

Appendix

Mammalian adipogenesis regulator (Areg) cells use retinoic acid signaling to be non- and anti-adipogenic in age-dependent manner

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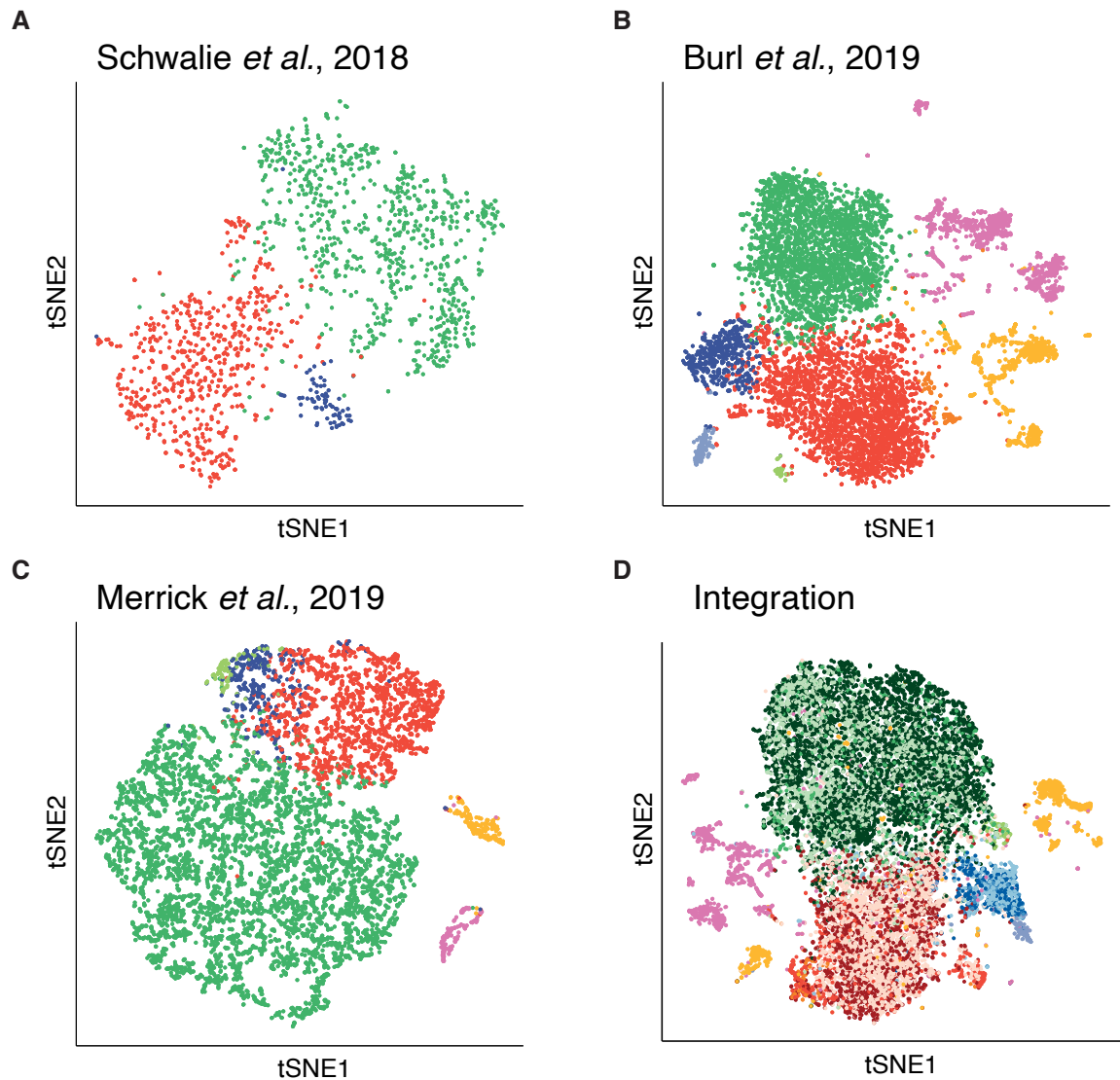
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Schwalie et al. iWAT

- P1 (ASCs)
- P2 (preAs)
- P3 (*F3+* ASPCs)

Merrick et al. iWAT

- Group 1 (ASCs)
- Group 2 (preAs)
- Group 3 (*F3+* ASPCs)
- *Serpina1b*⁺
- *Cited1*⁺
- *Cilp*⁺

Burl et al. iWAT

- ASC2 (ASCs)
- ASC1 (preAs)
- ASC1 (*F3+* ASPCs)
- *Cilp*⁺

Non ASPCs

- Immune cells
- Endothelial cells

ASCs

- preAs
- *F3+* ASPCs
- *Cilp*⁺
- *Serpina1b*⁺
- *Cited1*⁺

Immune cells

- Immune cells
- Endothelial cells

Appendix Figure S1. The *F3+* cluster exists in distinct, publicly available single-cell RNA-seq datasets

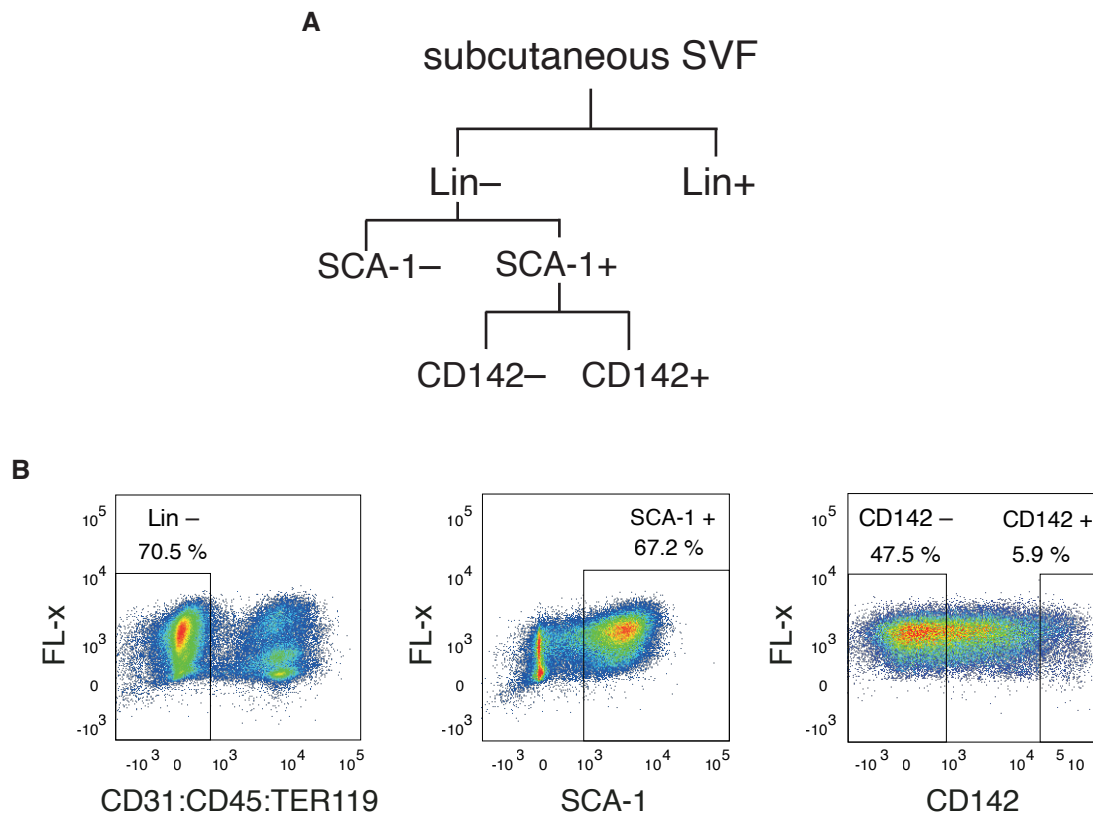
(A) t-SNE cell map of the scRNA-seq dataset published in Schwalie *et al.*, 2018;

(B) t-SNE cell map of the scRNA-seq dataset published in Burl *et al.*, 2019;

(C) t-SNE cell map of the scRNA-seq dataset published in Merrick *et al.*, 2019;

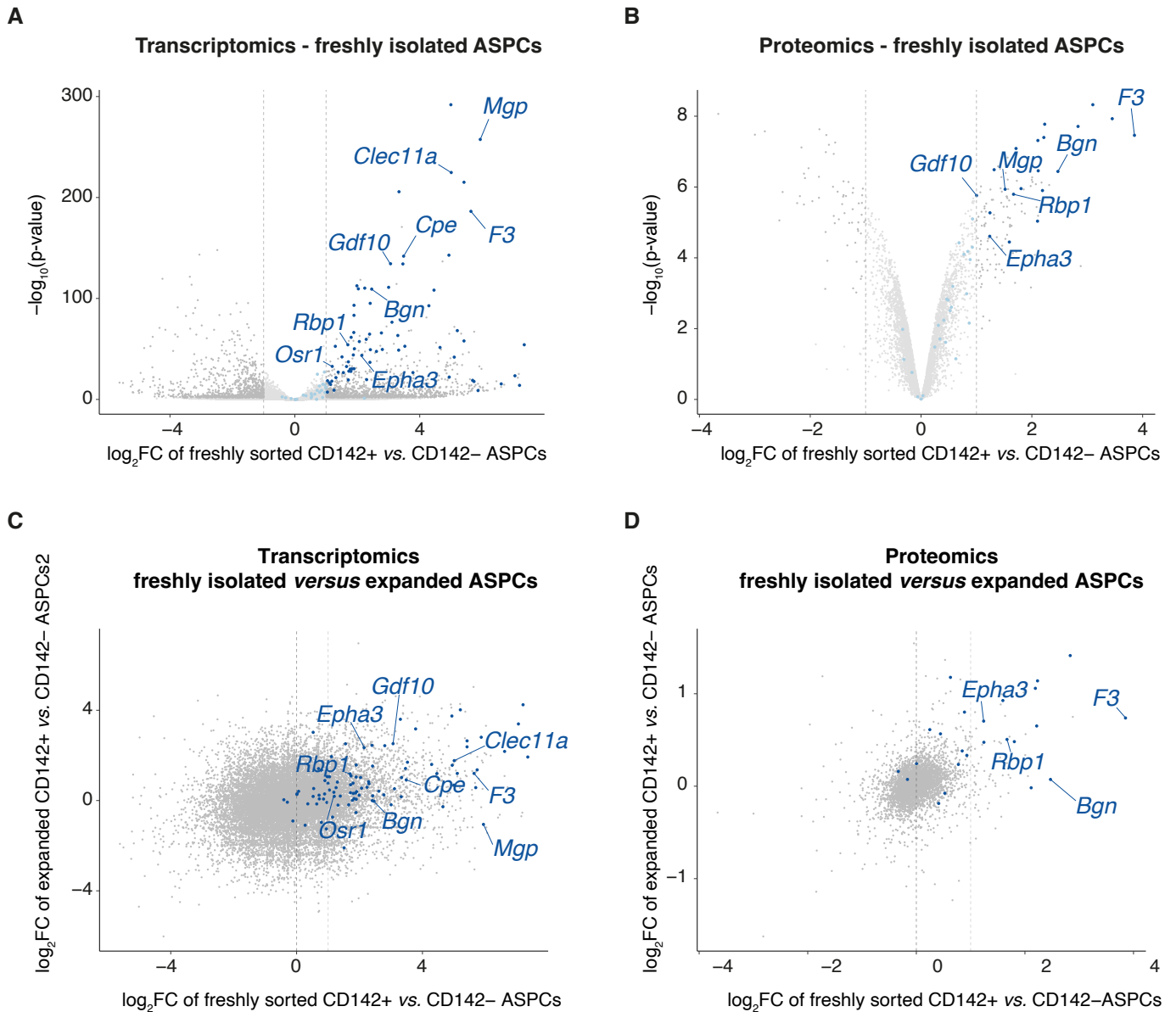
(D) t-SNE cell map of the integration analysis of the three datasets presented in A-C; the t-SNE is coloured by the clustering results identified in each dataset individually;

Three main ASPC subpopulations colour code: adipose stem cells (ASCs) — green, pre-adipocytes (PreAs) — red, *F3*(CD142)+ ASPCs — blue.



