Supplementary Figure 1

**a** Relative expression of Lats1, Lats2, Yap, and Taz in vWAT of WT and ob/ob mice. P = 0.084.

**b** LATS1 and LATS2 immunoblot analysis of proteins in vWAT of WT and ob/ob mice. Relative density is normalized to HSP90.

**c** Relative density of pYAP/YAP and pTAZ/TAZ is normalized to HSP90 in vWAT of WT and ob/ob mice.

**d** Relative density of pYAP/YAP and pTAZ/TAZ is normalized to HSP90 in vWAT of WT and ob/ob mice.

**e** Relative expression of Lats1, Lats2, Ya, and p1 in scWAT of WT and ob/ob mice. P = 0.076.

**f** LATS1 and LATS2 immunoblot analysis of proteins in scWAT of WT and ob/ob mice. Relative density is normalized to HSP90.

**g** Relative density of pYAP/YAP and pTAZ/TAZ is normalized to HSP90 in scWAT of WT and ob/ob mice.

**h** Relative density of pYAP/YAP and pTAZ/TAZ is normalized to HSP90 in scWAT of WT and ob/ob mice.

**i** Relative expression of Lats1, Lats2, Ya, and p1 in vWAT of WT and ob/ob mice. P = 0.070.

**j** Relative density of pYAP/YAP and pTAZ/TAZ is normalized to HSP90 in vWAT of WT and ob/ob mice.

**k** Relative density of pYAP/YAP and pTAZ/TAZ is normalized to HSP90 in vWAT of ND and HFD mice.

**l** Relative expression of Lats1, Lats2, Ya, and p1 in vWAT of ND and HFD mice.

**m** Relative density of pYAP/YAP and pTAZ/TAZ is normalized to HSP90 in vWAT of ND and HFD mice.
Supplementary Figure 1. Hippo pathway is inactivated in obesity-induced AT fibrosis.

a-d, Male WT or ob/ob mice were analyzed at 12 weeks old. **a**, RT-qPCR analysis of genes involved in Hippo pathway and fibrotic response in vWAT (n=5 mice). **b**, Immunoblot analysis of LATS1/2, YAP/TAZ, p-YAP and p-TAZ in vWAT. **c**, Quantification of protein expression from vWAT shown in (b) (n=4 mice). **d**, Quantification of phosphorylation level of YAP/TAZ (n=4 mice). e-j, Female WT or ob/ob mice were analyzed at 12 weeks old. **e**, RT-qPCR analysis of genes involved in Hippo pathway and fibrotic response in (e) scWAT or (h) vWAT (n=6 mice). **f**, Immunoblot analysis of LATS1/2, YAP/TAZ, p-YAP and p-TAZ in (f) scWAT or (i) vWAT, respectively. **g**, Quantification of protein expression from (g) scWAT or (j) vWAT shown in (f) and (i) (n=6 mice). **k**, RT-qPCR analysis of genes involved in Hippo pathway and fibrotic response in vWAT (n=6) obtained from male mice fed a HFD or ND for 18 weeks. **l**, Immunoblot analysis of LATS1/2, YAP/TAZ, p-YAP and p-TAZ in vWAT of ND and HFD mice (n=3 mice). **m**, Left, quantification of protein expression from scWAT or vWAT shown in (l); right, quantification of phosphorylation level of YAP/TAZ (n=3 mice). Data are means ± SEM. Two-tailed unpaired student’s t-test; *P < 0.05, **P < 0.01, ***P < 0.001. Exact P values are provided in a Source Data file.
Supplementary Figure 2

a) Heatmap showing correlations between gene expression levels. The heatmap includes genes such as ACTA2, COL1A1, COL1A2, COL6A1, COL6A2, COL6A3, COL1A3, COL6A2, COL6A3, CENPA, AXL, BIRC3, BIRC5, C5, CAV1, CAV2, CCM1, DAB2, DD3, DLC1, DUSP1, ECT2, EMP2, ETV5, FG2, FLNA, FSCN1, FSTL1, GADD45B, GAS6, GGH, GLS, HEXB, HIMR, ITG4B, ITG5B, LHRP2, MARCKS, NDRG1, NDRG2, PDLIM2, PMP22, SCHIP1, SERPINE1, ZG1, SH2D4A, SHGBP1, SLIT2, STN1, TGN2, THBS1, TK1, TNNT2, TNFS1, TOP2A, TSPAN3.

b) Heatmap showing correlations between gene expression levels. The heatmap includes genes such as WWTR1, YAP1, AGFG2, AMOTL2, ANKRD1, ASAP1, AXL, BCC1, BIRC3, BIRC5, C5, CAV1, CAV2, CCM1, DAB2, DD3, DLC1, DUSP1, ECT2, EMP2, ETV5, FG2, FLNA, FSCN1, FSTL1, GADD45B, GAS6, GGH, GLS, HEXB, HIMR, ITG4B, ITG5B, LHRP2, MARCKS, NDRG1, NDRG2, PDLIM2, PMP22, SCHIP1, SERPINE1, ZG1, SH2D4A, SHGBP1, SLIT2, STN1, TGN2, THBS1, TK1, TNNT2, TNFS1, TOP2A, TSPAN3.

