(zratio) SRT1720:UT	(zratio) SRT1720:UT	(p value)
NM_008216.2	Has2	11.98	4.20	4.95E-05
NM_010217.1	Ctgf	10.15	8.70	0
NM_010496.2	Id2	10.03	8.07	0.499
NM_139307.2	Vasn	7.85	*0.93*	0.0005
NM_145535.1	Sdcbp2	7.34	*3.64*	0
NM_010495.2	Id1	7.30	9.15	0
NM_011607.2	Tnc	6.90	*-0.53*	0
NM_011580.3	Thbs1	**6.65**	**-3.54**	0
NM_001039090.1	Skil	6.59	*3.29*	3.03E-09
NM_009397.2	Tnfaip3	**6.48**	*1.34*	2.43E-05
NM_009943.2	Cox6a2	-6.40	*-0.47*	6.31E-09
NM_025378.2	Ifitm3	-6.50	*-0.08*	0
NM_019738.1	Nupr1	-6.71	*3.46*	0
NM_016974.1	Dbp	-6.95	*-1.17*	0
NM_011333.3	Ccl2	**-7.65**	**1.86**	0
NM_007837.2	Ddit3	-8.04	*2.03*	3.22E-12
NM_007836.1	Gadd45a	-8.12	*0.40*	9.08E-14
NM_011315.3	Saa3	**-8.33**	**-3.43**	0.533
NM_013654.2	Ccl7	**-8.39**	*1.38*	0

Boldface, NF-κB target genes; *Italics*, no significant changes versus untreated (UT) cells.

Table S7, related to Figure 4. Partial list of signaling proteins involved in NF-κB activation with altered phosphorylation in response to SRT1720.

Wild-type (WT) and *Sirt1*-KO MEFs were incubated with vehicle or 3 mM SRT1720 for 18 h, after which total lysates were subjected to phospho-antibody microarray analysis. The signal intensities of phosphorylated and total forms of each protein were determined, and the ratio (phospho/total) of each protein was calculated between vehicle (UT)- and SRT1720-treated MEFs. 95% confidence interval (CI) was determined to demonstrate the significance of the signal alteration for each protein. The impact of site-specific phosphorylation on the biological function of these proteins is shown.

	Ratio (MEF WT)		**Ratio (MEF KO)**		
	SRT/UT	**95% CI**	**SRT/UT**	**95% CI**	**Function***
IκBα (Ser(P)-32/36)	1.66	[1.41-1.91]	0.80	[0.61-0.99]	Inactivation (Winston et al., 1999)
IKK-α/β (Ser(P)-180/181)	0.91	[0.86-0.96]	0.85	[0.39-1.31]	Activation (Karin, 1999)
IKK-α (Thr(P)-23)	0.34	[0.26-0.42]	0.92	[0.81-1.03]	Activation (Ozes et al., 1999)
NF-κB-p105/p50 (Ser(P)-907	0.60	[0.54-0.66]	1.00	[0.67-1.33]	Stability (Demarchi et al., 2003)
NF-κB-p65 (Ser(P)-311)	0.24	[0.22-0.26]	0.72	[0.63-0.81]	Activation (Chang et al., 2011)

* Supporting references

SUPPLEMENTAL REFERENCES

Chang, Y., Levy, D., Horton, J.R., Peng, J., Zhang, X., Gozani, O., and Cheng, X. (2011). Structural basis of SETD6-mediated regulation of the NF-κB network via methyl-lysine signaling. Nucleic Acids Research *39*, 6380-6389.

Cheadle, C., Vawter, M.P., Freed, W.J., and Becker, K.G. (2003). Analysis of Microarray Data Using Z Score Transformation. The Journal of Molecular Diagnostics *5*, 73-81

Demarchi, F., Bertoli, C., Sandy, P., and Schneider, C. (2003). Glycogen synthase kinase-3β regulates NF-κB1/p105 stability. Journal of Biological Chemistry *278*, 39583-39590.

Karin, M. (1999). Positive and negative regulation of IκB kinase activity through IKKβ subunit phosphorylation. Science *284*, 309-313.

Kim, S.Y., and Volsky, D.J. (2005). PAGE: Parametric analysis of gene set enrichment. BMC Bioinformatics *6*, 1471-2105

Lee, C., Raffaghello, L., Brandhorst, S., Safdie, F.M., Bianchi, G., Martin-Montalvo, A., Pistoia, V., Wei, M., Hwang, S., Merlino, A.*, et al.* (2012). Fasting Cycles Retard Growth of Tumors and Sensitize a Range of Cancer Cell Types to Chemotherapy. Science Translational Medicine *4*, 124ra127

Ozes, O.N., Mayo, L.D., Gustin, J.A., Pfeffer, S.R., Pfeffer, L.M., and Donner, D.B. (1999). NF-κB activation by tumour necrosis factor requires the Akt serine-threonine kinase. Nature *401*, 82-85

Winston, J.T., Strack, P., Beer-Romero, P., Chu, C.Y., Elledge, S.J., and Harper, J.W. (1999). The SCFβ-TRCP-ubiquitin ligase complex associates specifically with phosphorylated destruction motifs in IκBα and β-catenin and stimulates IκBα ubiquitination in vitro. . Genes & Development *13*, 270-283