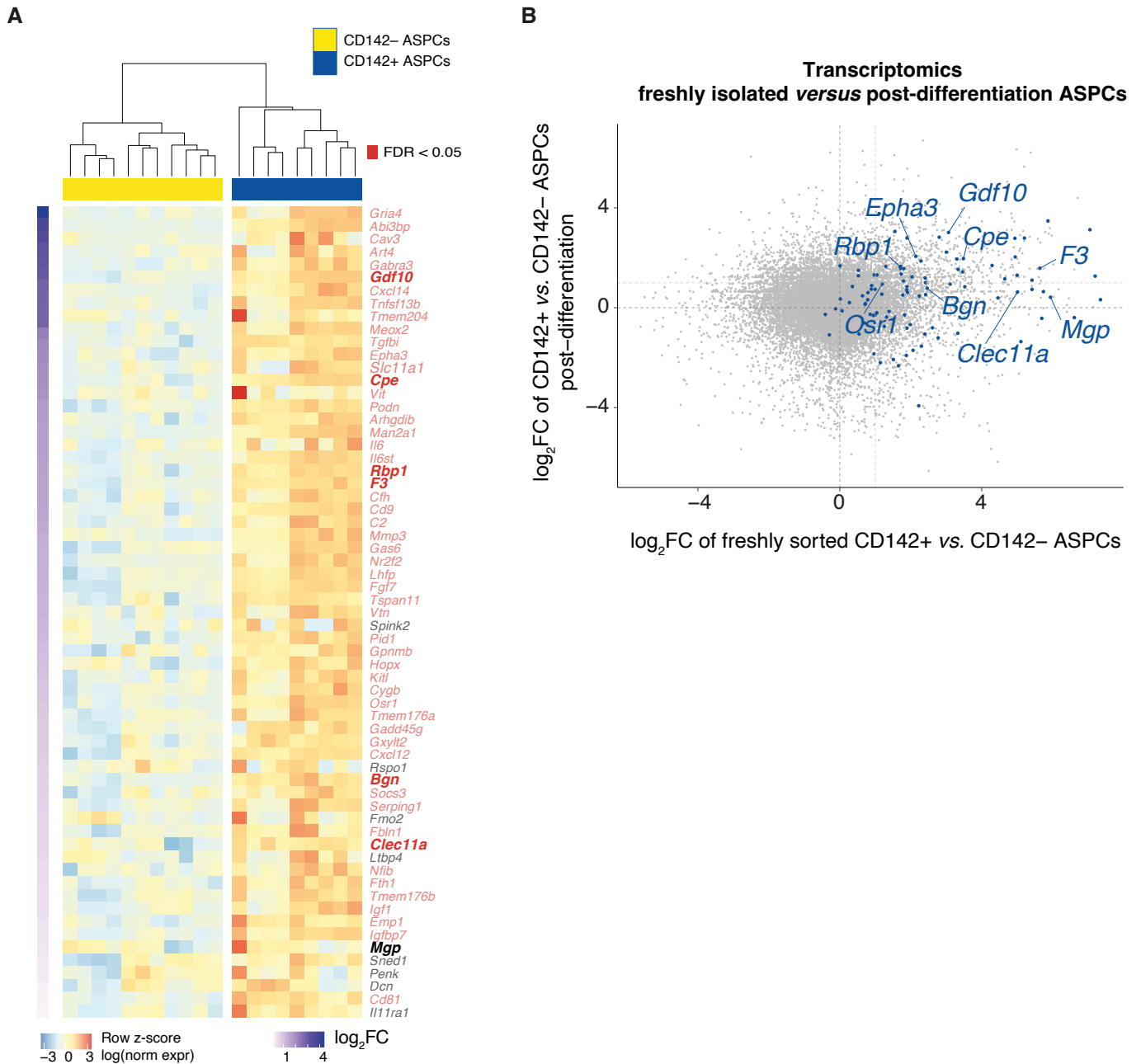
Appendix Figure S2. FACS-based gating strategy to isolate different ASPC fractions

- (A)** Schematic of the FACS-based selection strategy of mouse subcutaneous ASPCs;
- (B)** FACS-based gating strategy to isolate CD142+ and CD142- ASPCs defined as Lineage negative (Lin-: CD31- CD45- TER119-) SCA-1+ SVF cells (**Methods**).



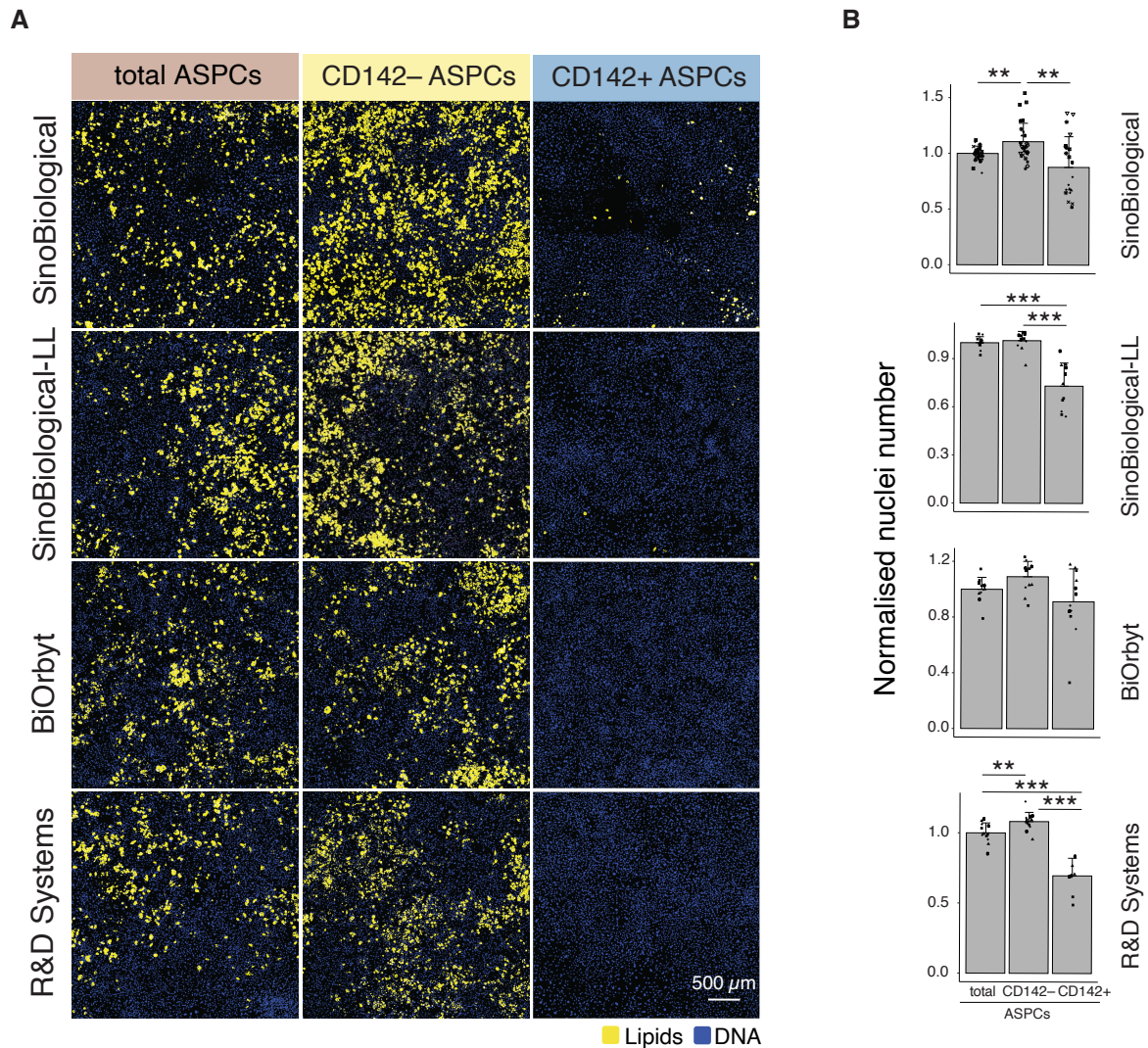
Appendix Figure S3. Top CD142+ markers are robust in freshly isolated CD142+ ASPCs across multiple omic datasets and some markers are maintained upon expansion

- (A) Volcano plot displaying differential gene expression results based on bulk RNA-seq data of freshly isolated CD142+ versus CD142- ASPCs; top CD142+ markers (**Appendix Table S1**) are highlighted in blue; significantly differentially expressed genes ($\log_2\text{FC} > 1$, $\text{FDR} < 0.05$) are highlighted in darker colours;
- (B) Volcano plot displaying differential protein abundance analysis based on mass spectrometry data of freshly isolated CD142+ versus CD142- ASPCs; top CD142+ markers (**Appendix Table S1**) are highlighted in blue; significantly differentially abundant proteins ($\log_2\text{FC} > 1$, $\text{FDR} < 0.05$) are highlighted in darker colours;
- (C) Scatter plot showing the correlation of $\log_2\text{FC}$ gene expression assessed by bulk RNA-seq of CD142+ over CD142- ASPCs in freshly isolated ASPCs (x-axis) versus expanded ASPCs (y-axis); top CD142+ markers (**Appendix Table S1**) are highlighted in blue;
- (D) Scatter plot showing the correlation of $\log_2\text{FC}$ protein abundance assessed by mass spectrometry of CD142+ over CD142- ASPCs in freshly isolated ASPCs (x-axis) versus expanded ASPCs (y-axis); top CD142+ markers (**Appendix Table S1**) are highlighted in blue.