c) Scatter plot showing the correlation between COL6A3 (TPM) and WWTR1 (TPM). The correlation coefficient is p=0.0269 and R^2=0.1956.

d) Scatter plot showing the correlation between COL6A3 (TPM) and YAP1 (TPM). The correlation coefficient is p=0.0444 and R^2=0.1643.

e) Scatter plot showing the correlation between COL6A3 (log2cpm) and WWTR1 (log2cpm). The correlation coefficient is p=0.0278 and R^2=0.1077.

f) Scatter plot showing the correlation between COL6A3 (log2cpm) and YAP1 (log2cpm). The correlation coefficient is p=0.0091 and R^2=0.1479.
Supplementary Figure 2. Genes involved in Hippo pathway and in AT fibrosis are positively correlated in humans.

a, b, Heatmap analysis of genes involved in ECM remodeling (a) and Hippo pathway (b, molecular signatures database: cordenonsi_YAP_conserved_signature) extracted from the RNAseq data (TPM values) of abdominal scWAT of 9 healthy lean individuals (Lean) and 8 obese normal glucose tolerance subjects (Obese NGT) (Gene Expression Omnibus (GEO) repository, accession number GSE141432). c-f, Correlation analysis between genes involved in Hippo pathway and ECM remodeling, using publicly available transcriptomic data. c, d, RNAseq of scWAT of 25 subjects (GSE141432). e, f, RNAseq of scWAT of 45 subjects (GSE152991). TPM, transcripts per million. CPM, read counts per million. Linear regression analysis (c-f); significance was calculated by two-tailed unpaired student’s t test.
Supplementary Figure 3

- **a** L1L2-FF L1L2-AKO Lats1
- **b** L1L2-FF L1L2-AKO Lats2
- **c** L1L2-FF L1L2-AKO
- **d** L1L2-FF L1L2-AKO
- **e** L1L2-FF L1L2-AKO
- **f** Survival (%) Observation period (days after birth)
- **g** Liver L1L2-FF L1L2-AKO
- **h** scWAT vWAT BAT L1L2-FF L1L2-AKO
- **i** L1L2-FF L1L2-AKO
- **j** Pro-CASP3 Cleaved CASP3 HSP90
- **k** L1L2-FF L1L2-AKO Picrosirius red staining
- **l** Acta2 Col1a1 Col1a2 Col1a3 Col3a1 Col3a2 Col6a1 Col6a2 Col6a3 Eln Fn1 Tgfb1 Tgfb2 Tgfb3 Itga3 Itga4 Itga5 Itga6 Itga7 Itga8 Itgah Itgai Itgb1 Itgb2
- **m** L1L2-FF L1L2-AKO
- **n** L1L2-FF L1L2-AKO
- **o** WT L1-AKO L2-AKO Adipocyte marker Fibrosis marker
- **p** WT L1-AKO L2-AKO H&E
- **q** WT L1-AKO L2-AKO Masson’s trichrome
Supplementary Figure 3. Characterization of Lats1/2-deficient mice.

**a, b**, mRNA expression of *Lats1* (**a**) and *Lats2* (**b**) in scWAT, BAT, liver, muscle, spleen, kidney and heart in male P7 L1L2-FF and L1L2-AKO mice (*n*=3). **c**, mRNA expression of *Lats1* and *Lats2* in mature adipocytes isolated from P7 L1L2-FF or L1L2-AKO scWAT (*n*=3). Three male mice were pooled as one sample. **d-k**, Characterization of 5-week-old male L1L2-AKO and L1L2-FF mice. **d**, Gross appearance of mice. **e**, Quantification of body weight L1L2-AKO (*n*=7) and L1L2-FF (*n*=10) mice. **f**, Survival analysis (*n*=19). **g**, Representative liver sections with H&E staining. **h**, Representative images of scWAT, vWAT, BAT. **i**, Plasma non-esterified free fatty acids (NEFA) levels (*n*=6 mice). **j**, Immunoblot analysis of Pro-Caspase 3 (Pro-CASP3) and Cleaved Caspase 3 (CASP3) in scWAT (*n*=3 mice). **k**, Representative scWAT sections with Picrosirius red staining. Independent experiments were performed twice with similar results. **l**, ECM remodeling genes extracted from the RNAseq data (TPM values) of scWAT adipocytes of P7 L1L2-FF (FF) and L1L2-AKO (KO) mice. **m, n**, Body size (**m**) and body weight (**n**) of P21 male L1L2-FF and L1L2-AKO mice (*n*=6). **o**, mRNA expression of *Lats1/Lats2*, adipocyte markers and fibrotic markers of scWAT in 5-week-old male L1-AKO or L2-AKO mice (*n*=5). **p, q**, Representative sections of scWAT with Masson’s trichrome (**p**) or H&E staining (**q**). Independent experiments were performed twice with similar results. Data are means ± SEM. Two-tailed unpaired student’s *t*-test in (**a-c, e, i, n**); One-way ANOVA with Bonferroni’s multiple-comparisons test in (**o**); *P* < 0.05, **P** < 0.01, ***P*** < 0.001; NS, not significant. Exact *P* values are provided in a Source Data file.
Supplementary Figure 4

(a) Western blot analysis of MST1, MST2, and HSP90 in WT and ob/ob mice. Relative density (Normalized to HSP90).

