

Appendix

Feeding activates FGF15-SHP-TFEB-mediated lipophagy in the gut

Sunmi Seok^a, Young-Chae Kim^{1, 2, a}, Yang Zhang^{1, b}, Bo Kong^c, Grace Guo^c, Jian Ma^b, Byron Kemper^a, and Jongsook Kim Kemper^{a, 3}

The appendix contains the following supplementary information: Appendix Figures S1-S9

Appendix Tables S1-S7

Appendix Figure S1. Feeding activation of intestinal autophagy is largely absent in FXR-KO mice

Appendix Figure S2. Identification of genome-wide intestinal SHP binding sites by ChIP-seq

Appendix Figure S3. Analysis of intestinal RNA-seq data

Appendix Figure S4. SHP occupancy at genes in mouse intestine potentially upregulated or downregulated by SHP binding

Appendix Figure S5. FGF19 activates lipophagy in intestinal cells and FGF19 lipid-lowering effects are dependent on autophagy

Appendix Figure S6. Effects of TFEB overexpression and FGF19 treatment on pre-mRNA and protein levels of p62/SQSTM1 in HT29 cells

Appendix Figure S7. Activation of PKC β in liver hepatocytes in response to FGF15/19 signaling and temporal feeding induction of intestinal autophagy via PKC-mediated phosphorylation in mice

Appendix Figure S8. FGF15/19 signaling increases nuclear localization of TFEB and SHP via PKC in HT29 cells

Appendix Figure S9. Effects of FGF19-induced phosphorylation of TFEB and SHP in nuclear localization and autophagy in HT29 cells.

Appendix Table S1. List of 856 genes with SHP-binding peak and down-regulated in SHP-KO (Up-regulated intestinal genes by SHP binding)

Appendix Table S2. List of 774 genes with SHP-binding peak and up-regulated in SHP-KO (Down-regulated intestinal genes by SHP binding)

Appendix Table S3. 34 unique motifs for down-regulated intestinal genes by SHP binding

Appendix Table S4. 63 unique motifs for up-regulated intestinal genes by SHP binding

Appendix Table S5. Information on antibodies

Appendix Table S6. Information on reagents

Appendix Table S7. Primer sequences used for ChIP-qPCR and qRT-PCR

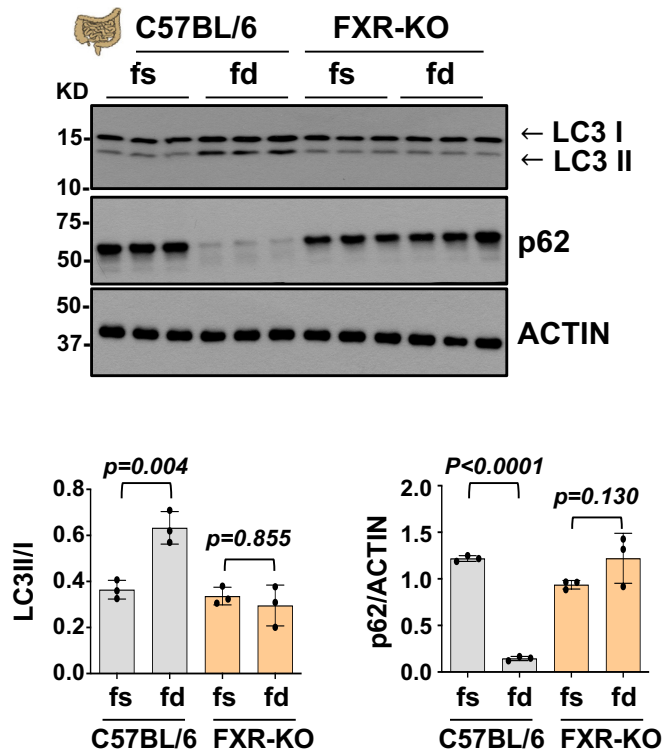


Figure S1. Feeding activation of intestinal autophagy is largely absent in FXR-KO mice. Mice as indicated were fasted overnight and then refed for 6 h. (Top) Levels of the indicated proteins in extracts of intestines from C57BL/6 or FXR-KO mice were detected by IB. (Bottom) LC3-I, LC3-II, and p62 band intensities were quantified and the relative ratios of LC3-II/LC3-I and p62/actin are shown (n=3 mice per each group). The mean and SD are plotted. Statistical significance was determined by two-way ANOVA with the Tukey post-test. P-values are indicated.

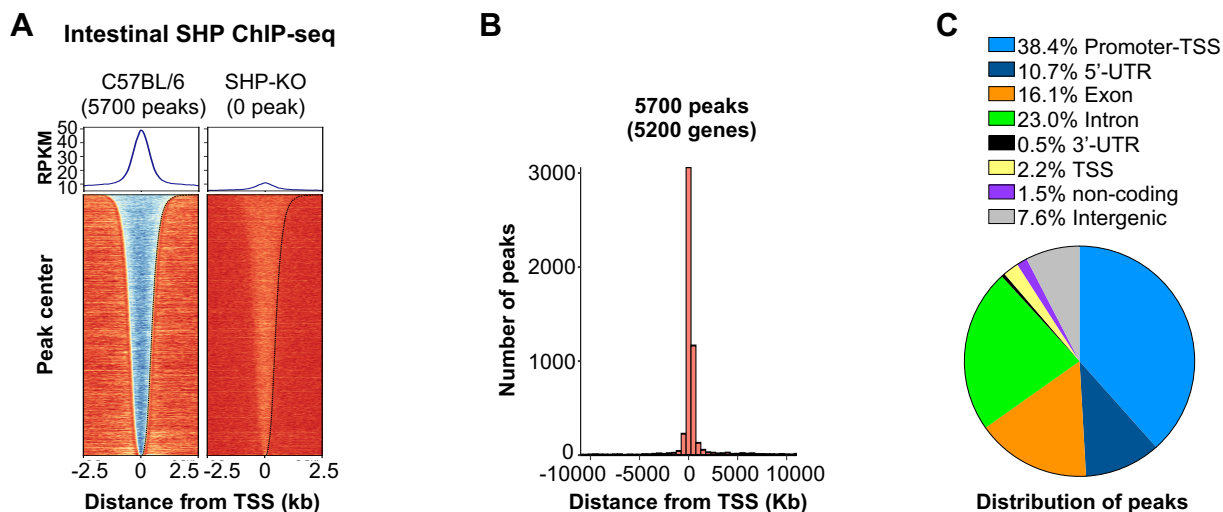


Figure S2. Identification of genome-wide intestinal SHP binding sites by ChIP-seq.

(A-C) C57BL/6 and SHP-KO mice were fasted for 12 h and re-fed for 4 h and intestinal ChIP-seq analysis was performed using SHP antibody (Santa Cruz Biotech, sc-30169).

(A) Heat map showing the distribution of SHP binding peaks relative to the TSS in C57BL/6 mice and the absence of statistically significant peaks in SHP-KO mice.

(B) Distances from the center of SHP binding peaks to the TSS. (C) Pie chart showing the genomic distribution of SHP binding peaks.

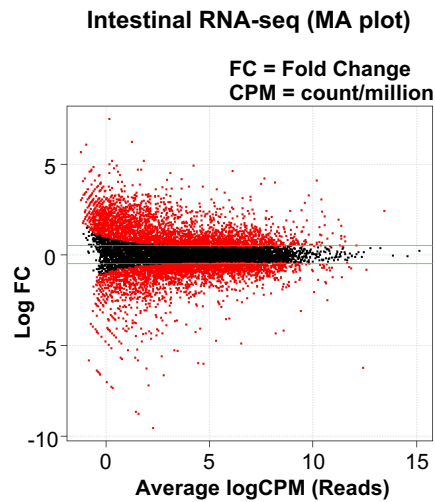


Figure S3. Analysis of intestinal RNA-seq data. Analysis of RNA-seq data published previously (Kim *et al.*, 2018, Gastroenterology 156: 1052-1065) using intestines from C57BL/6 and SHP-KO mice that were fasted for 12 h and re-fed for 6 h. MA (ratio intensity) plot of the RNA-seq data illustrating gene expression changes between C57BL/6 and SHP-KO mice.

