

Supplementary data

sFigure 1 | WAT basement membranes and pancreatic tissue immunofluorescence stainings.

(A) Representative immunofluorescence stainings for laminin $\beta 1$, $\beta 2$ and $\gamma 1$ chains in eWAT of laminin deficient mice and WT controls. (B) Double staining for pan-laminin and insulin visualises pancreatic islets and permits (C) their quantification. Data in C is from at least 3 mice; statistical significance was determined using a two-sided unpaired t-test.

sFigure 2 | Characterisation of interstitial progenitors and committed preadipocytes.

(A) Violin plots showing marker gene expression in committed preadipocytes and in interstitial progenitors. (B) Flow cytometry gating strategy for quantification of preadipocyte populations, and histogram showing high ICAM-1 expression specifically on committed preadipocytes; corresponding proportions of preadipocyte populations in (C) *Lama4*^{-/-} and (D) *Tek-cre:Lama5*^{-/-} eWAT. (E) Representative immunofluorescence staining for Ki67 to mark proliferating committed preadipocytes; boxed areas are shown at higher magnifications to the right. (F) Flow cytometry quantification of in vivo EdU incorporation into DPP4⁺ interstitial progenitors and ICAM-1⁺ committed preadipocytes. (G) In vitro doubling time of PDGFR α ⁺ committed preadipocytes from *Lama4*^{-/-} and WT littermate eWAT. (H) Gene set enrichment analysis (GSEA) of Gene Ontology (GO) including Biological Process (BP), Cellular Component (CC), and Molecular Function (MF) gene sets in interstitial progenitors and committed preadipocytes from WT and *Lama4*^{-/-} eWAT. Normalized enrichment scores (NES) are shown for significantly enriched GO terms. Dot size represents gene set size and colour indicates adjusted P value. Data in C, D, F and G are means $\pm$ SD from at least 3 experiments with at least 3/mice/genotype; statistical significance was determined using a two-sided unpaired t-test.

sFigure 3 | DPP4⁺ interstitial progenitors reside in the reticular septum.

Representative immunofluorescence staining of the (A, C) reticular septum and the (B) body of inguinal WAT (iWAT) of laminin deficient and *Cd18^{-/-}* mice. Arrow in C marks smooth muscle laminin $\alpha 5^+$ arteriole.

sFigure 4 | Immune cell characterization in laminin deficient mice.

Flow cytometry quantification total numbers and proportions of different immune cell populations in (A) *Lama4^{-/-}* and (B) *Tek-cre:Lama5^{-/-}* eWAT; cell numbers are normalized to adipose tissue weight. Data are means $\pm$ SD from at least 2-5 experiments with at least 3 mice/experiment; statistical significance was determined using a two-sided unpaired t-test. (C) Representative immunofluorescence showing distinction between CD209b⁺ and CD206⁺Lyve1⁺ macrophage populations (upper panels), and the absence of CD209b⁺ macrophages in contact with PDGFR α^+ committed preadipocytes (lower panels).

sFigure 5 | Quantification of extravasated monocytes in laminin deficient mice.

(A,D) Flow cytometry for Ly6C⁺ monocytes and (B, E) corresponding quantification expressed as total numbers and as a proportion of total CD45⁺ cells in (A,B) *Lama4^{-/-}* and (D,E) *Tek-cre:Lama5^{-/-}* eWAT. (C) Flow cytometry quantification of in vivo EdU incorporation into CD206⁺ macrophages. Volcano plots of differentially expressed genes in (F) Tim4^{neg} macrophages in *Lama4^{-/-}* versus WT eWAT (ND), and (G) Tim4⁺ macrophages in WT mice on ND and HFD. (H) Gene Ontology (GO) enrichment analysis of Biological Process (BP), Cellular Component (CC), and Molecular Function (MF) terms in Tim4⁺, MHC-II⁺ and Trem2⁺ macrophages from WT and *Lama4^{-/-}* eWAT. Normalized enrichment scores (NES) are shown for significantly enriched GO terms. Dot size represents gene set size and colour indicates adjusted *P* value . Data in B, C, E are means $\pm$ SD from at least 3-8 mice/ genotype in at least 3 experiments; statistical significance was determined using a two-sided unpaired t-test.

sFigure 6 | Characterisation of immune cell populations in *Cd18*^{-/-} eWAT.

(A) Flow cytometry of gating strategy for identification of myeloid populations and (B) corresponding quantification total CD11b⁺ cells, confirming the deletion of Mac-1 (CD18). (C) Quantification of eWAT weights, (D) Flow cytometry quantification of in vivo EdU incorporation into DPP4⁺ interstitial progenitors and ICAM-1⁺ committed preadipocytes. (E) Comparisons of in vitro doubling times of WT, *Cd18*^{-/-} and *Lama4*^{-/-}PDGFR α ⁺ committed preadipocytes. Data in B-E are means $\pm$ SD from at least 3 experiments with a minimum of 2 mice/genotype/experiment. statistical significance was determined using a two-sided unpaired t-test.

sFigure 7 | Adipocyte organoid cultures.

(A) Immunofluorescence staining for Tim4⁺ macrophages in in vitro adhesion assays employing different ratios of laminin 511:laminin 411. (B) Representative fluorescence imaging of WT and *Tek-cre:Lama5*^{-/-} spheroids for Bodipy and perilipin to mark lipid droplets, Pan-laminin (PNLM) to mark all basement membranes and DAPI to mark nuclei. (C) qPCR analysis of adipogenic gene *Pparg* expression following differentiation of 3D spheroid cultures. Data are means $\pm$ SD of at least 3 separate experiments with triplets in each; statistical significance was determined using a two-way ANOVA.

sFigure 8 | Proposed model of the vascular basement membrane–dependent macrophage–preadipocyte niche in white adipose tissue.

Laminin α 4-rich vascular basement membranes provide a specialized microenvironment that promotes interactions between CD206⁺Tim4⁺ resident macrophages and ICAM1⁺ committed preadipocytes. Cell–cell contact through the MAC-1 (CD11b/CD18)–ICAM-1 adhesion axis activates intracellular signaling pathways, including Src, ROCK, and p38 MAPK, in committed preadipocytes, leading to the induction of pro-adipogenic genes (including *Plin1*, *Adipoq*, and *Pparg*) and promoting adipocyte differentiation. Illustration created using Adobe Illustrator.