(b) Western blot analysis of MST1, MST2, and HSP90 in ND and HFD conditions. Relative density (Normalized to HSP90).

(c) Western blot analysis of MST1, MST2, and HSP90 in SVF and Adipocyte samples. Relative density (Normalized to HSP90).

(d) Western blot analysis of MST1, MST2, p-YAP, YAP, and HSP90 in M1M2-FF and M1M2-AKO conditions. Relative density (Normalized to HSP90).

(e) H&E and Masson's trichrome staining images of M1M2-FF and M1M2-AKO samples. Scale bars: 100 μm.

(f) Relative expression levels of various genes in M1M2-FF and M1M2-AKO samples. NS indicates no significant difference.
Supplementary Figure 4. Lats1/2 deficiency-induced AT fibrosis does not depend on MST1/2.

a, b, Immunoblot analysis of MST1 and MST2 in 12-week-old male ob/ob mice (a, n=4) and male mice fed a HFD for 18 weeks (b, n=3). Right, quantification of protein levels of MST1/2. The asterisks indicated non-specific bands. c, Immunoblot analysis of protein expression of MST1 and MST2 in scWAT adipocytes and SVF isolated from 5-week-old male WT mice. Right, quantification of relative protein expression of MST1 and MST2 (n=3 mice). d, Immunoblot analysis of the indicated protein in scWAT of 5-week-old male M1M2-FF and M1M2-AKO mice. Right, quantification of the indicated protein expression (n=3). e, Representative sections of scWAT with H&E or Masson’s trichrome staining. Independent experiments were performed twice with similar results. f, RT-qPCR for adipocyte and fibrotic marker gene expression in scWAT (n=6 mice). Data are means ± SEM. Two-tailed unpaired student’s t-test; *P < 0.05, **P < 0.01, ***P < 0.001; NS, not significant. Exact P values are provided in a Source Data file.
Supplementary Figure 5. LATS1/2 maintain adipocyte identity in a cell-autonomous manner.

a, Schematic overview of AAV infection of Cas9Tg/+ SVF undergoing differentiation. b, Immunoblot analysis of time-course expression of TAZ, p-YAP, and LATS1 in Vec or LATS1/2-gRNA (L1L2-gRNA1) transduced cells. Independent experiments were performed three times with similar results. c, RT-qPCR analysis of adipocyte identity and fibrosis markers of differentiated adipocytes transduced with Vec, L1L2-gRNA1 and L1L2-gRNA2 on day 2 post differentiation, and then analyzed 5 days later (n=3 biologically independent cell cultures). d, Immunoblot analysis of protein expression of adipocyte identity, fibrosis markers and Hippo signal proteins of cells in (c). e, Knock-down efficiency of L1L2-gRNA1 (left) and L1L2-gRNA2 (right) in differentiated adipocytes transduced with the indicated gRNAs (n=3 biologically independent cell cultures). f, mRNA expression of Lats1 and Lats2 in adipocyte and SVF from eight-week-old male L1L2-FF and L1L2-iAKO mice that were i.p. administered with a dose of tamoxifen (100 mg/kg) and then analyzed 6 days later (n=6 mice). g-i, Body weight (g), mRNA expression of adipocyte identity (h) and fibrosis markers (i) from eight-week-old male L1L2-FF and L1L2-iAKO mice that were i.p. administered with 3 doses of tamoxifen (100 mg/kg) every other day and then analyzed 4 weeks later (n=5 mice). j, The structure of the AAV-Adiponectin promoter (ADP) vector. k, Immunoblot analysis of AAV-ADP-GFP-Flag expression at the indicated tissues of WT mice whose scWAT were locally injected with the viruses. Independent experiments were performed twice with similar results. l, Comparison of FLAG expression levels driven by ADP (1.6 k) or ADP (1.1 k) promoter. Independent experiments were performed twice with similar results. m-q, Eight-week-old male mice injected with AAV-ADP-GFP, AAV-ADP-TAZ (4SA) or AAV-ADP-YAP (5SA) in
scWAT for 4 weeks. m, Immunoblot analysis of protein expression of TAZ(4SA) or YAP(5SA) in scWAT. n, Representative images of scWAT injected with AAV-ADP-GFP, AAV-ADP-TAZ (4SA) or AAV-ADP-YAP (5SA). o, Quantification of scWAT weight (n=7 mice). p, Representative scWAT sections with H&E staining. Independent experiments were performed twice with similar results. q, RT-qPCR analysis of fibrosis and adipocyte markers (n=6 mice). Data are means ± SEM. Two-tailed unpaired student’s t-test in (e), (g-i), (o), (q); One-way ANOVA with Bonferroni’s multiple-comparisons test in (c); Two-way ANOVA with Bonferroni’s multiple-comparisons test in (f); *P < 0.05, **P < 0.01, ***P < 0.001; NS, not significant. Exact P values are provided in a Source Data file.
Supplementary Figure 6

a) DAPI, p-SMAD2, Merge

b) p-SMAD2, SMAD2, HSP90

c) Tgfr1

d) Tgfr2

e) CD9

f) L1L2-FF, L1L2-AKO

g) L1L2-FF, L1L2-AKO

h) L1L2-FF, L1L2-AKO

i) L1L2-FF, L1L2-AKO

j) Vec, L1L2-gRNA1

k) L1L2-FF, L1L2-AKO

l) L1L2-FF, L1L2-AKO

m) L1L2-FF, L1L2-AKO

n) L1L2-FF, L1L2-AKO

o) NS

p) L1L2-FF, L1L2-AKO

q) L1L2-FF, L1L2-AKO

Scale bar, 50 μm

Body weight (g)