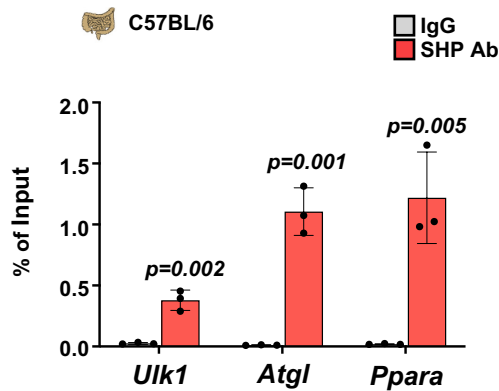
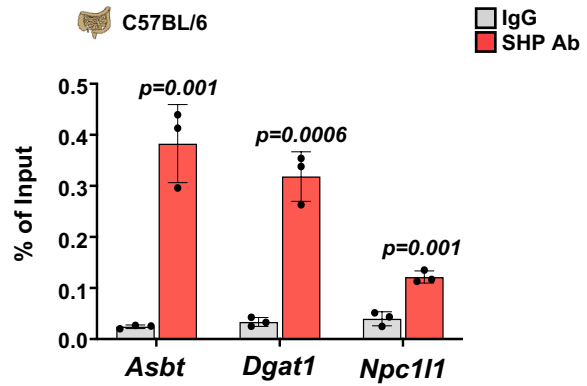
A**Intestinal ChIP : SHP occupancy****B****Intestinal ChIP : SHP occupancy**

Figure S4. SHP occupancy at genes in mouse intestine potentially upregulated or downregulated by SHP binding. SHP binding at selected genes potentially upregulated (A) or downregulated (B) by SHP binding was confirmed by intestinal ChIP assays using SHP antibody (Abcam, ab96605), (n=3 mice). The mean and SD are plotted. Statistical significance was determined by the two-tailed Student's t-test and p-values are indicated.

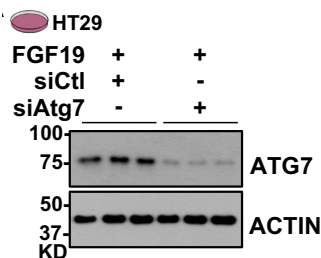
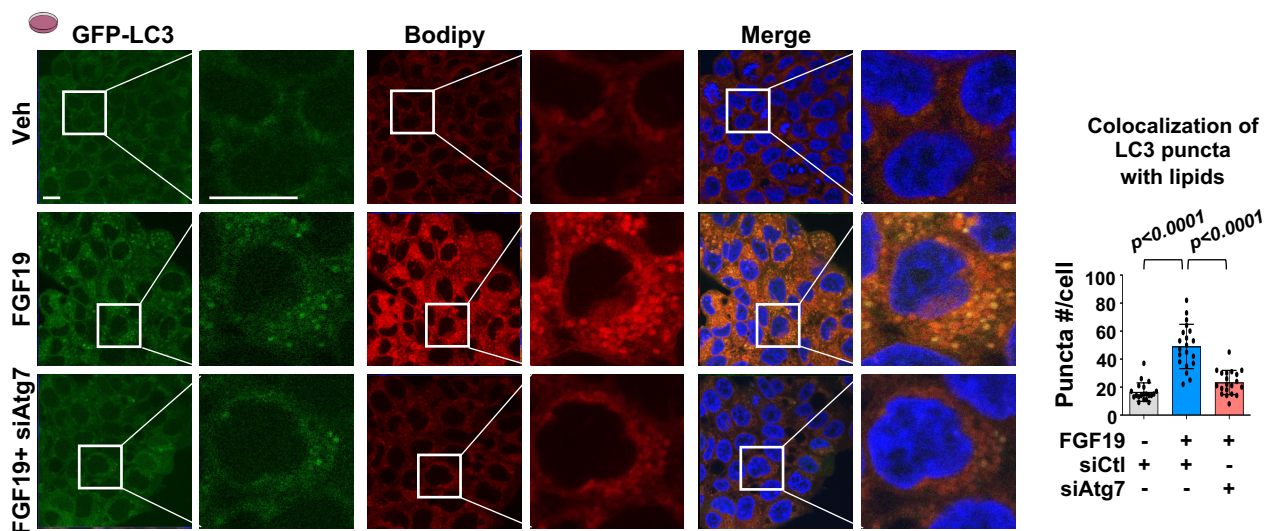
A**B**

Figure S5. FGF19 activates lipophagy in intestinal cells and FGF19 lipid-lowering effects are dependent on autophagy. (A) HT29 cells were transfected with *Atg7* siRNA or scrambled siRNA (siCtl) for 48 h followed by 50 ng/ml FGF19 for 12 h. ATG7 and control actin levels determined by IB (n=3 culture plates). (B) Representative images of LC3 puncta detected by GFP fluorescence and lipids stained with BODIPY are shown. Quantification of the number of puncta/cell that co-localize with lipids (right, n=20 cells). Scale bar, 20 μ m. The mean and SD are plotted. Statistical significance was determined by one-way ANOVA with the Tukey post-test. P-values are indicated.

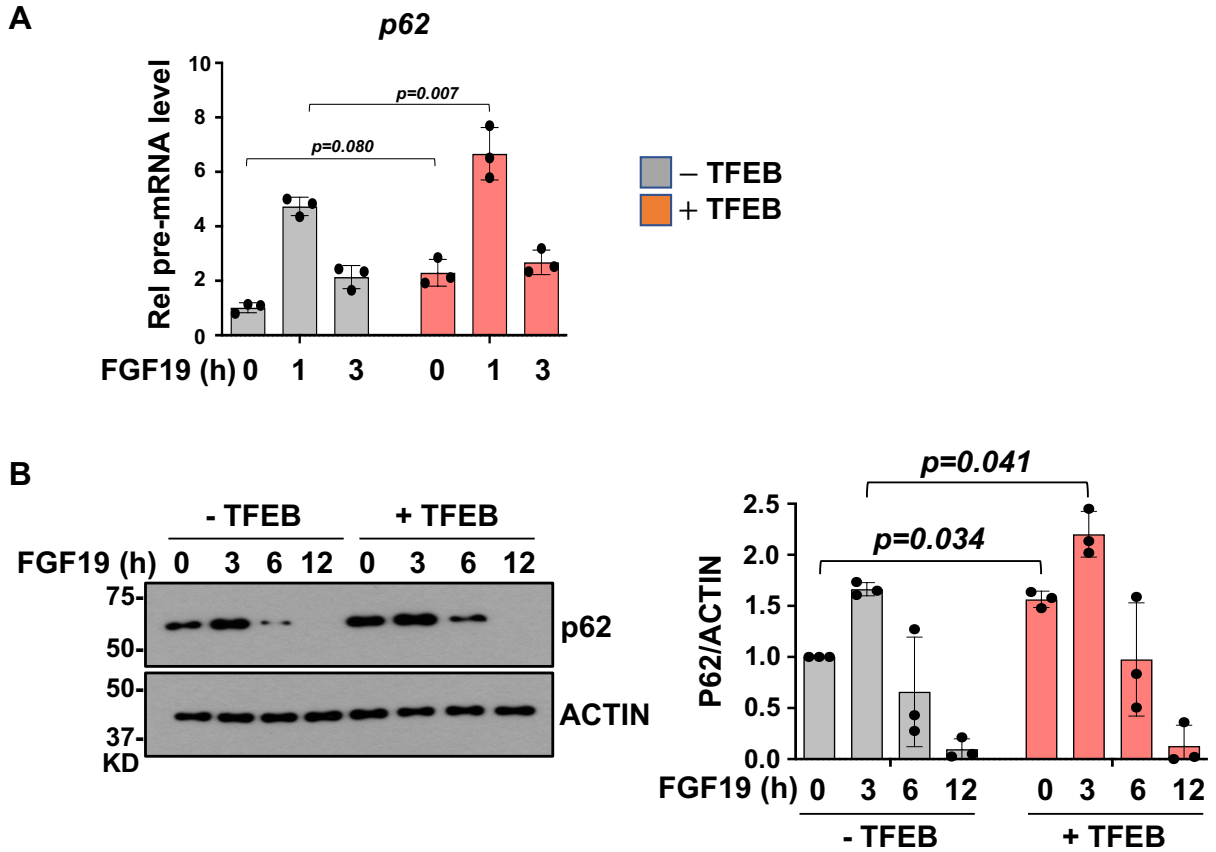


Figure S6. Effects of TFEB overexpression and/or FGF19 treatment on pre-mRNA and protein levels of p62/SQSTM1 in HT29 cells.