Appendix Figure S4. Many top CD142+ markers are maintained in CD142+ ASPCs upon exposure to an adipogenic cocktail

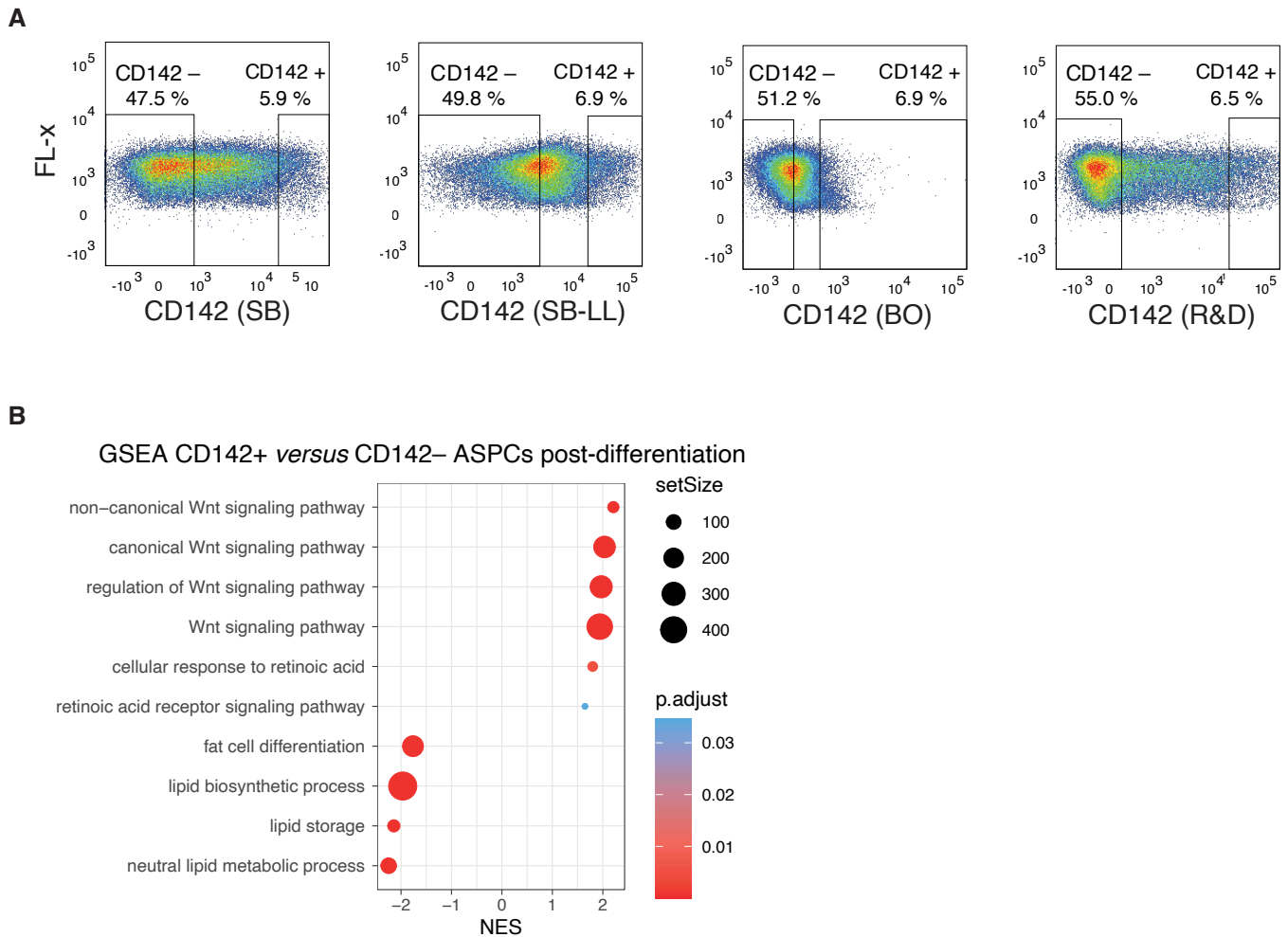
- (A) Gene expression heatmap across bulk RNA-seq samples of CD142+ and CD142- ASPCs post-differentiation (i.e., post exposure to an adipogenic cocktail); the selected genes are the top CD142+ markers (**Appendix Table S1**) with a positive log₂FC of CD142+ over CD142- ASPCs post-differentiation; the genes are ordered from greatest (top) to lowest (bottom) log₂FC and significantly differentially expressed genes (FDR < 0.05) are highlighted in red; log normalised expression is scaled by row;
- (B) Scatter plot showing the correlation of log₂FC gene expression based on bulk RNA-seq of CD142+ over CD142- ASPCs in freshly isolated (x-axis) *versus* post-differentiation (y-axis) ASPCs; top CD142+ markers (**Appendix Table S1**) are highlighted in blue.



Appendix Figure S5. CD142+ ASPCs are non-adipogenic regardless of the antibody used for isolation

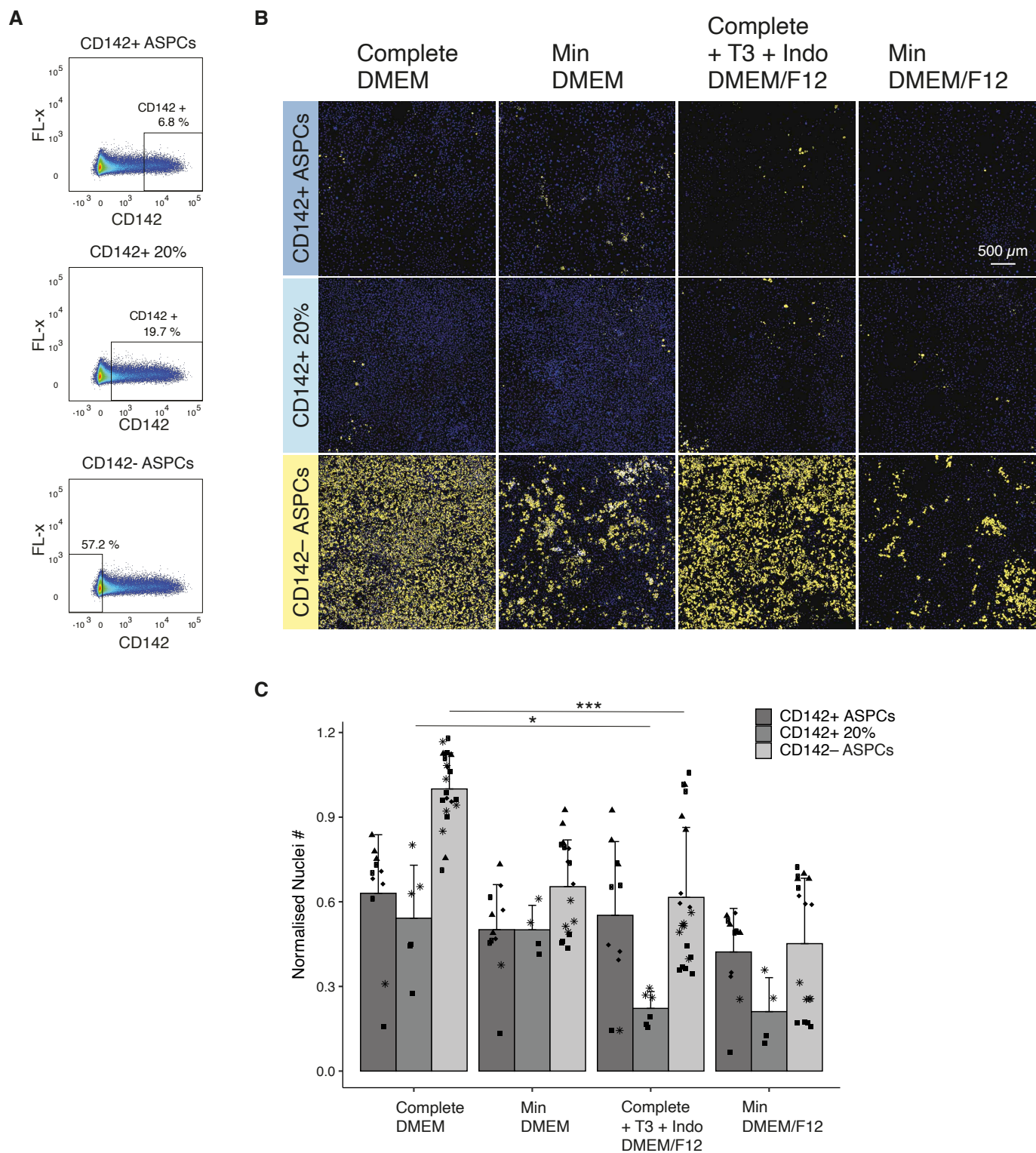
- (A)** Representative fluorescence microscopy images of total, CD142- and CD142+ ASPCs that were isolated with the use of distinct anti-CD142 antibodies, as also shown in **Appendix Fig. S6A**, after *in vitro* adipogenic differentiation; the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**);
- (B)** Bar plots showing the nuclei numbers for the cellular fractions shown in **A**, $n=9-15$, 3-4 biological replicates, 3-5 independent wells for each.

In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μ m; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure 6. FACS-based gating strategy to isolate CD142+ ASPCs using different antibodies and GSEA of CD142+ versus CD142– ASPCs post-differentiation