Analysis
Supplementary Figure 6. TGFβ signaling may contribute to Lats1/2-deficiency induced fibrosis.

a, Representative sections of scWAT stained for p-SMAD2 (red) and nucleus (DAPI, blue) during growth. Independent experiments were performed twice with similar results. b, Representative immunoblot analysis of phosphorylation levels of SMAD2 in scWAT during growth. Independent experiments were performed twice with similar results. c-e, mRNA expression of scWAT Tgfbr1 (c), Tgfbr2 (d) or CD9 (e) during growth (n=6 mice). f, g, mRNA expression of scWAT CD9 in 5-week-old L1L2-AKO (f, n=6 mice) or in L1L2-iAKO mice as indicated in supplementary Figure 5g (g, n=5 mice). h, Immunoblot analysis (left) and quantification (right) of the p-SMAD2 in scWAT of male L1L2-FF or L1L2-AKO mice at five weeks old (n=3 mice). i, mRNA expression of scWAT Tgfb1/2/3, Tgfbr1 and Tgfbr2 of L1L2-FF or L1L2-iAKO mice as indicated in supplementary Figure 5g (n=5). j, mRNA expression of Tgfb1/2/3 of differentiated adipocytes transduced with Vec or L1L2-gRNA1 AAV (n=3 biologically independent cell cultures). k, Immunoblot analysis (left) and quantification (right) of the non-canonical TGFβ pathways in scWAT of male L1L2-FF or L1L2-AKO mice at five weeks old (n=3 mice). l, Immunoblot analysis (left) and quantification (right) of phosphorylation levels of JNK in scWAT of male L1L2-FF or L1L2-iAKO mice four weeks after tamoxifen administration (n=3 mice). m-q, Three consecutive intragastric injections of tamoxifen (1.5 mg/mL) were given to newborn male L1L2-FF or L1L2-iAKO mice at P2–P3–P4 and all mice were analyzed at 5 weeks old. n, Gross pictures of mice. o, p, Body weight (o) or mRNA expression of fibrosis markers (p) of L1L2-FF (n=5) and L1L2-iAKO (n=4) mice. q, Representative Masson’s trichrome staining of scWAT sections of L1L2-FF and L1L2-iAKO mice. Data are means ± SEM. Two-
tailed unpaired student’s t-test in (f), (g), (h-l), (o); *$P < 0.05$, **$P < 0.01$, ***$P < 0.001$;

NS, not significant. Exact $P$ values are provided in a Source Data file.
Supplementary Figure 7

a) CAG-GFP vs CAG-TGFβ1 (T204D)

b) CAG-GFP vs CAG-Tgfb1 (2CS)

c) SMAD2 and p-SMAD2 expression

d) DIO-EF1A-GFP vs DIO-EF1A-TGFβ1 (T204D)

e) ADP-GFP vs ADP-TGFβ1 (T204D)

f) Tissue Weight (g)

h) Relative expression of various genes

i) Tissue Weight (g)

j) ADP-GFP vs ADP-TGFβ1 (T204D)

k) Relative expression of Yap1 and Taz
Supplementary Figure 7. Hippo pathway inactivation or TGFβ stimulation alone is not sufficient to induce AT fibrosis in adult mice.

a, Overexpression efficiency of AAV-CAG-TGFβR1 (T204D) in L1L2-FF or L1L2-iAKO scWAT transduced with AAV-CAG-GFP (n=5 mice) or AAV-CAG-TGFβR1 (T204D) (n=6 mice) followed by tamoxifen administration. B, Representative sections of indicated scWAT stained for p-SMAD2 (red) and nucleus (DAPI, blue). c, d, f, g, Left, immunoblot analysis of phosphorylation of SMAD2 (p-SMAD2) of the indicated mice; Right, quantification of relative phosphorylation levels of SMAD2 (n=3 mice). e, Overexpression efficiency of AAV-CAG-TGFβ1 (2CS) in scWAT (n=5 mice). h-j, Eight-week-old WT mice were subcutaneously injected with AAV-ADP-GFP or AAV-ADP-TGFβR1 (T204D). h, mRNA expression of fibrosis and adipocyte markers (n=6 mice). I, scWAT weight (n=6 mice). j, Representative scWAT sections with H&E staining. k, Knockdown efficiency of YT-gRNA in Cas9+/ SVF (n=4 biologically independent cell cultures). Data are means ± SEM. Two-tailed unpaired student’s t-test; *P < 0.05, **P < 0.01, ***P < 0.001; NS, not significant. Exact P values are provided in a Source Data file.
Supplementary Figure 8. *Lats1/2* deletion induces inflammatory responses in scWAT.