(A, B) HT29 cells were transfected with TFEB expression plasmids or control plasmids and incubated in serum-free medium for overnight, and then, cells were treated with 50 ng/ml FGF19 for the indicated times. Pre-mRNA (A) and protein (B) levels of p62/SQSTM1 determined by qRT-PCR and IB, respectively (n=3 culture plates). In (B), band intensities were quantified, and the relative ratios of p62/actin are shown. Statistical significance was determined by two-way ANOVA with Tukey's post-test. The p-values are indicated.

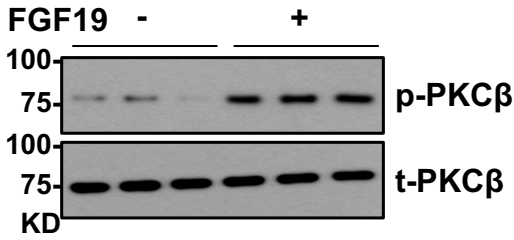
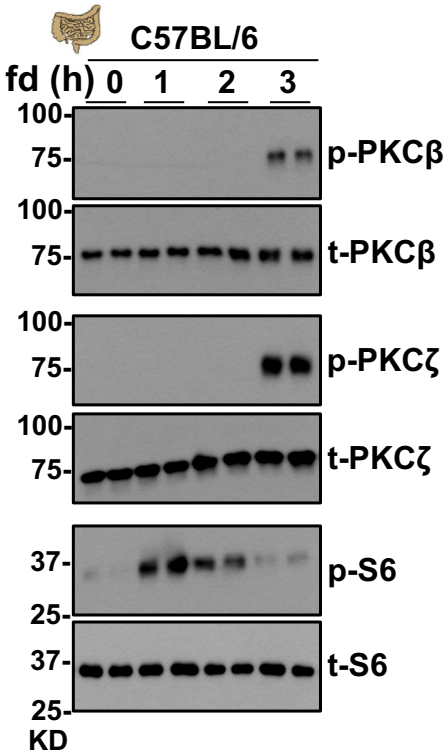
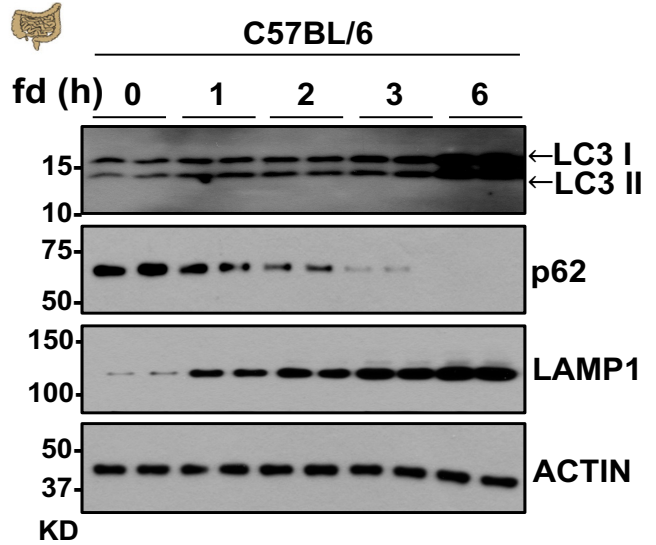
A**B****C**

Figure S7. (A) Activation of PKC β in liver hepatocytes in response to FGF15/19 signaling. Primary mouse hepatocytes were treated with 50 ng/ml FGF19 for 30 min and phospho-PKC β and total PKC β were detected by IB (n=3 culture plates). **(B, C) Temporal feeding induction of intestinal autophagy via PKC-mediated phosphorylation in mice.** C57BL/6 mice were re-fed for indicated times after fasting overnight and the indicated proteins were detected by IB (n=2 mice).

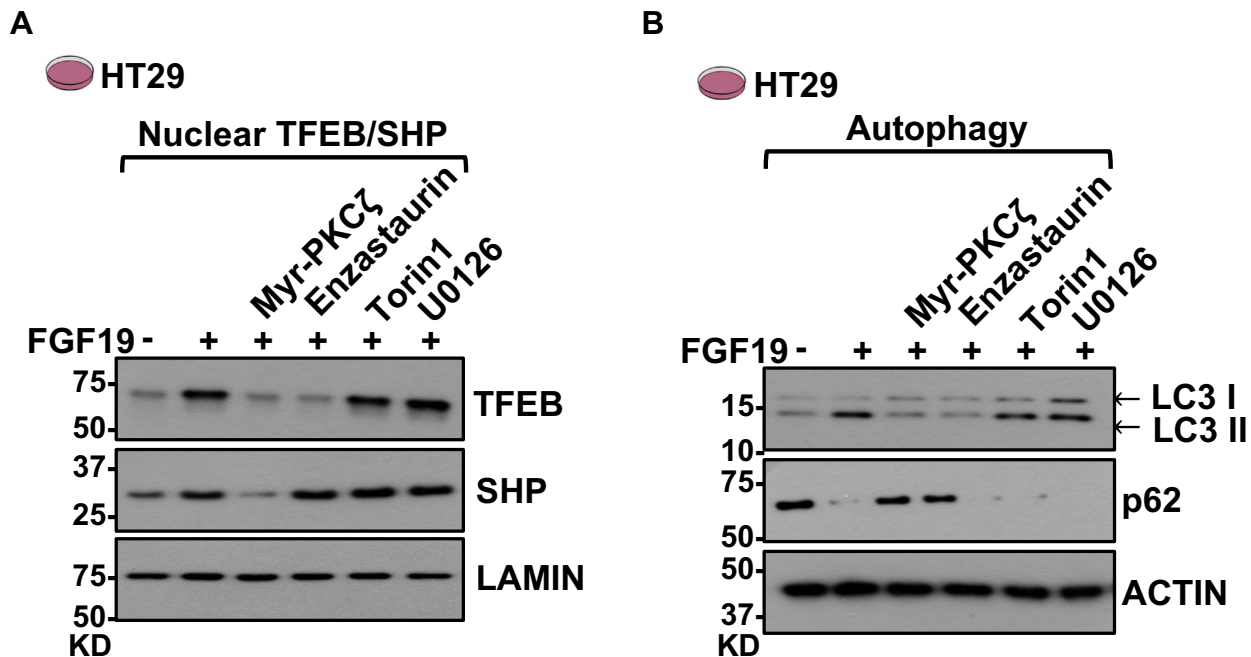


Figure S8. FGF15/19 signaling increases nuclear localization of TFEB and SHP via PKC in intestinal cells. HT29 cells were cultured overnight in serum free media and were treated with the inhibitors, Myr-PKC ζ (PKC ζ), Enzastaurin (PKC β), Torin (mTOR), and U0126 (ERK) for 30 min followed by treatment with vehicle or 50 ng/ml FGF19 for 12 h. The indicated proteins in the nuclear fractions (A) or cell extracts (B) were detected by IB. Similar results were obtained in a second experiment.