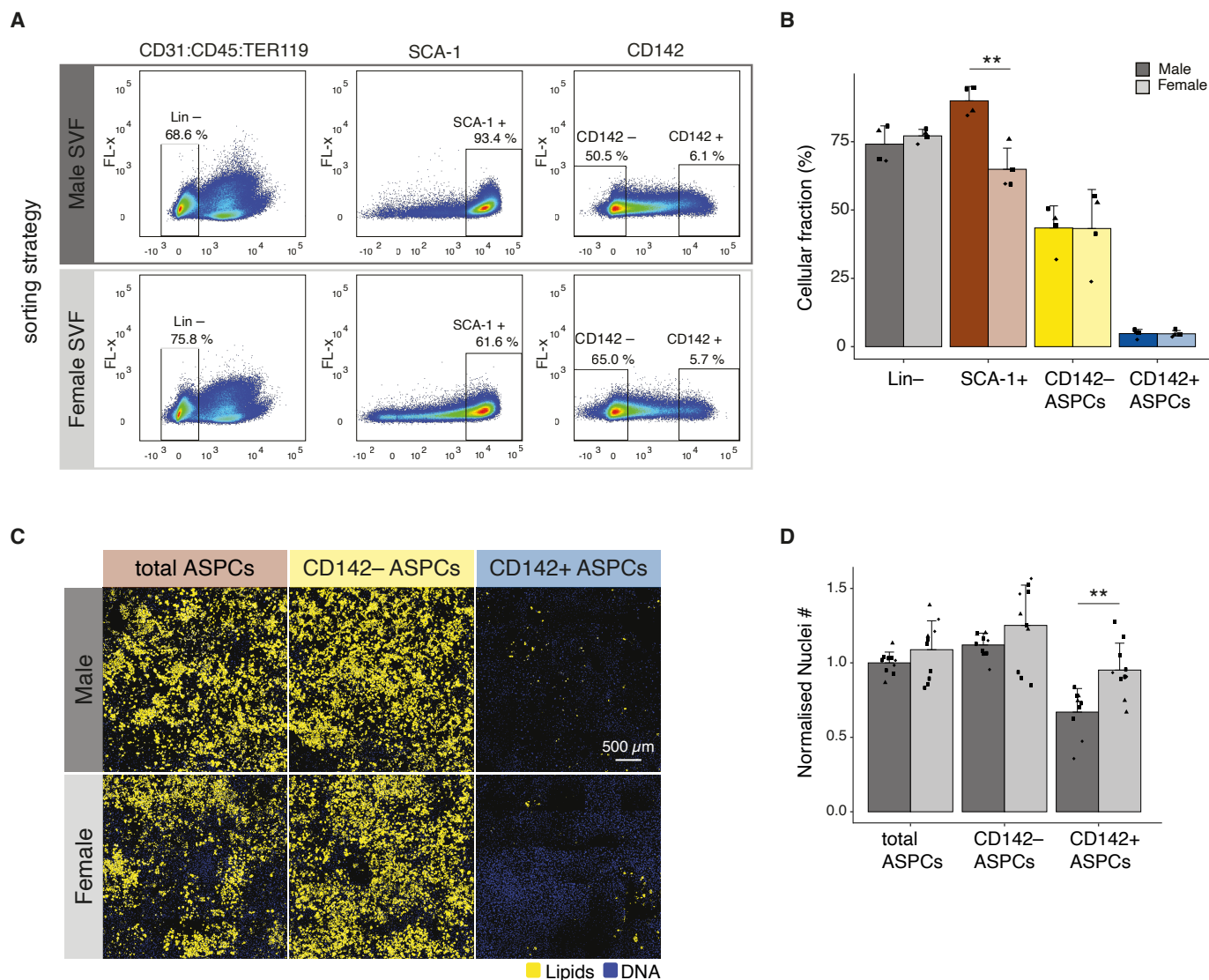
- (A)** FACS-based gating strategy to isolate CD142+ ASPCs defined as 5-7% CD142+ ASPCs, and CD142– ASPCs defined as around 50% cells negative for the CD142 marker, using four different antibodies: “SB” – SinoBiological anti-CD142–PE (as already displayed in **Appendix Fig. S2B**), “SB-LL” SinoBiological anti-CD142 coupled to PE in house (**Methods**), “BO” – BiOrbyt anti-CD142–PE and “R&D” – R&D Systems anti-CD142–PE; the experiment was repeated four times with similar results, shown here is one representative biological replicate;
- (B)** GSEA results performed on differential gene expression between CD142+ versus CD142– ASPCs post-differentiation; NES: normalized enrichment score.



Appendix Figure S7. CD142+ ASPCs are non-adipogenic regardless of the stringency of the gating strategy

- (A) FACS-based gating strategy to isolate CD142+ (5-7%), 20% CD142+ and CD142- ASPCs; the experiment was repeated at least five times with similar results, shown here is one representative biological replicate;
- (B) Representative fluorescence microscopy images of CD142+, 20% CD142+ and CD142- ASPCs isolated following the sorting strategy shown in **A**, after *in vitro* adipogenic differentiation with the indicated adipogenic differentiation cocktails (**Methods**); the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**);
- (C) Bar plots showing the nuclei numbers for the cellular fractions shown in **B**, marker shapes correspond to different biological replicates, n=8-17, 3-5 biological replicates, 2-5 independent wells for each;

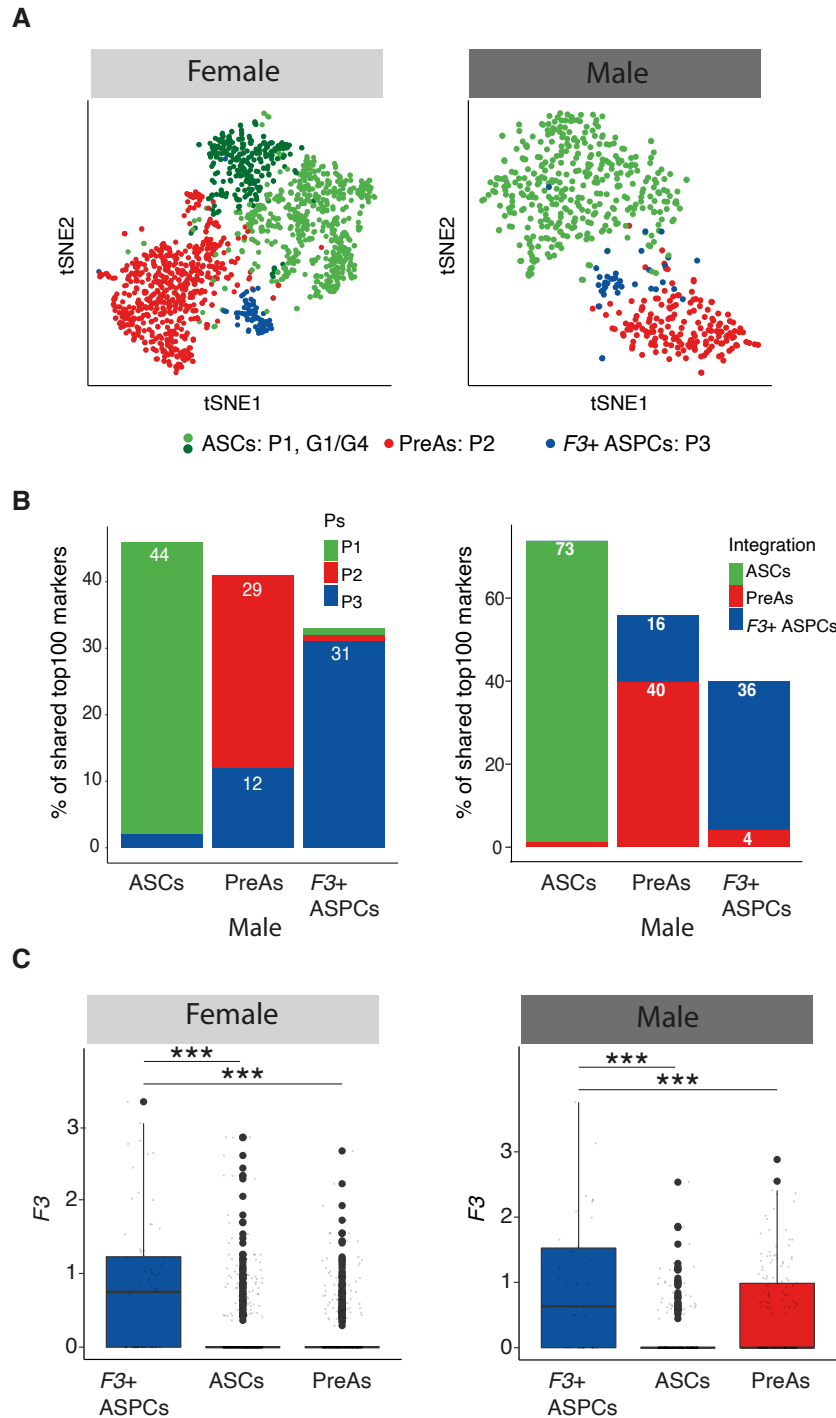
In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μ m; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S8. CD142+ ASPCs are non-adipogenic in male and female mice

- (A)** FACS-based gating strategy of male and female Lin⁻, SCA-1⁺ (ASPC), CD142⁻ and CD142⁺ ASPC cellular fractions within the SVF; the experiment was repeated four times with similar results, shown here is one representative biological replicate;
- (B)** Bar plots showing the percentage of the cellular fractions gated as shown in **A**, the experiment was repeated n=4;
- (C)** Representative fluorescence microscopy images of male- and female-derived ASPCs, CD142⁻ and CD142⁺ ASPCs, isolated following the sorting strategy shown in **A** after *in vitro* adipogenic differentiation; the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**): n=9-10, 4 biological replicates, 2-3 independent wells for each;
- (D)** Bar plots showing the nuclei numbers for the cellular fractions shown in **C**, marker shapes correspond to different biological replicates, n=9-10, 4 biological replicates, 2-3 independent wells for each;

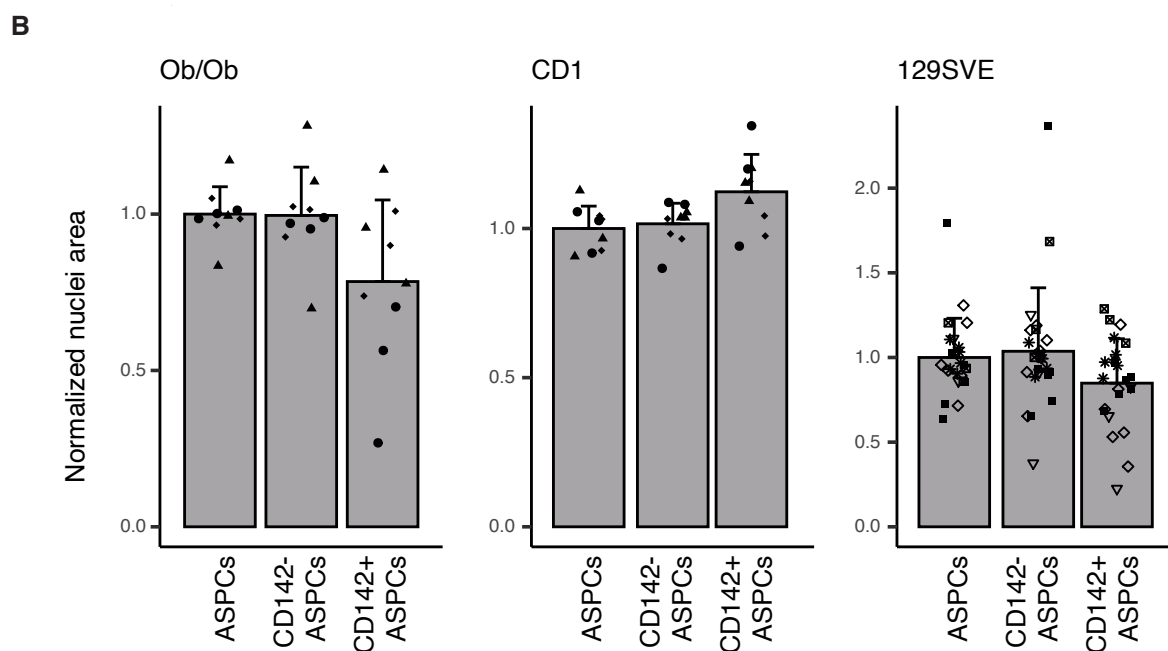
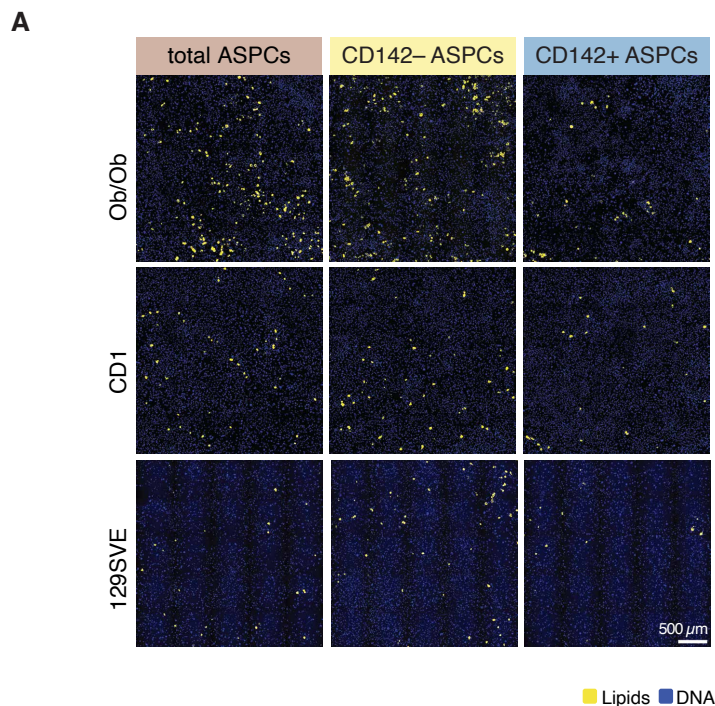
In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μ m; bar colours: total ASPCs - brown, CD142⁻ ASPCs — yellow, CD142⁺ ASPCs — blue, female — lighter colours, male — darker colours; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S9. scRNA-seq validates the molecular existence of *F3*(CD142)+ ASCs in male and female mice