**a**, Gating strategy of flow cytometry for macrophages. **b**, L1L2-FF or L1L2-iAKO mice that were locally injected with AAV-ADP-FLPo and AAV-EF1A-FIO-TGFβR1 (T204D). mRNA expression of inflammatory genes in scWAT (n=6 mice). **c**, L1L2-FF mice that were locally injected with AAV-ADP-GFP or AAV-ADP-TGFβR1 (T204D). mRNA expression of inflammatory genes in scWAT (n=6 mice). **d**, mRNA expression of inflammatory markers in L1L2-FF or L1L2-iAKO mice (n=5). **e**, mRNA expression of *Ccl2* in adipocytes of pooled P7 male L1L2-FF or L1L2-AKO scWAT (n=3). One dot represents a mean value of biologically independent samples from four male mice. **f**, Overexpression efficiency of AAV-CAG-GFP (n=7 mice), AAV-CAG-CCL2 and AAV-CAG-CCL7 (n=5 mice) in scWAT. **g**, h, F4/80⁻ and F4/80⁺ cells were magnetically sorted from pooled L1L2-FF or L1L2-AKO scWAT SVF and analyzed for mRNA expression of (g) *Ccl2* or (h) *Ccl7* (n=3). One dot represents a mean value of biologically independent samples from three male mice. Data are means ± SEM. Two-tailed unpaired student’s *t*-test; *P* < 0.05, **P* < 0.01, ***P* < 0.001. Exact *P* values are provided in a Source Data file.
Supplementary Figure 9. *Lats1/2* deletion promotes the cell fate conversion from adipocyte to myofibroblast.

**a,** Representative sections of scWAT of P28 L1L2-AKO^{mTmG} mice stained for αSMA (AF647, red), nucleus (DAPI, blue) and membrane GFP (green). Arrowheads show examples of GFP^αSMA^+ cells. Independent experiments were performed three times with similar results. **B,** Representative sections of scWAT of P28 L1L2-AKO^{mTmG} mice stained for Ki-67 (eFluor 660, red), nucleus (DAPI, blue). Arrowheads show examples of GFP^Ki-67^+ cells. Independent experiments were performed twice with similar results. **C,** Representative sections of scWAT of L1L2-iAKO^{mTmG} mice stained for αSMA (AF647, red), nucleus (DAPI, blue) and membrane GFP (green). Independent experiments were performed twice with similar results. **D,** Representative sections of scWAT of L1L2-iAKO^{mTmG} transduced with AAV-CAG-TGFβR1 (T204D) and stained for αSMA (AF647, red), nucleus (DAPI, blue) and membrane GFP (green). Arrowheads indicate GFP^αSMA^+ cells. Independent experiments were performed twice with similar results. **e,** Immunoblot analysis of αSMA protein expression in sorted DPP4^+ or DPP4^- cells from WT scWAT SVF. Independent experiments were performed twice with similar results. **f,** Immunoblot analysis of αSMA protein expression in sorted DPP4^ICAM1^- or DPP4^ICAM1^+ cells from WT scWAT SVF. Independent experiments were performed twice with similar results. **g,** mRNA expression of adipocyte markers of P14 L1L2-FF or L1L2-AKO scWAT (n=6 mice). **h,** GFP^DPP4^+ or GFP^DPP4^- cells sorted from L1L2-AKO^{mTmG} scWAT SVF were plated and stained for Ki-67 (eFluor 660, purple) and nucleus (DAPI, blue). Independent experiments were performed twice with similar results. **i,** Quantification of Ki-67^+ cell percentages of GFP^DPP4^+ (n=10 visual fields) or GFP^DPP4^- (n=12 visual fields) cells. **j,** RT-qPCR analysis of *Dpp4* mRNA expression in differentiated
adipocytes (from Cas9Tg/+ scWAT SVF) transduced with Vec or L1L2-gRNA (n=3 biologically independent cell cultures). k, Sorted GFP+DPP4− cells from P21 L1L2-AKO<sup>LSL-CAS9-EGFP</sup> scWAT SVF were treated with TGF-β1 (10 ng/ml) for 0 h or 24 h. Cells were stained with αSMA (red), DPP4 (purple), nucleus (DAPI, blue) and GFP (green). Independent experiments were performed twice with similar results. l, The other two recipient mice as in Fig. 5g. Donor cells were from scWAT SVF of L1L2-AKO<sup>mTmG</sup> (upper) or L1L2-AKO<sup>LSL-CAS9-EGFP</sup> (lower) mice. Data are means ± SEM. Two-tailed unpaired student’s t-test; *P < 0.05, **P < 0.01, ***P < 0.001. Exact P values are provided in a Source Data file.
Supplementary Figure 10

(a) Images of mouse tissues stained with specific markers.

(b) Western blot analysis showing relative expression levels of proteins including HSL, PLIN1, αSMA, TAZ, YAP, and HSP90.

(c) Diagram illustrating the Induction cocktail leading to Adipogenesis and Fibrosis pathways.