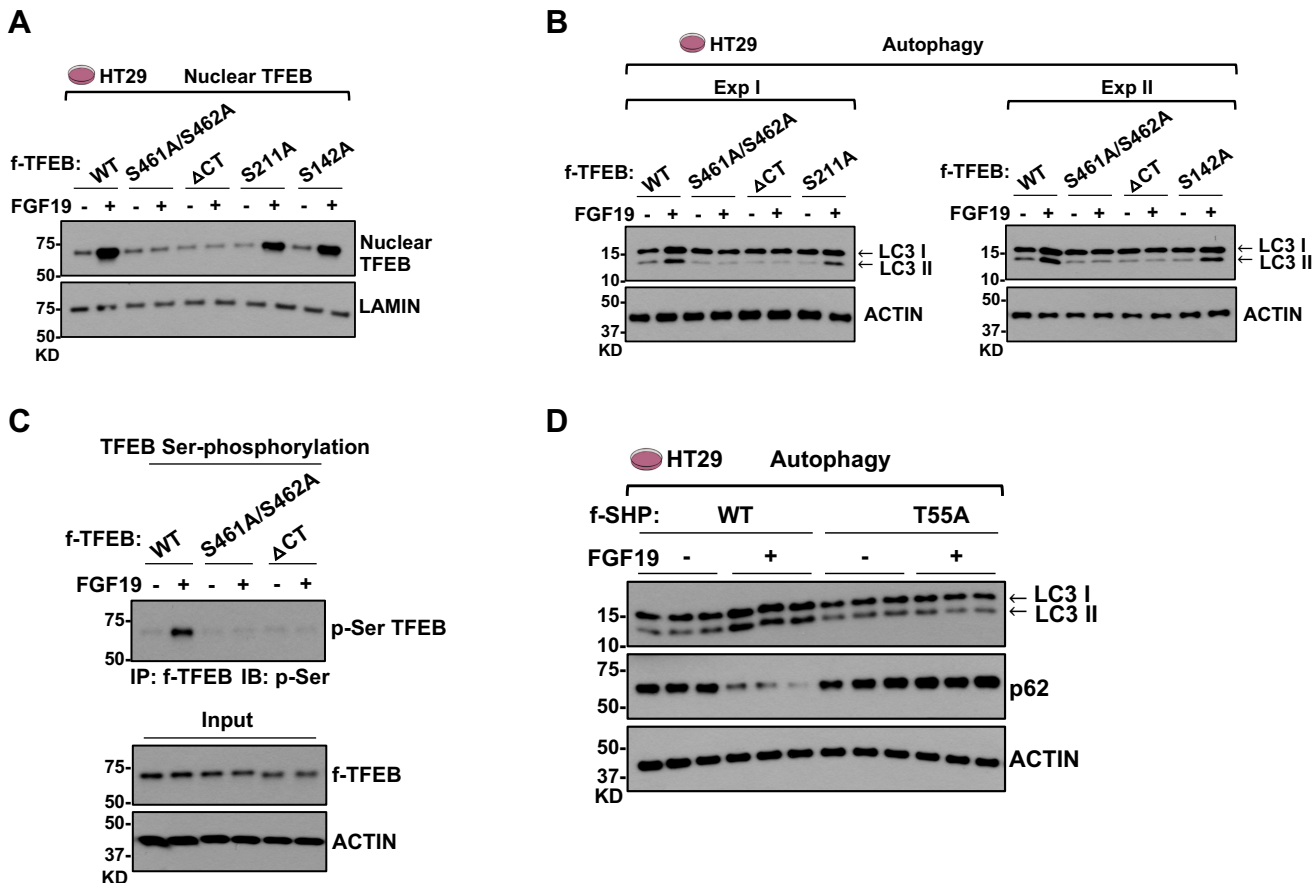


Figure S9. Effects of FGF19-induced phosphorylation of TFEB and SHP in nuclear localization and autophagy in HT29 cells.

(A, B) FGF19 treatment increases nuclear localization of both TFEB and SHP via PKC-mediated phosphorylation. The results of the experiments in the main manuscript (Fig. 8G) were repeated and confirmed. Briefly, HT29 cells were transfected with expression plasmids for flag-tagged TFEB WT and its mutants as indicated, and 24 h later, cells were treated with FGF19 (50 ng/ml) for 12 h and nuclear extracts or whole cell extracts were prepared. CT indicates C-terminal deletion (15 amino acids) in TFEB. The indicated proteins in (A) nuclear fractions or (B) cell extracts were detected by IB.

(C) FGF19-induced Ser phosphorylation of TFEB is substantially reduced by a S461A/S462A mutation or a C-terminal 15 amino acids-deletion mutant. HT29 cells were transfected with flag-tagged TFEB WT or indicated plasmids and 24 h later, cells were treated with FGF19 for 30 min. Flag-TFEB in whole cell extracts was immunoprecipitated by M2 antibody and then, p-Ser levels were detected by IB using p-Ser antibody.

(D) Increased autophagy after FGF19 treatment was largely absent with the T55A SHP mutant. HT29 cells were transfected with expression plasmids for flag-tagged SHP WT and T55A mutant, and 24 h later, cells were treated with FGF19 (50 ng/ml) for 12 h, and IB analysis was done (n=3 plates).

List of 856 genes with SHP-binding peak and down-regulated in SHP-KO
(Up-regulated intestinal genes by SHP binding)