- (A) t-SNE cell map of an scRNA-seq dataset of female (**left**) or male (**right**) mouse subcutaneous ASCs: adipose stem cells (ASCs) — green, pre-adipocytes (PreAs) — red, *F3*(CD142)+ ASCs — blue;
- (B) Percentage of the top 100 markers (**left**) of the three clusters (P1 (equivalent to ASCs) in green, P2 (equivalent to PreAs) in red and P3 (equivalent to *F3*(CD142)+ ASCs) in blue) published in Schwalie *et al.*, 2018 (analysis performed on female ASCs, t-SNE shown in panel **A left**), or (**right**) of the top 100 markers of the three main ASC populations based on the integration of adult (male and female) ASCs (**Fig. 1A**, Ferrero, Rainer and Deplancke, 2020) that overlap with the top markers of the same three subpopulations identified in male ASCs (t-SNE shown in panel **A right**);
- (C) Boxplots showing the distribution of log normalised expression of *F3* in female (**left**) and male (**right**) ASCs across the three main ASC subpopulations;

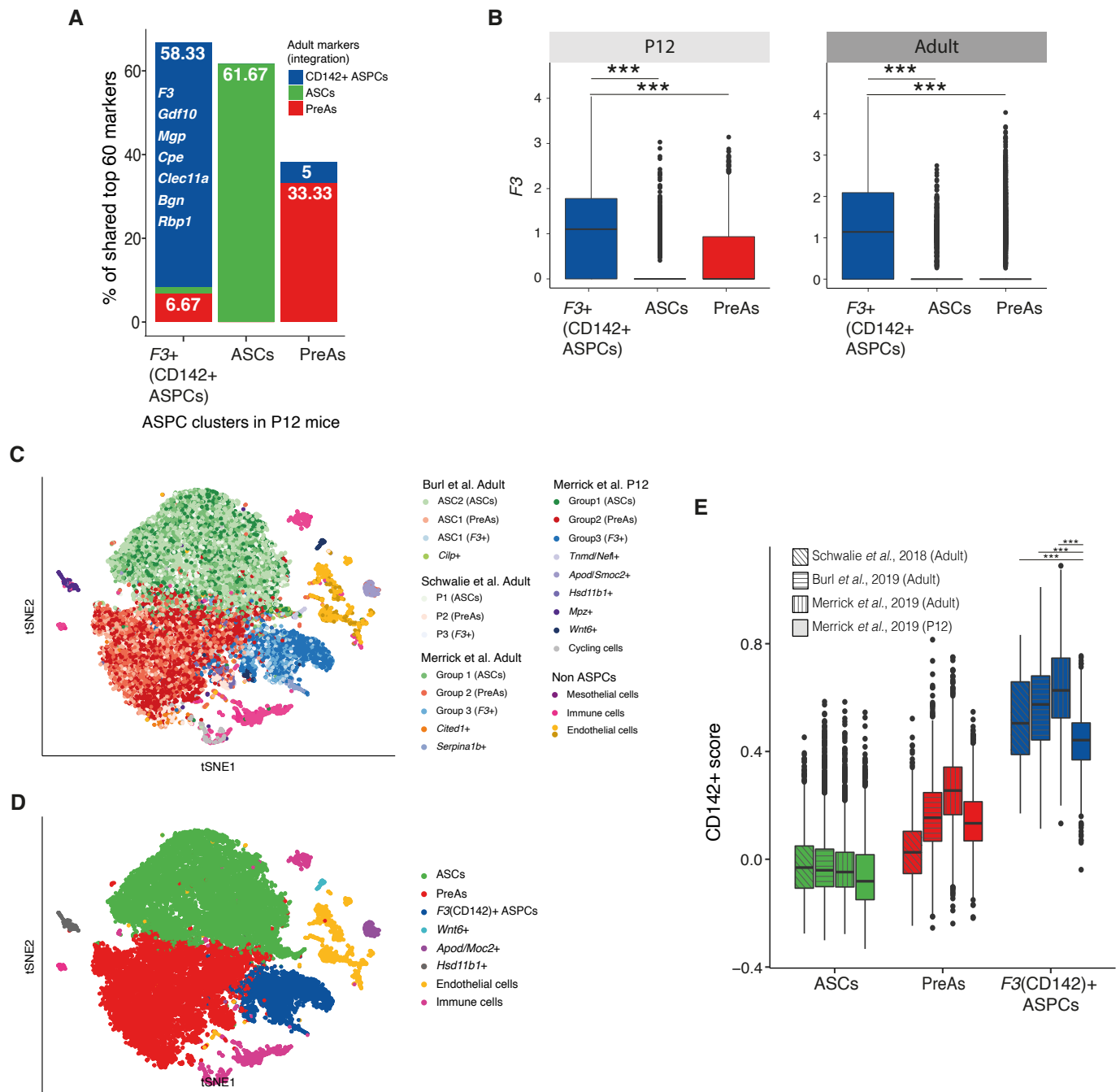
Adipose stem cells (ASCs) — green, pre-adipocytes (PreAs) — red, *F3*(CD142)+ ASCs — blue; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, ANOVA and Tukey HSD *post hoc* test, for statistical details see **Methods**.



Appendix Figure S10. CD142+ ASPCs are non-adipogenic regardless of the mouse strain

- (A) Representative fluorescence microscopy images of total, CD142- and CD142+ ASPCs isolated from different mouse strains (**Methods**), after *in vitro* adipogenic differentiation;
- (B) Barplot showing the normalized nuclei area for the cellular fraction and mouse strains shown on **A** and **Fig. 1M**; marker shapes correspond to different biological replicates, n=9-24, 3-5 biological replicates, 3-6 independent wells for each.

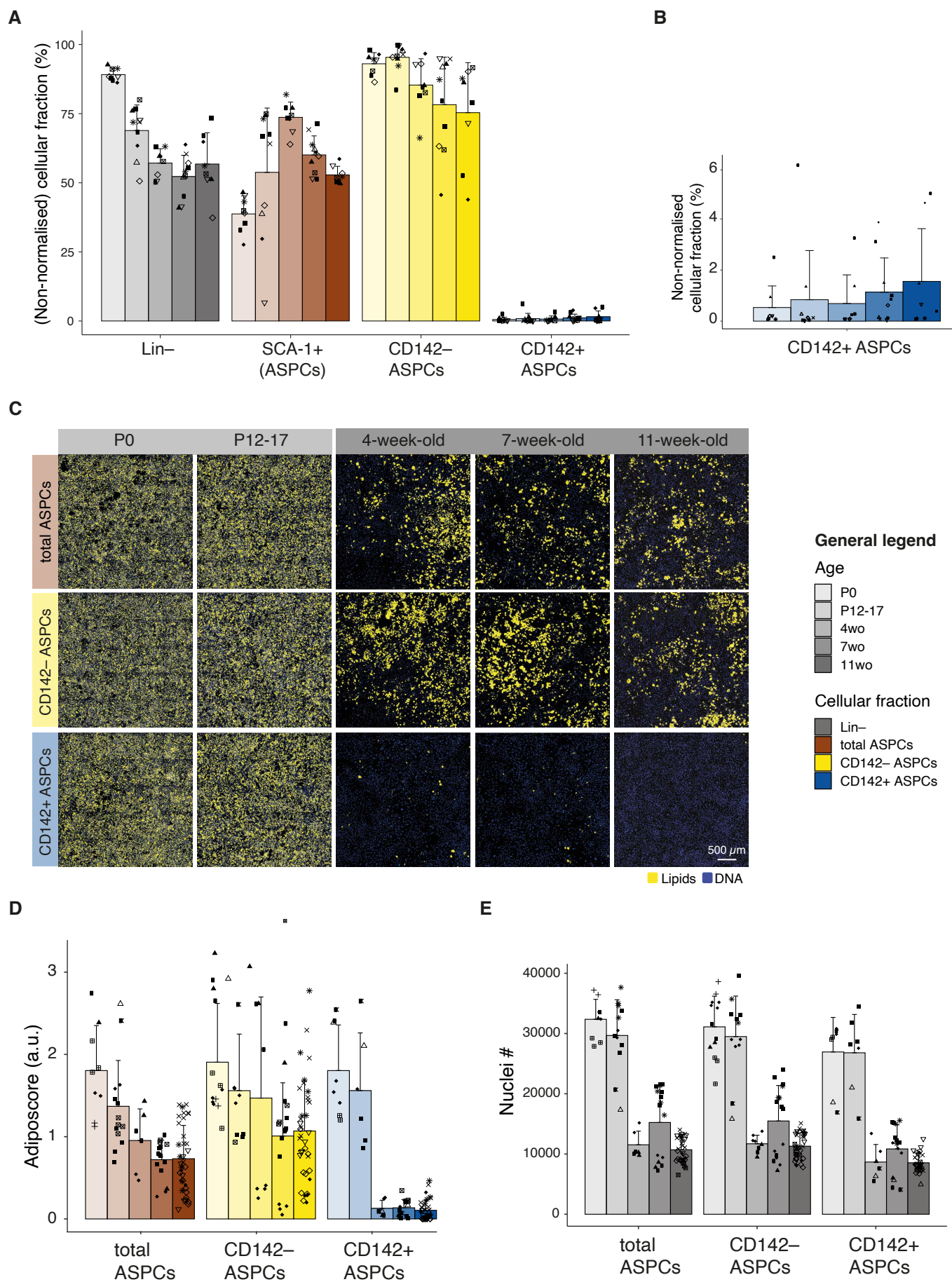
* $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S11. The CD142+ score of F3(CD142)+ ASCs is less pronounced in P12 mice

- (A) Percentage of the top 60 markers (**Methods**) of the three main ASPC subpopulations (adipose stem cells (ASCs) in green, pre-adipocytes (PreAs) in red, F3(CD142)+ ASCs in blue) based on the integration of adult ASCs (**Fig. 1A**, Ferrero, Rainer and Deplancke, 2020) that overlap with the top 60 markers of the same three subpopulations identified in P12 ASCs;
- (B) Boxplots showing the distribution of log normalized expression of *F3* in P12 (**left**) and adult (**right**) ASCs across the main ASPC subpopulations;
- (C) t-SNE map of the integration analysis of adult- (Schwalie *et al.*, 2018, Burl *et al.*, 2019, Merrick *et al.*, 2019) and P12- (Merrick *et al.*, 2019) derived ASCs coloured by the clustering results identified in each dataset individually;
- (D) t-SNE map of the integration analysis described in **C** coloured by the clustering results, showing the main three ASCs subpopulations as well as other ASPC subgroups, endothelial and immune cells;
- (E) Boxplot showing the distribution of “CD142+ score” (**Appendix Table S1**) in the four different datasets used in the integration described in **C** across the three main ASPC subpopulations.