(d) Tables showing KO efficiency and Relative expression for different genotypes (L1L2-FF, L1L2-AKO, L1L2YT-FF, L1L2YT-AKO).

(e) Immunofluorescence images with scale bars indicating 200 μm and 50 μm.

(f) Graphs showing average intensity for different genotypes.

(g) Graphs illustrating Relative protein level and Relative expression for different proteins.

(h) Graphs showing KO efficiency and Relative expression for different genotypes.

(i) Graphs illustrating Relative expression for different proteins.

(j) Graphs showing Relative protein level and Relative expression for different genotypes.

(k) Graphs showing Relative protein level and Relative expression for different genotypes.

(l) Graphs showing Relative protein level and Relative expression for different genotypes.

(m) Graphs showing Relative protein level and Relative expression for different genotypes.

(n) Graphs showing Relative protein level and Relative expression for different genotypes.

(o) Graphs showing Relative protein level and Relative expression for different genotypes.

(p) Graphs showing Relative protein level and Relative expression for different genotypes.

(q) Graphs showing Relative protein level and Relative expression for different genotypes.

(r) Graphs showing Relative protein level and Relative expression for different genotypes.
Supplementary Figure 10. The LATS1/2-YAP/TAZ-TEADs axis regulates adipocyte identity.

a, Representative images of scWAT of the indicated mouse strains. b, Immunoblot analysis of HSL, Perilipin 1 and αSMA protein expression in scWAT of the indicated mouse strains (n=3 mice). c-g, Primary Cas9<sup>Tg<sup>+<sub>/+</sub></sup> SVF treated with TGFβ1 or adipogenic cocktail. c, Experimental scheme. d, Immunoblot analysis of adipocyte markers in differentiated Cas9<sup>Tg<sup>+/+</sub></sup> SVF. e, Oil red staining of differentiated Cas9<sup>Tg<sup>+/+</sub></sup> SVF transduced with Vec, L1L2-gRNA or L1L2YT-gRNA. Independent experiments were performed twice with similar results. f, Immunostaining of αSMA<sup>+</sub> primary SVF cells treated with TGFβ1 (10 ng/mL) for 24 h. Independent experiments were performed twice with similar results. g, Quantification of average intensity of αSMA in (f) (n=6 visual fields). h, i, RT-qPCR analysis of the knockdown efficiency of (h) Lats1/2 and Tead1/2/3/4 and (i) the Hippo pathway downstream gene expression in differentiated adipocytes (n=3 biologically independent cell cultures). j, Immunoblot analysis of the specificity of SMAD2 antibody. Independent experiments were performed twice with similar results. k, Immunoblot analysis of SMAD2 protein expression in differentiated adipocytes from SVF of L1L2-FF or L1L2-AKO scWAT. Independent experiments were performed twice with similar results. l, mRNA expression of Smad2 in differentiated adipocytes from SVF of L1L2-FF or L1L2-AKO scWAT (n=3 biologically independent cell cultures). m-p, Primary Cas9<sup>Tg<sup>+/+</sub></sup> SVF transduced with Vec or L1L2-gRNA for 4 days. m, Experimental timeline. n, SMAD2 protein expression. o, Quantification of SMAD2 protein expression (n=3 biologically independent cell cultures). p, mRNA expression of Smad2 (n=3 biologically independent cell cultures). q, r, Primary Cas9<sup>Tg<sup>+/+</sub></sup> SVF transduced with L1L2-gRNA on day 0, followed by YT-gRNA on day 1. q, Experimental timeline. r, Immunoblot
analysis of SMAD2 protein expression. Independent experiments were performed twice with similar results. Differentiated adipocytes were transduced with Vec, L1L2-gRNA1, MYC-Ub and SMAD2-HA. Immunoprecipitation (IP) followed by western blot assay was performed. Independent experiments were performed twice with similar results. Data are means ± SEM. One-way ANOVA with Bonferroni’s multiple-comparisons test in (g), (h) and (i); Two-tailed unpaired student’s t-test in (l), (o) and (p); *P < 0.05, **P < 0.01, ***P < 0.001; NS, not significant. Exact P values are provided in a Source Data file.
Supplementary Figure 11

a) Relative expression of Yap1 and Wwtr1 in scWAT (YT-FF) and vWAT (YT-iAKO) with high fat diet (HFD).

b) Body weight comparison between Veh, VP, and HFD over 8 weeks after tamoxifen injection.

c) Relative expression of Coll1a1, Coll6a1, Mmp2, Mmp14, Fn1, Lox, A20, and Yap1 in vWAT (YT-FF) and vWAT (YT-iAKO) with HFD.

d) Picrosirius red staining of vWAT (YT-FF) and vWAT (YT-iAKO) with HFD.

e) Picrosirius red staining of vWAT (Veh) and vWAT (VP) with HFD.

f) DAPI, Colla1, and Merge staining of scWAT and vWAT with HFD.

g) Relative expression of Wwtr1, Colla1, Tim1, Mmp1, Fn1, Lox, and Yap1 in vWAT (Veh) and vWAT (VP) with HFD.

h) Blood glucose levels in Veh and VP with HFD.