Rb1cc1, Snhg6, Ube2w, Ogfr1l, Phf3, Mgat4a, Rpl31, Rnf149, Mfsd9, Glis, Nab1, Ankrd44, Coq10b, Mars2, 9130024F11Rik, Bmpr2, Fam117b, Pard3b, Atic, Mreg, Rpl37a, Cul3, Cab39, Ncl, Ptma, Dgkd, Arl4c, Sh3bp4, Pam, Tnfrsf11a, Phlpp1, Ralb, Ptpn4, Mgat5, Rassf5, Nucks1, Btg2, Adipor1, 5730559C18Rik, Ddx59, Kif14, Nr5a2, Zbtb41, Ivn51abp, Slc19a2, Mxzl1, Uap1, Uhmk1, Igsf8, Psen2, H3f3a, Wdr26, Lbr, Ppp2r5a, Dtl, Slc30a1, Irf6, Plekhg1, Sash1, Rab32, Utrn, Stx11, Aig1, Reps1, Pex7, Sgk1, Rnf146, Hddc2, Cdk19, Fig4, Cd164, Foxo3, Bend3, Cd24a, Gopc, Serinc1, Smpd13a, Sowahc, Dnajb12, Ddit4, Psap, Arid5b, Cdk1, Ube2d1, Ipmk, Vpreb3, Zfp280b, Slc19a1, Cirbp, Rps15, Mex3d, Btbd2, Mknk2, Eef2, Celf5, Ntn4, Eea1, Btg1, Atp2b1, Tbc1d15, Rab3ip, Cpm, BC048403, Tmem5, Ctdsp2, Mars, Naca, Cdk2, Camk2b, Ppia, H2afv, Tns3, Ppp3r1, Etaa1, Actr2, Vps54, Cct4, Asb3, Cpeb4, Wwc1, Mat2b, Slu7, Fabbp6, Ublcp1, Rnf145, Sqstm1, Mgat4b, Maml1, Ube2b, Skp1a, Sowaha, Grap, Ulk2, Akap10, 2410006H16Rik, Hs3st3b1, Aurkb, Naa38, Cldn7, Ube2g1, Ovca2, Rpa1, Crk, Cpd, Gm11437, Pigw, Appbp2, Ppm1d, Rnft1, Dhx40, Ypel2, Scpep1, Phospho1, Sp2, Pip4k2b, Rpl23, Fbxl20, Cdk12, BC030867, Fmnl1, Ern1, Axin2, Gna13, Fam20a, Slc25a19, Grb2, H3f3b, Sec14l1, Engase, Cbx2, Cbx8, Bahcc1, Narf, Rab10, Rhob, Pum2, Sdc1, Hpcal1, 5730507C01Rik, Asap2, Klf11, Id2, Rps7, Sh3yl1, Hbp1, Nampt, Sypl, Twistnb, Pnpla8, Arhgap5, Nfkbia, Foxa1, Trappc6b, Mia2, Rps29, Arf6, Nin, Ppm1a, Sgpp1, Mthfd1, Fntb, Mpp5, Zfp361l, Sipa11l, Dcaf4, Acot2, Fos, Jdp2, Calm1, Rin3, Papola, Bcl11b, Ccdc85c, Yy1, Zfyve21, Ncapg2, Klf6, Gtpbp4, Larpa4b, Elmo1, Foxq1, Slc22a23, Nedd9, Cd83, Myl1p, Kdm1b, Cks2, Gadd45g, Auh, Nfil3, Thoc3, 4833439L19Rik, Mxd3, B230219D22Rik, Pitx1, Smad5, Hnmpa0, Golm1, Ptch1, Fastkd3, Med10, Erap1, Arrdc3, Lysmd3, Cetn3, Homer1, Jmy, Arsb, Gcnt4, Hexb, Tmem174, Ocln, Pik3r1, Gbbp1, Itga2, Ptprg, Top2b, Thrb, Kcnk5, Zfp503, Rps24, Zmiz1, Anxa11, Erc2, Tmem110, Sh3bp5, Ankrd28, Ncoa4, Gmfb, Cgrrf1, Ktn1, Tmem55b, Homez, Lats2, Zdhhc20, Nupl1, Mtmr6, Kpna3, Zfp395, Cdca2, Itm2b, Lrch1, Tpt1, Ccdc122, Akap11, Dis3, Klf5, Cln5, Fbxl3, Slain1, Dnajc3, Ipo5, Stk24, Tm9sf2, Tmtc4, Rpl37, Dnajc21, Amacr, Ank, Laptm4b, Matn2, Osr2, Pabpc1, Klf10, Lrp12, Oxr1, Ext1, Mal2, Deptor, Mtbp, Zhx1, Fbxo32, Trib1, Myc, Ptp4a3, Ifi27, H1f0, Rpl3, Cacna1i, Fam83f, Tnrc6b, Polr3h, Tnfrsf13c, Arfgap3, Ppara, Celsr1, Tmem117, Asb8, Cacnb3, Eif4b, Rarg, Sp1, Pcbp2, Atf7, Srl, Tfpaf, Abat, Carhsp1, Cebpdl, Crkl, Klhl24, Ehhdh, Rfc4, Ii1rap, Ppp1r2, Pak2, Ubxn7, Tfr, Snx4, Sec22a, Dirc2, Gtf2e1, Lrrc58, Gsk3b, Nr1i2, Arhgap31, B4galt4, Plcx2d, Cd47, Bbx, Nsun3, Chmp2b, Gbe1, Usp25, Cxadr, Bach1, Hunk, Hlcs, Hmg1, Ripk4, Scaf8, Tiam2, Dact2, Flywch2, Pdpk1, Noxo1, Rps2, Sox8, Neur1b, Dusp1, Zbtb9, A1413582, Fkbp5, Kctd20, Abcg1, Kifc1, Vars, Cd2ap, Hsp90ab1, Tmem63b, Xpo5, Mocs1, Rftn1, Satb1, Rab5a, Ubxn6, Uhrf1, Rpl36, Fbxl17, Myl12b, Myl12a, Memo1, Yipf4, Crim1, Strn, Map4k3, Zfp361l, Prkce, Epas1, Msh6, Kif5b, Rock1, Polr2d, Paip2, Ndfip1, Dcp2, Ythdc2, Ccdc112, Ap3s1, Csnk1g3, Lmnb1, Slc26a2, Adb2, Apcdd1, Malt1, Impa2, Mex3c, Rpl17, Pstpip2, Nfatc1, Soccs6, Ppp1ca, Rnaseh2c, Incenp, Fads2, Prpf19, Zfp91, Ostf1, D030056L22Rik, Klf9, Fam189a2, Cdc371l, Jak2, Uhrf2, Sgms1, Atad1, Pank1, Fgfbp3, Kif11, Hhex, Cep55, Plce1, Tm9sf3, Frat1, Frat2, Erlin1, Scd1, Pcgf6, Add3, Smc3, Pdcd4, Atml1, Pd2d8, Eif3a, Nmt2, Prpf18, Gm13375, Pdss1, Abi1, Acbd5, Lrrc26, Qsox2, Agpat2, Rxra, Setx, Set, Fam78a, Slc25a25, Fam102a, Rpl12, Psmd5, Phf19, Rpl35, Epc2, Acvr1c, Cobll1, Tik1, Slc25a12, Hat1, Itga6, Pdk1, Cdca7, Ola1, Wipf1, Rbm45, Plekha3, Nckap1, Itgav, Ptprij, Madd, Chrm4, Caprin1, Fbxo3, Hipk3, Itpka, Sema6d, Bcl2l11, Mertk, Sirpa, Ptptra, Rnf24, Pcpa, Bmp2, Snx5, Ralgapa2, Fam110a, Sox12, Id1, E2f1, Eif2s2, Zhx3, Prex1, Cse1l, Ptpn1, Bmp7, Zfp704, Pag1, Fabbp5, Mtrf1, Pde7a, Nceh1, Atp11b, Sclt1, Nbea, Rnf13, P2ry1, Kpna4, Pdgc, Arhgef11, Efn3, Efn4, Ube2q1, S100a10, Mcl1, Txnip, Rnf115, Pde4dip, Notch2, Hipk1, Cttnbp2nl, Rap1a, Gnai3, Fam102b, Snx7, Usp53, Camk2d, Tifa, Gar1, Tbck, Ppa2, Pkn2, Lmo4, Hs2st1, Cyr61, Ankrd13c, Rps20, Trp53inp1, Cpne3, Bach2, Casp8ap2, Rnf38, Anp32b, Trim14, Nipsnap3b, Slc44a1, Klf4, Susd1, Akna, Ptpird, Psp1, Haus6, Dennd4c, Rps6, Cdkn2b, Usp1, Cachd1, Leprot, Mier1, Slc35d1, Ssbp3, Tmem59, Zfyve9, Osbp19, Spata6, Ipp, Gbbp1l1, Tie1, Tmem125, Edn2, Rbbp4, Marcks1, Hdac1, Serinc2, Snhg12, Sesn2, Waf2, Nudc, Hmg1, Cd52, Pdik1l, Nipal3, Rpl11, Id3, Zfp46, Pink1, Nbl1, Rcc2, H6pd, Rpl22, Fzd1, Cldn12, Dbf4, Rundc3b, Napepld, Slc5a6, Slc4a1ap, Fosl2, Ywhah, Tacc3, Mxd4, Sorcs2, Jakmip1, Hs3st1, Fbxl5, Prom1, Qdpr, Dhx15, Rel1, Klf3, Rpl9, Appb2, Gnmda2, Commdb, Usp46, Rest, Dck, Ccng2, Bmp3, Zfp644, Rpl5, Fam69a, Pcgf3, Fgfr1l, Ung, Kctd10, Acads, Unc119b, Rplp0, Vsig10, Tbx3, Rpl6, Mapkapk5, Aacs, Gusb, Cldn3, Vps37d, Fam20c, Snx8, Wipi2, Rac1, Lmtk2, Bri3, Arpc1b, Rpl21, Cdx2, Hmgb1, Brca2, N4bp2l1, Wasl, Tmem229a, Parp12, Mkn1, Ezh2, Zfp282, Cbx3, Tax1bp1, Znrf2, Lancl2, Krcc1, Rnf103, Suctg1, Mob1a, Snrpg, Pcyox1, Mxd1, Rab43, Cnbp, Klf15, Slc6a6, Nr2c2, Eif4e3, Bhlhe40, Mtmr14, Hrh1, Bid, Ptms, Etv6, Cdkn1b, Eps8, Plekha5, Aebp2, Etnk1, Stk38l, Leng9, Zfp524, Bbc3, Calm3, If2bp1, Bloc1s3, Relb, Rps19, Pfah1b3, B3gnt8, Fbl, Dyrk1b, Wdr62, Wtip, Lsm14a, Cebpg, Cebpa, Nudt19, Ccne1, Pold1, Rps11, Nubc1, Nipa2, Slco3a1, Prc1, Blm, Rps17, Mex3b, Capn5, Gdpd5, Fchsd2, Smpd1, Rrp8, Rpl27a, Nrip3, Ipo7, Swap70, Arntl, Sox6, Pik3c2a, Rps15a, Thumpd1, Mettl9, Ypel3, Dctpp1, Fam53b, Mki67, Ascl2, Kcnq1, Cdkn1c, Ccnd1, Insr, Evi5l, Rab20, Rasa3, Fbxo25, Kbtbd11, Adam9, Bag4, Gsr, Eri1, M16793, Lonrf1, Frg1, Slc25a4, Ifi2, Wwc2, Klhl2, Psd3, Uba52, Rpl18a, Tom1, Nr3c2, Rbmxl1, Otud4, Tbc1d9, Tecr, Gipc1, Lyl1, Vps35, Chd9, Mt2, Nup93, Herpud1, Cpne2, Dok4, Mmp15, Hsd11b2, Pskh1, Atnx11, Maf, Cotl1, Map1lc3b, Tmem123, Fam76b, Fut4, 4931406C07Rik, S1pr2, Acp5, Anln, Rps9, Dpy19l1, H2afx, Bud13, Fdx1, Kdelc2, Ireb2, Scaper, Tspan3, Ptpn9, 1700017B05Rik, Nptn, Neo1, Fem1b, Cln6, Kbtbd13, Spg21, Ppib, Adam10, Ccpg1, Mapk6, Elovl5, Eef1a1, Myo6, Ttk, Ctsh, Stag1, Topbp1, Rbm15b, Dag1, Dclk3, Cmtm7, Ctdspl, Rpsa, Rpl14, Snrk, Abhd5, 1110059G10Rik, Sacm1l, Suv39h1, Cul4b, Efnb1