F3(CD142)+ ASCs — blue, adipose stem cells (ASCs) — green, pre-adipocytes (PreAs) — red; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, one-way ANOVA and Tukey HSD *post hoc* test (**B**, **E** null hypothesis: no difference between means of F3+ ASCs in each dataset), for statistical details see **Methods**.

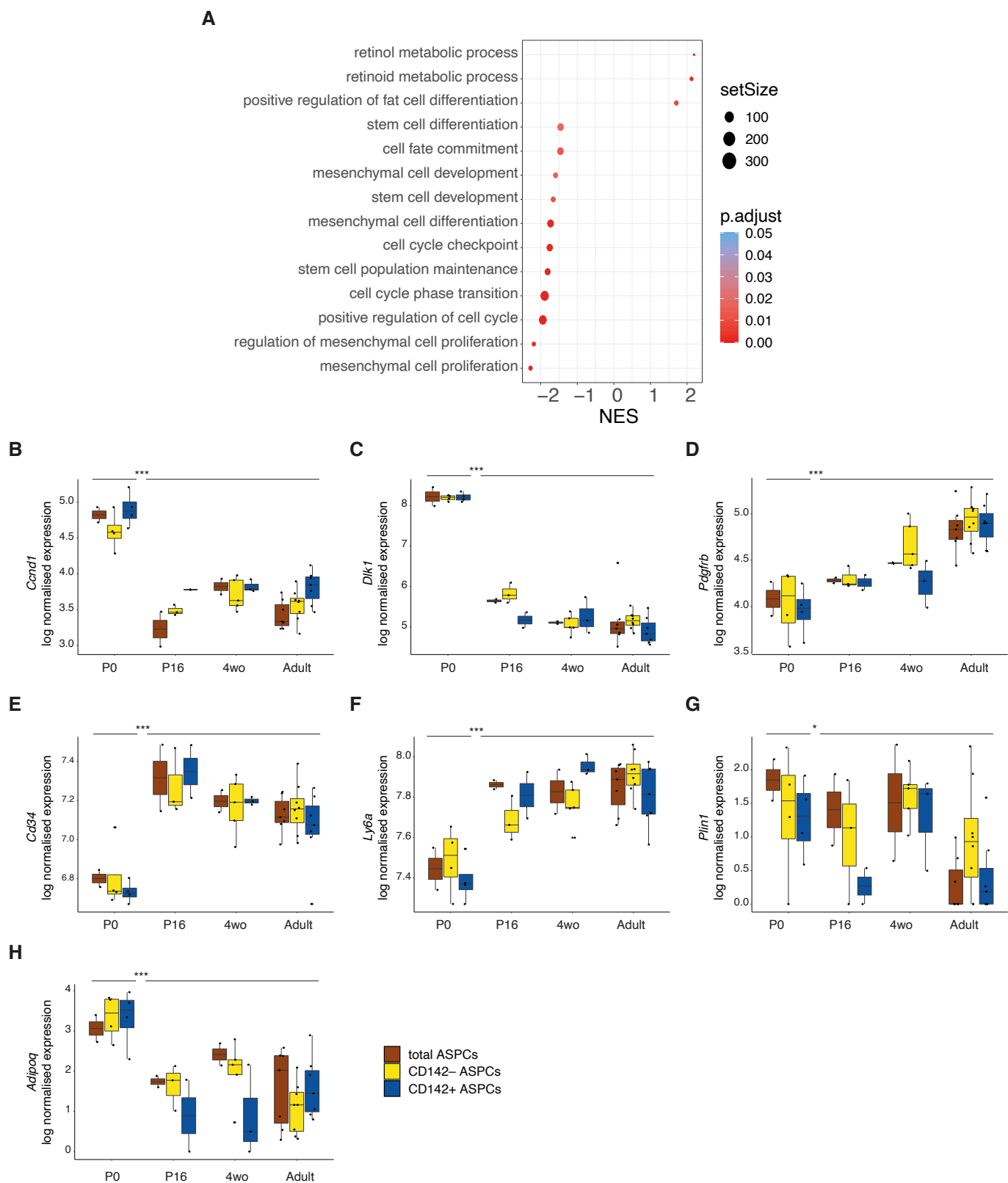


Appendix Figure S12 | See next page for caption

Appendix Figure S12. The non-adipogenic character of CD142+ ASPCs is age-dependent

- (A) Bar plot showing the non-normalised parental percentage of cellular fractions gated for FACS-based isolation of the indicated subpopulations at different ages: P0, P12-17, 4wo, 7wo and 11wo, marker shapes correspond to different biological replicates, n=8-10;
- (B) Bar plot showing the non-normalised cellular fractions gated for FACS-based isolation of P0-, P12-17-, 4wo-, 7wo- and 11wo-derived CD142+ ASPCs, marker shapes correspond to different biological replicates, n=8-10;
- (C) Representative fluorescence microscopy images of P0-, P12-17-, 4wo-, 7wo- and 11wo-derived total, CD142- and CD142+ APSCs after in vitro adipogenic differentiation; the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**);
- (D) Fraction of differentiated cells per ASPC type shown in **C**, as quantified the “adiposcore”; bar colour shading corresponds to the individual ages as indicated, marker shapes correspond to different biological replicates, n=6-35, 3-13 biological replicates, 2-6 independent wells for each;
- (E) Bar plot showing the nuclei numbers for the cellular fractions shown in **C**; marker shapes correspond to different biological replicates, n=6-35, 3-13 biological replicates, 2-6 independent wells for each;

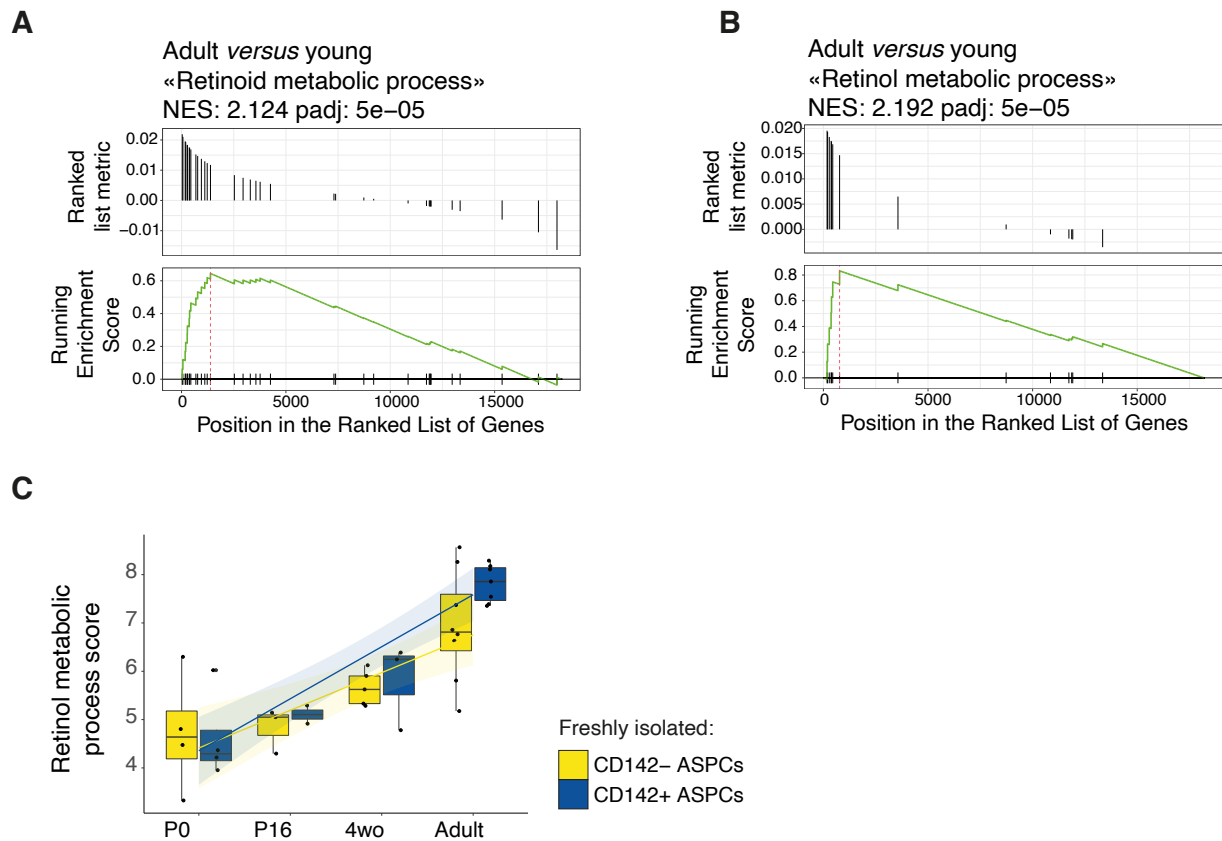
In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μ m, bar colours: total ASPCs - brown, CD142- ASPCs - yellow, CD142+ ASPCs - blue; shades are indicative of age (young - light, adult - dark); *P $\leq$ 0.05, **P $\leq$ 0.01, ***P $\leq$ 0.001, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S13. Gene expression profiling of P0 ASPCs suggests that these cells are in a “naïve” state