i) Blood glucose levels in Veh and VP with ITT.

j) High fat diet (HFD) on Body weight (g) over 20 weeks.

k) Body weight (g) comparison between WT mice and ob/ob with HFD.

l) Body weight (g) comparison between AAV injection and ITT.

m) Relative expression of ADP-GFP, ADP-TAZ (4SA), and HSP90 in vWAT (YT-FF) and vWAT (YT-iAKO) with HFD.

n) Blood glucose levels in ADP-GFP and ADP-TAZ (4SA) with ITT.

o) DAPI, Colla1, and Merge staining of scWAT and vWAT with HFD.

p) Blood glucose levels in GFP and TAZ (4SA) with ITT.

q) Blood glucose levels in ADP-GFP and ADP-TAZ (4SA) with ITT.
Supplementary Figure 11. Targeting the Hippo pathway in precursor cells and adipocytes to treat obesity-induced AT fibrosis and metabolic dysfunction.

**a-d**, YT-FF and YT-iAKO mice were fed a HFD for 19 weeks. **a**, Knockout efficiency of *Yap1* or *Wwtr1* in scWAT or vVAT (n=6 mice). **b**, Body weight of YT-FF and YT-iAKO mice under a high-fat diet before and after 5 doses of tamoxifen (Tam) (50 mg/kg) treatment (n=6 mice). **c**, mRNA expression of fibrosis markers in vWAT (n=6 mice). **d**, Representative vWAT sections with Picrosirius red staining. **e**, Representative vWAT sections with Picrosirius red staining of 12-week-old *ob/ob* mice administrated with Veh or VP. Independent experiments were performed three times with similar results. **f**, Representative sections of vVAT stained for Collα1 (red) and nucleus (DAPI, blue) of *ob/ob* mice that were i.p. injected with Veh or VP. Independent experiments were performed twice with similar results. **g-j**, Mice fed a HFD for 18 weeks were i.p. injected with Veh or VP (25 mg/kg) for 5 doses every other day. **g**, Experimental scheme. **h**, GTT (n=6 mice). **i**, ITT (n=6 mice). **j**, Representative scWAT sections with Picrosirius red staining in scWAT and vVAT. Independent experiments were performed three times with similar results. **k-q**, WT mice were fed a HFD for 10 weeks followed by an injection with AAV-ADP-GFP, AAV-ADP-TAZ (4SA) in scWAT for 4 weeks. **k**, Experimental scheme. **l**, Immunoblot analysis of protein expression of TAZ(4SA) in scWAT. **m**, Body weight (n=6 mice). **n**, mRNA expression of fibrosis markers (n=6). **o**, Representative scWAT sections with IF staining of Collα1. **p**, GTT (n=6 mice). **q**, ITT (n=6 mice). Data are means ± SEM. Two-tailed unpaired student’s *t*-test in (a), (b), (c), (m), (n); Two-way ANOVA with Bonferroni’s multiple-comparisons test in (h), (i), (p), (q); *P < 0.05, **P < 0.01, ***P < 0.001. Exact *P* values are provided in a Source Data file.
Supplementary Table 1

Sequences of qRT-PCR primers used in this study.