List of 774 genes with SHP-binding peak and up-regulated in SHP-KO
(Down-regulated intestinal genes by SHP binding)

Cspp1, Npas2, Spats2l, Gm20257, Nrp2, Mettl21a, Igfbp2, Tns1, Slc23a3, Serpine2, Atg16l1, Ramp1, Per2, Traf3ip1, Asb1, Kif1a, Stk25, Tmem163, Ccnt2, Tmcc2, Dstyky, Mdm4, Ppfia4, Npl, Sec16b, Dnm3, Degs1, Hlx, Gpatch2, Ptpn14, Lpgat1, Lrp11, Hivep2, Ah1i, Tcf21, Rspo3, Ncoa7, Slc16a10, 1700021F05Rik, Sgpl1, Adora2a, Susd2, Col6a2, Rnf126, Ptbp1, Cfd, Sbn2, Dazap1, Adamts15, Dot1l, Lmn2, Gadd45b, Zfr2, Nfic, Fbxo7, Syn3, Cdk17, Zdhhc17, Phlda1, Zfc3h1, Msr3, Arhgef25, Mbd6, Baz2a, Stat2, Itga7, Smtm, Rnf215, Tbc1d10a, Ap1b1, Npc1l1, Ogdh, Tbrg4, Slc1a4, Mtif2, C1qtnf2, Trim41, Trim7, Hnrmph1, Col23a1, Irf1, Slc22a21, Zfp692, Rnf187, Guk1, 2610507101Rik, Mrip1, Flcn, Rai1, Slc47a1, Pigl, Zkscan6, Gas7, Ntn1, Myh10, Vamp2, Chd3, Tmem88, Kdm6b, Atp1b2, Nlgn2, Slc2a4, Arrb2, Mink1, Camta2, Spns2, Rtn4r1l, Tlcd2, Slc43a2, Nxn, Nufip2, Phf12, Rab34, 2610507B11Rik, Wsb1, Tbx2, Mtmr4, Cuedc1, Cacna1g, Mycbpap, Rsad1, Samd14, Fam117a, Ngfr, Arhgap23, Cacnb1, Igfbp4, Hap1, Acl, Naglu, Tubg2, Mpp2, Fam171a2, Itga2b, Gpatch8, Dcackd, Acbd4, Mapt, Itgb3, Ace, Slc16a6, Gga3, Itgb4, Galk1, Sphk1, Rbdf2, Srsf2, Socs3, Gaa, P4hb, Pcyt2, Sirt7, Rfng, Slc16a3, Hexdc, Dnajc27, Itsn2, Klhl29, E2f6, Kcnf1, Odc1, Pxdn, Prkar2b, Egl3, Prpf39, 9330151L19Rik, Actn1, Srsf5, Zfp410, Abcd4, Eml5, Ccdc88c, B930059L03Rik, Dync1h1, Amn, Ckb, Klc1, Bf1, Ptpm2, Gng4, Hist1h1c, E2f3, Foxf2, Tubb2b, Txndc5, Ptpdc1, Ror2, Cplx2, Unc5a, Uimc1, Dbn1, Fam193b, Agtpbp1, Gas1, Nkd2, Slc9a3, Ahrr, Ssbp2, Acot12, Rasgrf2, F2r, Hmgcr, Gfm2, Nnt, Vdac2, Arf4, Glt8d1, Syt15, Fermt2, Bmp4, Samd4, Sall2, Pabpn1, Nfatc4, Rcbtb1, Dleu2, Gata4, Ephx2, Zc3h13, Dgkh, Gpr180, Fam105a, Sdc2, Spag1, Rims2, Taf2, Sqle, St3gal1, Gpr20, Gsdmd, Scrib, Plec, Dgat1, Gpt, Mfsd3, Txn2, Gga1, Gcat, Ankrd54, Sox10, Pick1, Pla2g6, Atf4, Sgsm3, Sreb2, Scube1, Smc1b, Fbln1, CerK, Trabd, Mapk12, Sbf1, Odf3b, Slc38a2, Adcy6, Tuba1c, Prpf40b, Nckap5l, Csrnp2, Fignl2, Itga5, Ppp1r1a, Slx4, Glis2, Pam16, Vasn, Txndc11, Rm3, Tmem191c, Scarf2, Comt, Cldn5, Clcn2, Senp2, Bcl6, 4632428C04Rik, Tmem44, Dlg1, Adcy5, Boc, Nfkbiz, Son, Setd4, Bace2, Pde10a, Dll1, Fam120b, E4f1, Caskin1, Zfp598, Tpsg1, Rbhd1f, Rhot2, Wdr90, Ggnbp1, Itpr3, Pacsin1, Spdef, Mapk13, Brpf3, Brd4, Akap8, Zfp414, Rgl2, Col11a2, H2-Ab1, Notch4, Prrt1, Tnxb, Msh5, Clic1, Ddah2, Ddx39b, H2-Q1, H2-Q4, Ier3, Ppp1r18, Dhx16, Prr3, H2-B1, 2410017117Rik, Gm8909, Trim39, Runx2, Abcc10, Pex6, Tbc1d5, Plin4, Ptpm3, Safb2, Nrtn, Efna5, Cdc42ep3, Morn2, Plekhh2, Ttc7, Zeb1, Gpr17, Dnajc18, Psd2, Ndufa2, Yipf5, Zfp608, Prrc1, Slc6a7, Ablim3, Zbtb7c, Atp9b, Mbp, Zfp236, Fam69c, 1810055G02Rik, Chka, Acy3, Cam31, Tmem151a, Sssca1, Malat1, Neat1, Syvn1, Cdc42bpq, Map4k2, Nrnx2, Esrra, Gpr137, 2700081O15Rik, Slc3a2, Nxf1, Cpsf7, Ddb1, Fam122a, Asah2, Rbp4, Hps1, Cnnm1, Nkx2-3, Entpd7, Kazald1, Slk, Dusp5, Tcf7l2, Gfra1, Csf2ra, Sec61a2, Pfkfb3, Arrdc1, Inpp5e, Surf4, Olfm1, Tor1b, Fnbp1, Ncs1, Ass1, Dnm1, Zbtb43, Nr6a1, Fmnl2, Grb14, Stk39, Klhl23, Sp5, Gorasp2, Cybrd1, Agps, Osbpl6, Fnbp4, Slc39a13, Mdk, Creb3l1, Phf21a, Gm13889, Ino80, Ltk, Pla2g4b, Vps39, Ckmt1, Shf, Fbn1, Slc27a2, Slc20a1, Gpcpd1, Gm5535, Rbck1, Snta1, Acss2, Cep250, Rbm39, Srsf6, Zfp335, Ncoa5, Slc2a10, Zmynd8, Ptgis, Slc9a8, Snai1, Ctsz, Car2, Pex5l, Ttc14, Spry1, Lar1b, Pgrmc2, Smad9, Ccnl1, Dclk2, Trim46, Crtc2, Cgn, Plekho1, Sec22b, Ppm1j, Slc6a17, Gstm5, Scarna2, Prpf38b, Rnpc3, Agl, Arhgap29, Sec24d, Prss12, Ank2, Gm5105, Fubp1, Rabggtb, Srsf11, Chd7, Ndufaf4, Cnr1, Topors, Nfx1, Enhf, Dnajb5, Unc13b, Rmrp, Tmem8b, Reck, Fbxo10, Stx17, A1427809, Col27a1, Tle1, E130102H24Rik, Ak4, Prkaa2, Acot11, Podn, Cc2d1b, Mob3c, Ipo13, Foxo6, Scmh1, Zfp69, Mfsd2a, BC002163, Mrps15, Stk40, Col8a2, AU040320, Sdc3, Ptpm3, Map3k6, Trp1, Arid1a, Zbtb40, Wnt4, Hspg2, Eif4g3, Sh2d5, Otud3, Tas1r2, Crocc, Zbtb17, Spen, B330016D10Rik, Tmem51, Tardbp, Casz1, Cttnbip1, Slc2a5, Gpr153, Prdm16, Mib2, Tmem88b, Ccnl2, Mxra8, Dvl1, Agrn, Klhl17, Dmtf1, 5031425E22Rik, Rint1, Kcnh2, Cdk5, Fastk, Cnpy1, Dpysl5, Tmem214, Zfp513, Krtcap3, Fgfr3, Ablim2, D5Ert2579e, Atp8a1, Guf1, Sgcb, Rasl11b, Cep135, Cox18, Uso1, Sec31a, Gfi1, Fbrsl1, Ep400, Mn1, Sgsm1, Mvk, Trpv4, Mlec, Cux2, Camkk2, Diabolo, Vps33a, Clip1, Ccdc62, Abcb9, Pitpnm2, Rilpl1, Ddx55, Scarb1, Bri3bp, Sfsfap, Tbl2, Nsun5, Pom121, Trip6, Ggfyf1, Pdgfra, Cyth3, Usp42, Aimp2, Nptx2, Met, Arf5, Calu, Smo, Luc7l2, Casp2, Zfp956, Krba1, Tril, Aqp1, Tmem150a, Vamp5, Tmsb10, Mgl1, Podxl2, Prickle2, Pdzr3, Itpr1, Gt(ROSA)26Sor, Sec13, Syn2, Zfp239, Cacna1c, Zfp384, Ltbr, Vwf, Tulp3, Dusp16, Rerg, 2810474O19Rik, Leng1, Leng8, Brsk1, U2af2, Zc3h4, Ppp5c, Dmpk, Six5, Ercc1, Clasrp, Bcam, Cadm4, Zfp428, Megf8, Numb1, Ltbp4, Plekhg2, Zfp36, Paf1, Rinl, Capn12, Aplp1, Gramd1a, Lrp3, Plekhf1, Jsd2, Atf5, Cpt1c, Irf3, Snmp70, Rasip1, Lmtk3, Grin2d, Nav2, Tarsl2, Synm, Chd2, Polg, Cib1, Unc45a, Hddc3, Man2a2, Slc28a1, 2900076A07Rik, Pcf11, Inpp1l, Trim30d, Fam160a2, Apbb1, Trim3, Mrpl17, Coq7, Tnrc6a, Atxn2l, Taok2, Setd1a, Stx4a, Rgs10, Tial1, Fgfr2, Bet1l, B4galnt4, Eps8l2, Muc6, Igf2, Tssc4, Tpcn2, Col4a2, Col4a1, Cars2, Mcf2l, Slc25a15, Ank1, Hook3, Rbpms, Ppp1r3b, Hand2, Palld, Tmem161a, Rab3a, Unc13a, Cherp, Rasd2, Zfp827, Scoc, Ptger1, Rfx1, Dcaf15, Cacna1a, Ier2, Hook2, N4bp1, Nkd1, Gnao1, Ciapin1, Ccdc102a, Cngb1, Hsf4, Zdhhc1, Slc12a4, Esrp2, Slc7a6os, Nfat5, Chst4, Znr1, Necab2, Atp2c2, Irf8, Banp, Zfpm1, Tubb3, Galnt2, Sipa1l2, Kank2, Elavl3, Barx2, 4930581F22Rik, Tbrg1, Mcam, Rexo2, Dnaja4, Chma3, Cspg4, Scamp5, Clk3, Sema7a, Loxl1, Parp6, Myo9a, Rasl12, Rbpms2, Fbxl22, Tpm1, Bnip2, Sltm, Aldh1a2, Myo5c, Bckdhb, Trpc1, Amotl2, Uba5, Pcbp4, Dock3, Ifrd2, Sema3f, Rbm6, Mst1r, Bsn, Celsr3, Dhx30, Scap, Ptpn23, Pth1r, Clasp2, Trak1, Nktr, Zfp651, Gpc4, G530011O06Rik, Slc10a2, Abcg5, Abca1, Acat2