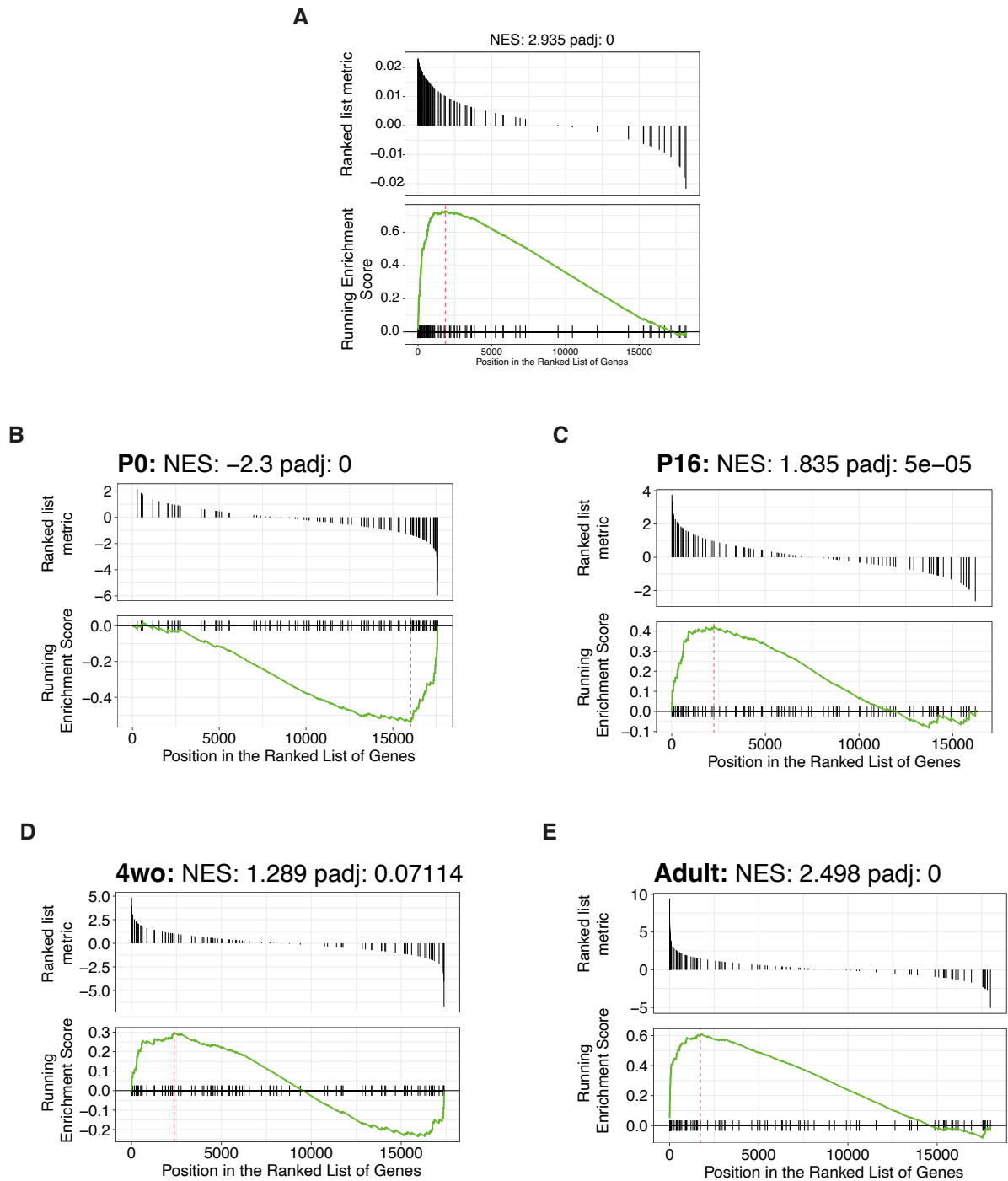
- (A)** Significantly enriched terms found by GSEA performed on the genes driving the first principal component of the PCA representation of bulk RNA-seq samples of P0, P16-, 4-, 7- and 11wo-derived total, CD142- and CD142+ ASPCs (**Fig. 2F**); negative and positive NES (Normalized enrichment score) indicates enrichment in young or adult samples respectively;
- (B-H)** Boxplots showing the distribution of the log normalized expression of **(B)** *Ccnd1* (coding for Cyclin D1), **(C)** *Dlk1* (coding for PREF-1), **(D)** *Pdgfrb*, **(E)** *Cd34*, **(F)** *Ly6a* (coding for SCA-1), **(G)** *Plin1* (coding for Perilipin), **(H)** *Adipoq* (coding for Adiponectin) in total ASPCs, CD142- and CD142+ ASPCs at the indicated ages;

* $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S14. Genes linked to retinol/retinoid metabolic processes are enriched in adult compared to young mouse-derived ASPCs

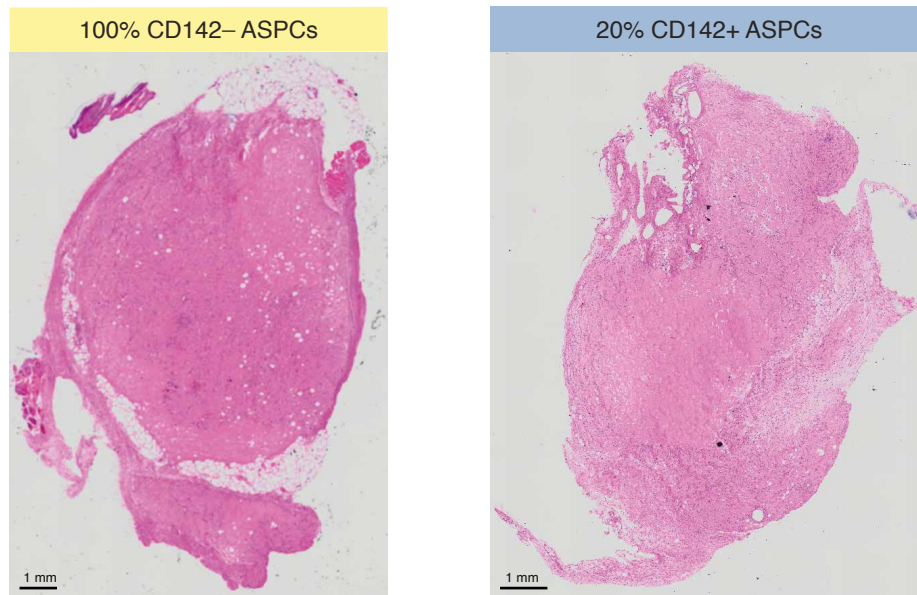
- (A) The term “retinoid metabolic process” (GO:0001523) was identified as significantly enriched in freshly isolated adult- compared to young mouse-derived ASPCs when performed on the genes driving the PC1 of the PCA representation based on bulk RNA-seq samples of P0-, P16-, 4-, 7- and 11wo-derived ASPCs (see **Fig. 2F**);
- (B) The term “retinol metabolic process” (GO:0042572) was identified as significantly enriched in adult- compared to young mouse-derived ASPCs when performed on the genes as described in **A**;
- (C) Boxplot showing the distribution of the “retinol metabolic process score” based on the expression of the genes linked to “retinol metabolic process” (GO:0001523) for CD142- and CD142+ ASPCs at the indicated ages.



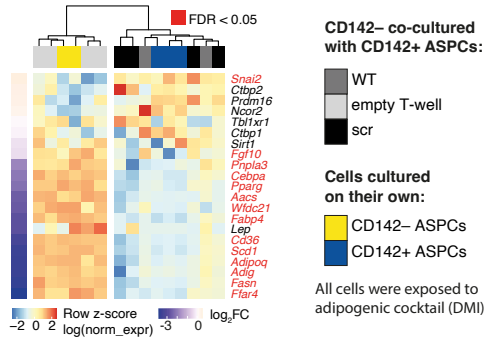
Appendix Figure S15. The molecular “CD142+ signature” appears in CD142+ ASPCs in an age-dependent manner

- (A)** GSEA of the top CD142+ markers (**Appendix Table S1**) performed on the genes driving PC1 of the PCA representation of bulk RNA-seq samples of P0-, P16-, 4-, 7- and 11wo-derived freshly isolated total, CD142- and CD142+ ASPCs (see **Fig. 2F**);
- (B-E)** GSEA of the top CD142+ markers (**Appendix Table S1**) performed on the differential expression data between CD142+ and CD142- ASPCs of P0 (**B**), P16 (**C**), 4wo (**D**) and adults (7 and 11wo) (**E**).

A

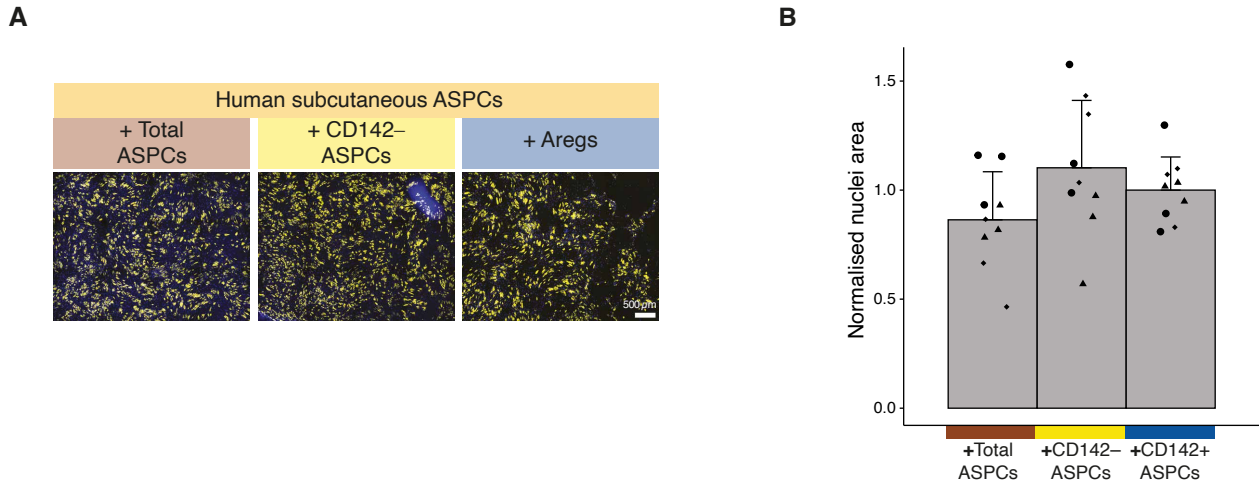


B



Appendix Figure S16. The CD142+ ASPCs phenotype is maintained *in-vivo*

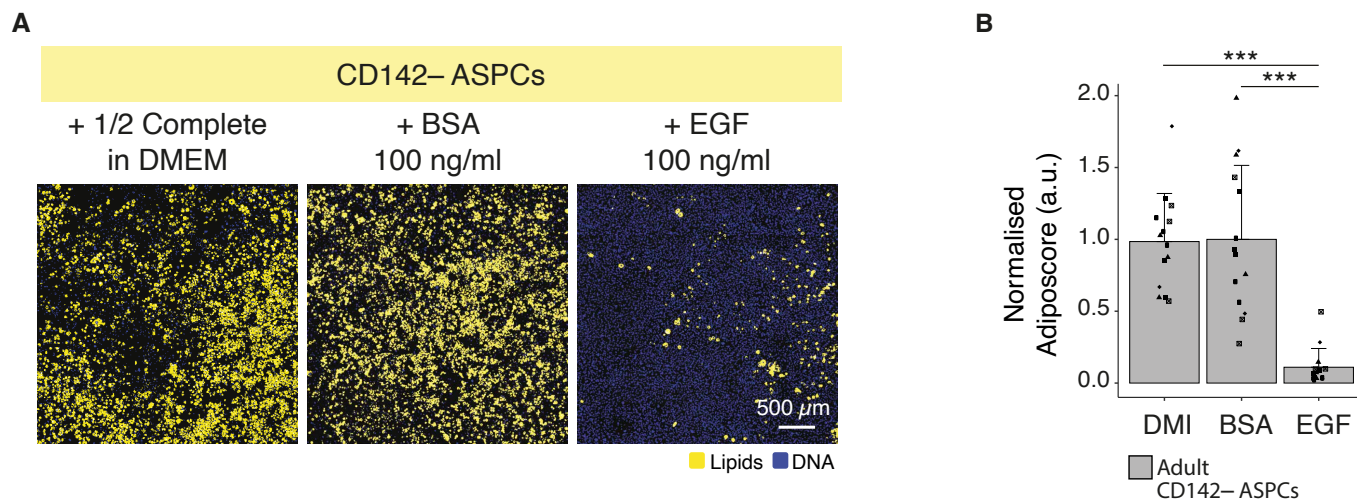
- (A) Full histology cuts of Matrigel implants after fixation with 4% PFA and dehydration shown on **Fig. 3D**. The plugs were injected with 100% CD142- ASPCs (left) or 80% CD142- ASPCs and 20% CD142+ ASPCs (right). Both images show plugs that were dissected from a female mouse after Matrigel injection and 3 weeks on high-fat diet. In pink: Matrigel, in white: adipocytes. Scale bar = 1mm. Staining H&E Fat;
- (B) Expression heatmap listing genes linked to “white fat cell differentiation” (GO:0050872) across bulk RNA-seq samples of CD142+ and CD142- ASPCs after adipogenic differentiation (i.e., after exposure to an adipogenic cocktail, **Methods**); the genes are ordered from top to bottom by the log₂FC of CD142+ over CD142- ASPCs after adipogenic differentiation; significantly differentially expressed genes (FDR < 0.05) are coloured in red; log normalised expression scaled by row.