| Gene     | Forward primer       | Reverse primer       | Note                                      |
|----------|----------------------|----------------------|-------------------------------------------|
| Fn1      | GATGTCCGACGACACTATTTCACCA | CCTTGCCGACTTCAGCCACT |                                           |
| Mmp2     | GGACAGTGGTCCGCCAGAA | CCGACGGTGAGCAAGGAAGG |                                           |
| Mmp14    | CAGTATGCTACCTACTCCCAAG | GCCCTGCTGCTACTTGTAAA |                                           |
| Lox      | CAGCCACATAGTGCGATGGT | GCCGTATCCAGGTCCGTC    |                                           |
| Acta2    | GTGCCAGACACATGGAACGTA | TCGGATACCTTACGTCAGGA |                                           |
| Tagln    | CAAAAGGGTCCATCTACCGG | ATCTGGGGCGGCCATACCA  |                                           |
| Col1a1   | TAAAGGTCCTCAGGATGAAA | GGTCCTGCACTCCTACAT   |                                           |
| Col3a1   | CGTAAAGCATGGAACGACG | CCGATGACGGGAGCAGAC   |                                           |
| Col6a1   | CAGTATGCTACCTACTCCCAAG | GCCCTGCTGCTACTTGTAAA |                                           |
| Tgfb1    | ATCTGCGCTGGTACCTTGGG | AGCCCTGATCCGTCCTCCT  |                                           |
| Tgfb2    | TCGGACATGTTATGGGACGTA | CCCGTATGCGAGTCTAGG    |                                           |
| Tgfb3    | CCTGGCCCTTGCAGATTCTG | TTGATGGCGGACAGTCCACAC |                                           |
| Tgfr1    | CCAGTCCTGCTGCTTTCGCT | GCCATACAAATGGCCTGTCCT |                                           |
| Tgfr2    | GATGTCAAGGCGCAGCCG | TGTTCTTGCAGTCTTTCTC  |                                           |
| Smad2    | ATGCTGCTTACCTTGGGCATC | AACCGTCTGTTATTTAGG    |                                           |
| Timp1    | CCAGACACACACTTGACACGC | ATGACTGCGGTGGATGCTAGG |                                           |
| Ccl2     | TTAAATGTCAGTGCGGCCAA | GCATTCTCAGATTACGCGGA |                                           |
| Ccl7     | CCAAAAGCTTGGTCCGGAACCAA | GCATTGCTCAGATTACGCGGA |                                           |
| Mcc1     | CTCTGTTACGTATTTGGACGC | CGGAATTTCGAGGTAGAGAGC |                                           |
| Mgl1     | TGAGAAAGGCTTTAAGAAGTGGG | GACCACCTGATGATGATG    |                                           |
| Nos2     | GAGCAACTATCGCTGGTGTTG | TCAGAGTCTGCCCATTGCG  |                                           |
| F4/80    | TGACTCTACCTTGGTTCCTTA | CTGCCCAATACGCTTCATTT  |                                           |
| CD11b    | CCTTCATCAACACACCAGATGCG | CGAGGTGGCTCTATCAAAAACCAAGC |                                           |
| Pparg    | CAAGAAATACAAAGTGTCGATCAA | GAGCTGGGCTTTTCCAGAATAT |                                           |
| Fabp4    | ACACCGAGATTCTCCTAAACTG | CCACTCTGATGATGCTTTCC |                                           |
| Adipsoq  | GCACCTGCAAGTTTACTGCAA | GTAGGTGAAGAAGGGCCTTGT |                                           |
| Glut4    | GTGACTGGAGCCCACTGCTTCA | CCAAGCCAGTGGCATTAGG  |                                           |
| Hsl      | CCAGCCCTAGAGGGCTTACTG | CTCCCTGACGACTATCG    |                                           |
| Lats1    | ATCTCGCGGAATCCTCCTGTG | GTGCGCTCCGAGGTAAGTTAAAA |                                           |
| Lats1    | GAAAGACGGTTTCTGCTCCGAA | ATCGTGCCAGATTTCAGGA  | for detection of KO efficiency of L1-gRNA1 |
| Lats1    | GGAGTTCCAGAATGTTGCTGGG | GGAACGTTTCCATTGCGGAA | for detection of KO efficiency of L1-gRNA2 |
| Lats2    | GGCTTCTCCACCGGGACAT | AATCCAGTGCAAGGCGAAAA |                                           |
| Lats2    | TGGAGCAGGGAAAATGGCCAAA | CGTGAGTGTCAGCTTACAA  | for detection of KO efficiency of L2-gRNA1 |
| Lats2    | GCAGACGGCCAGTGGAGGTA | CTCGCTAGTTTGCAACCACCC | for detection of KO efficiency of L2-gRNA2 |
| Gene   | Sequence 1 | Sequence 2 |
|--------|------------|------------|
| Yap1   | TGAGATCCCTGATGATGTACCAC | TGTTGTGTCTGATCGTGTGAT |
| Wwtr1  | CCGTTCCGGGGATAAAAGAT  | GTTGAGAGGGCCTCGAGGTT |
| 36B4   | TCACTGTGCCAGCTCAGAAC | ATCAGCTGCACATCCACTCAGA |
| Tead1  | AAGCGATTTGACACCAACGACGC | CCTGTCTTTCCCGTCCTGA |
| Tead2  | ATGGGGGATCCCCGGACTGG  | ATGATCTTGGACGCGCCACA |
| Tead3  | AAGGGTCTGGACAACCGATGC | CTGTTTTCTTGTCTGGTTT |
| Tead4  | TGGAGCTCTCTCCCGACTCCCC | TGCAGTCAGCTCATCCGCAC |
| Ctgf   | GGACACCTAAAATCGCCAAAGC | ACTTAGCCCTGTATGCTCCACA |
| Cyr61  | TAAGTGCTGCCAAACAAACTC | CAGATCCCTTTCCAGACCGGT |
| Ankrd1 | TGCAGATGATATAAACGGAGC | GTGGATCAAGCATATCTCGGA |

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## Supplementary Table 2

**Sequences of gRNAs used in this study.**

| gRNA     | Sequence                        | Note |
|----------|---------------------------------|------|
| Lats1-gRNA1 | AGACGTCTCTGCTCCGAAATC           |      |
| Lats1-gRNA2 | ACGTTTCCATGGCGAATGA             |      |
| Lats2-gRNA1 | GAGTGTCACGGTTAAAGCG             | 68   |
| Lats2-gRNA2 | GCTGGGTGGTGCAAAACTACG           |      |
| Yap-gRNA  | GATCAGACAACACATGGC              |      |
| Taz-gRNA  | TCACGTCATAGGACTGGG              |      |
| Tead1-gRNA | CCGATTGACAACGACGCGGA            |      |
| Tead2-gRNA | ATCTTGCGACGGCCACAGGG            |      |
| Tead3-gRNA | GTCTGGACAGTGAGGGG               |      |
| Tead4-gRNA | AGCTCTCCCGACTCCCCGA             |      |
| Smad2-gRNA | CTTGCCATTCCTCCCGCCAG           |      |