Table S3.

**34 unique motifs for down-regulated intestinal
genes by SHP binding**

MOTIF Name	p-value	MOTIF Name	p-value
Tp53_DBD_3	3.21E-05	ESR1	3.65E-03
TP53	3.21E-05	RARA_DBD_1	3.99E-03
ZEB1	1.85E-04	Zic2_secondary	4.24E-03
TFAP2B_DBD_2	2.98E-04	TCF4_full	4.33E-03
TFAP2B(var.2)	2.98E-04	SP3_DBD	4.37E-03
Tcf1_secondary	4.91E-04	SP3	4.37E-03
SP2	8.19E-04	Mrg2_2302.1	4.87E-03
Tp73_DBD	1.15E-03	CTCF	4.91E-03
TP73	1.15E-03	Esrra_DBD_2	4.97E-03
TFAP2B_DBD_3	1.44E-03	Esrra	4.97E-03
TFAP2B(var.3)	1.44E-03	MYC	5.13E-03
Zfp281_primary	1.71E-03	ZIC3_full	5.51E-03
NRF1_full	2.00E-03	Zic3_DBD	5.51E-03
Nr2e3	2.35E-03	ZIC3	5.51E-03
Zfp740_primary	2.42E-03		
KLF16_DBD	2.44E-03		
NHLH1_DBD	2.47E-03		
KLF16	2.58E-03		
Gfi1b	3.57E-03		
ESR1_DBD	3.65E-03		

Table S4.

63 unique motifs for up-regulated intestinal genes by SHP binding

MOTIF Name	p-value	MOTIF Name	p-value	MOTIF Name	p-value
BHLHE41_full	3.12E-06	Bhlhb2_DBD_2	3.11E-05	SMAD3_DBD	8.11E-04
BHLHE41	3.12E-06	USF1_DBD	3.11E-05	SMAD3	8.11E-04
MLX_full	5.55E-06	BHLHB3_full	3.57E-05	Id2	9.61E-04
Mlx_DBD	5.55E-06	TFEC_DBD	4.08E-05	Myod1	9.69E-04
MLX	5.55E-06	TFEC	4.08E-05	Plagl1_secondary	1.30E-03
ARNTL_DBD	9.24E-06	Zscan4_primary	4.57E-05	Nkx2-5(var.2)	1.46E-03
TFE3_DBD	9.24E-06	Gm397_primary	4.57E-05	Mlxip	2.57E-03
SREBF2_DBD	1.41E-05	TBX1_DBD_4	5.16E-05	RARG_full_3	2.58E-03
SREBF2(var.2)	1.41E-05	Arnt	6.53E-05	Tcf21_DBD	2.60E-03
MLXIPL_full	1.69E-05	MTF1_DBD	9.52E-05	Tcf21	2.60E-03
Srebf1_DBD	1.69E-05	MTF1	9.52E-05	Rxra_primary	2.90E-03
TFEB_full	1.69E-05	MITF	1.31E-04	Smad3_secondary	3.00E-03
MLXIPL	1.69E-05	USF2	1.39E-04	Rarg_DBD_3	3.20E-03
TFEB	1.69E-05	Ascl2_primary	1.46E-04	Rarg(var.2)	3.20E-03
Srebf1(var.2)	1.69E-05	TFE3	2.52E-04	GCM1_full_1	3.67E-03
BHLHB2_DBD	1.99E-05	Arntl	2.99E-04	YY2_full_2	3.80E-03
Bhlhb2_DBD_1	1.99E-05	Mtf1_primary	3.11E-04	TBX21_DBD_3	4.48E-03
BHLHE40	1.99E-05	USF1	3.14E-04	POU6F2_DBD_2	4.94E-03
Bhlhb2_primary	2.77E-05	Ascl2_DBD	7.22E-04	MEIS2_DBD_2	5.10E-03
ZNF306_full	3.01E-05	Ascl2	7.22E-04	MEIS2	5.10E-03
				GATA1::TAL1	5.28E-03
				EOMES_DBD_2	5.31E-03
				YY1	5.37E-03

Table S5. Information on antibodies

Antibody	Company	# of catalogue	dilutions (IB)
SHP	Abcam	ab96605	1:5000
β-tubulin	Santa Cruz Biotechnology	sc-5274	1:3000
LAMIN A	Santa Cruz Biotechnology	sc-518013	1:3000
PKC	Santa Cruz Biotechnology	sc-13149	1:3000
LAMP1	Abcam	ab24170	1:5000
p-PKCb	Abcam	ab194749	1:5000
ACTIN	Cell Signaling	#4970	1:5000
LC3-I/II	Cell Signaling	#4108	1:5000
p62	Cell Signaling	#5114	1:5000
TFEB	Cell Signaling	#37785	1:5000
ULK1	Cell Signaling	#8054	1:5000
p-PKCz	Cell Signaling	#9378	1:5000
PKCz	Cell Signaling	#9368	1:5000
p-S6	Cell Signaling	#4857	1:5000
S6	Cell Signaling	#2217	1:5000
p-ERK	Cell Signaling	#9101	1:5000
ERK	Cell Signaling	#4695	1:5000
APOB48	ThermoFisher	MA5-35458	1:5000
anti-Rabbit IgG secondary	Cell Signaling	#7074S	1:5000
anti-Mouse IgG secondary	Santa Cruz Biotechnology	sc-516102	1:10000

Antibody	Company	# of catalogue	dilution (ChIP)
SHP	Abcam	ab96605	1:100
	Santa Cruz Biotechnology	sc-30169	3μg
RNA pol II	Novus biologicals	NB200-598	2μg
LSD1	Abcam	ab17721	2μg
Histone H3	Abcam	ab1791	1μg
ZKSCAN3	Abcam	ab223477	3μg
TFEB	Cell Signaling	#37785	1:50
G9A	Millipore	07-551	2μg
H3K9-me2	Millipore	07-030	2μg

Antibody	Company	# of catalogue	dilution (IHC)
LC3-I/II	Cell Signaling	#4108	1:100

Antibody	Company	# of catalogue	dilution (IF)
anti-Perilipin 2	Progen	GP40	1:100
anti-Guinea pig IgG H&L	Abcam	ab150187	1:500

Table S6. Information on reagents

Reagents	Company	# of catalogue
ON-TARGETplus siRNAs for <i>Atg7</i>	Dharmacon	L-020112-00-0005
ON-TARGETplus siRNAs for Tfeb	Dharmacon	L-009798-00-0005
ON-TARGETplus siRNAs for Shp	Dharmacon	L-003410-00-0005
ON-TARGETplus siRNAs for Lamp1	Dharmacon	L-013481-02-0005
Myr-PKC ζ	Thermo fisher scientific	77749
Enzastaurin	Tocris	5994
Torin1	Tocris	4247
U0126	Tocris	1144
PMSF	Sigma	P7626
Aprotinin	Calbiocam	616370
Leupeptin	Calbiocam	108976
Pepstatin A	AG scientific	P1519
NaF	Sigma	S6776
Na ₃ VO ₄	Sigma	450243 or S6508
Na ₂ MoO ₄	Sigma	M1003
Na ₄ P ₂ O ₇	Sigma	221368

Table S7. Primer sequences used for ChIP-qPCR and qRT-PCR.

A. List of primer sequences for mouse ChIP-qPCR

Gene	Forward (5'-3')	Reverse (5'-3')
<i>Slc9a3(NHE3)</i>	ACTGCCAGTGGGCTAGATGT	CAAACAGTCCTGCTGCGTTA
<i>Slc10a2(Asbt)</i>	TTACCTCACCTGGTGGCAGT	CCGCTCCTCCTGATTCTATG
<i>Acly</i>	TAGACCCGCACAAACTTCC	GGTGCTTACGGACAGAGAGC
<i>Dgat1</i>	GGGGGTAGATATGGGCACT	CCCTGAGATACGAGGCTCTG
<i>Npc1l1</i>	ATGCCCCGTTCTATCCCTTTC	CAGGTTGGGTGGGACTTG
<i>Acat2</i>	CGAGCTGTGGAGTCTGTGG	CAGAGCGACAAGATGAATGC
<i>Ulk1</i>	CTGACACACTCCAGCTCCAA	CGCGGATCTTTTGTTTCC
<i>Wipi2</i>	CATCCACCTGTCCTGAGACC	CATGAAAAGCGACAGAAGCA
<i>Nampt</i>	ACAGGGAGGACTGGATGAGA	CTAGGTGCAAGGGAGTTGGA
<i>Arntl (Bmal1)</i>	TCCGTTGCTCTCTCTGATT	ACCTCCGTCCTGACCTACT
<i>Psen2</i>	AAAACACTGGGTGCAGAAGG	GCAGAGGCTCCTGAAATCAC
<i>Ppara</i>	AATGTGCCTGTAGGGTCTGG	TGGGAACGAAATGTGTGCTA
<i>Pnpla2(Atgl)</i>	GCTCATGGCATTCTTTGGT	TCCTCTCTCCCTCGCTCATA
<i>Rock1</i>	GAAACAGCAGCACTCACCA	GGGGACAGTTTTGAGACTCG

B. List of primer sequences for mouse qRT-PCR

Gene	Forward (5'-3')	Reverse (5'-3')
<i>Slc9a3(NHE3)</i>	ACAGAAGCGGAGGAATAGCA	TATCAATTCTGCCCCAGAG
<i>Slc10a2(Asbt)</i>	GACTCGGGAACGATTGTGAT	GGTTCAATGATCCAGGCACT
<i>Acly</i>	AGGTCTCTCTGCAGCCATGT	AAGCTTTCCTCGACGTTTGA
<i>Dgat1</i>	GCTTCTGCAGTTTGGAGACC	CTCATGGAAGAAGGCTGAGG
<i>36b4</i>	CGACATCACAGAGCAGGC	CACCGAGGCAACAGTTGG
<i>Npc1l1</i>	GCAAGGTGATCAGGAGGTTGA	ATCCTCATCCTGGGCTTTGC
<i>Acat2</i>	CAGAGCGACAAGATGAATGC	CGAGCTGTGGAGTCTGTGG
<i>Ulk1</i>	TTCTGTGAGTCTGGCTCCT	GAAACCCACATCAGCTTGT
<i>Wipi2</i>	TGAATTGAGGGAGAGGATGG	GCAAAGCAAGGAGACTCCAC
<i>Nampt</i>	AGGGGTTAGCAGGTGATCCT	TAGTGCGGGCTAGCCTAAAA
<i>Arntl (Bmal1)</i>	CGAAGACAATGAGCCAGACA	AAATAGCTGTCGCCCTCTGA
<i>Psen2</i>	GGAGGATGGAGAGAGCACTG	CCACCACGATCATACACAGC
<i>Ppara</i>	TCTGTGGGCTCACTGTTCTG	AACTACCTGCTCAGGGCTCA
<i>p62 (pre-mRNA)</i>	CGAGTGCTGTGCTGTTT	TCGCCACAGCTACTACTCC