Appendix Figure S17. Mouse Aregs exert an inhibitory effect on human-derived subcutaneous ASPCs

- (A)** Representative fluorescence microscopy images of human-derived stromal vascular fraction (SVF), containing the ASPCs after *in vitro* adipogenic differentiation when co-cultured in a transwell set up with mouse total, CD142- or CD142+ ASPCs; the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**);
- (B)** Bar plots showing the nuclei area for cellular fractions shown in **A**; marker shapes correspond to different biological replicates, n=9, 3 biological replicates, 3 independent wells for each.

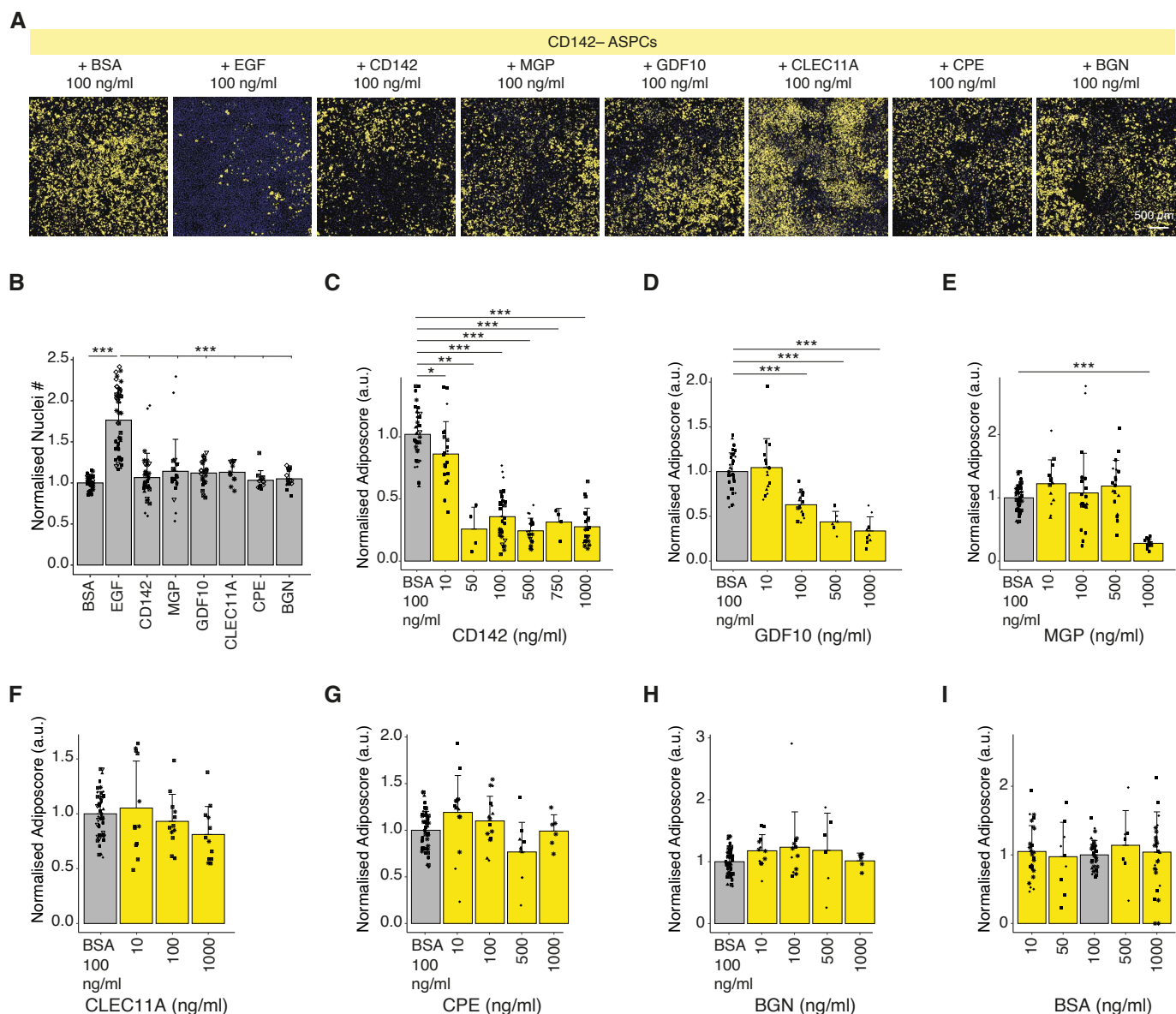
In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μm ; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S18. Positive and negative controls of chemical inhibition of adipogenesis

- (A)** Representative fluorescence microscopy images of CD142– ASPCs after *in vitro* adipogenic differentiation with the differentiation cocktail (1/2 Complete in DMEM, **Methods**) supplemented with control recombinant proteins BSA (negative control) and EGF (positive control) of inhibition of adipogenesis at 100 ng/ml; the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**);
- (B)** Fraction of differentiated CD142– ASPCs treated with the indicated recombinant proteins shown in **A**, marker shapes correspond to different biological replicates, n=12, 4 biological replicates, 3 independent wells for each;

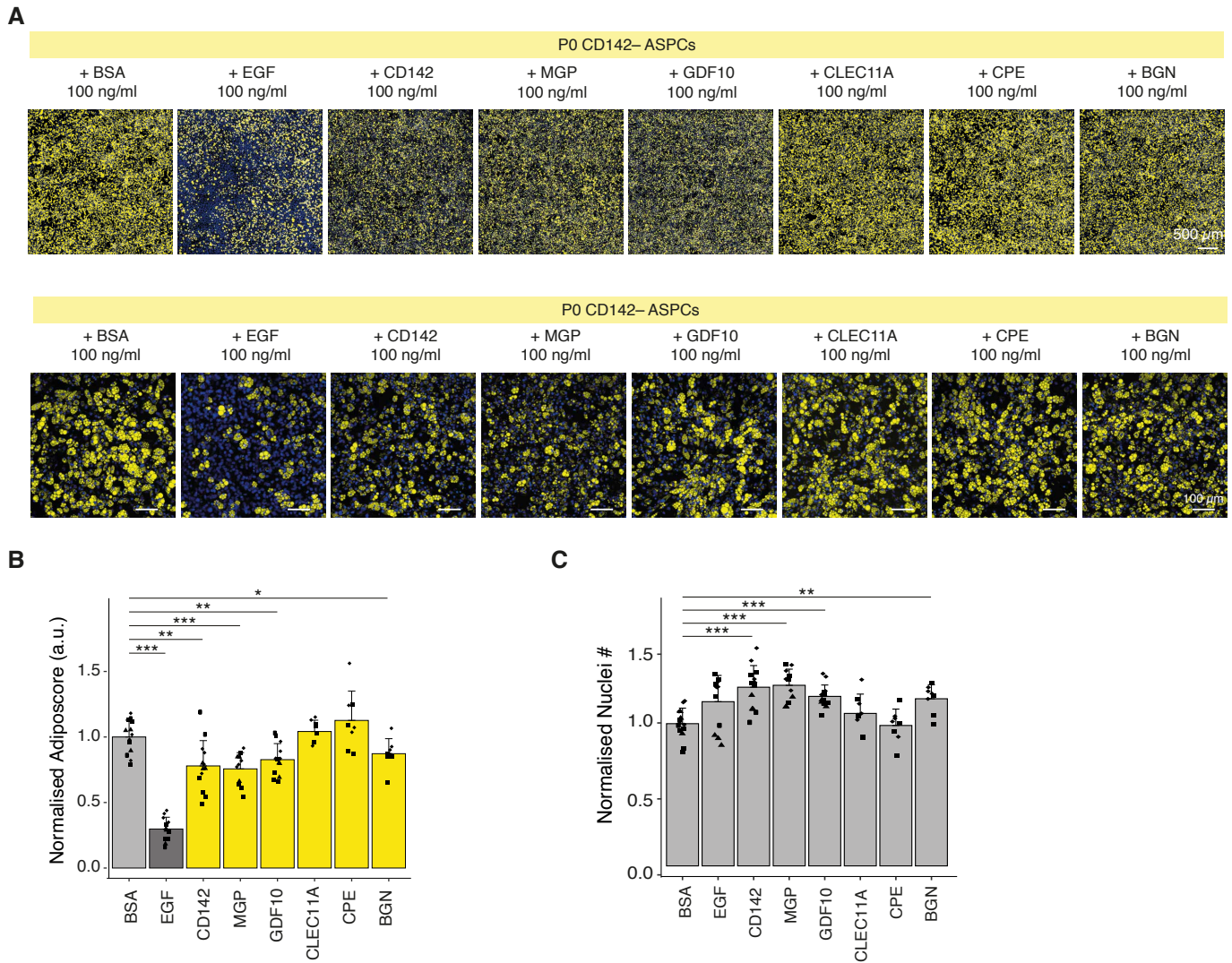
In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μ m; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S19. Adipogenic inhibition of adult mouse-derived CD142–ASPCs using recombinant proteins

- (A)** Representative fluorescence microscopy images of adult-derived CD142–ASPCs after *in vitro* adipogenic differentiation; the induction cocktail was supplemented with recombinant proteins corresponding to controls and to the selected CD142+ ASPC (Areg)-specific candidates: BSA (negative control), EGF (positive control), CD142, MGP, GDF10, CLEC11A, CPE and BGN at 100 ng/ml; the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**);
- (B)** Bar plots showing the nuclei numbers for cellular fractions shown in **A**, marker shapes correspond to different biological replicates, $n=11-59$, 2-10 biological replicates, 3-9 independent wells for each;
- (C-I)** Bar plots showing the fraction of differentiated adult mouse-derived CD142–ASPCs treated with the indicated recombinant proteins at the indicated concentrations as quantified by the “adiposcore”; marker shapes correspond to different biological replicates.

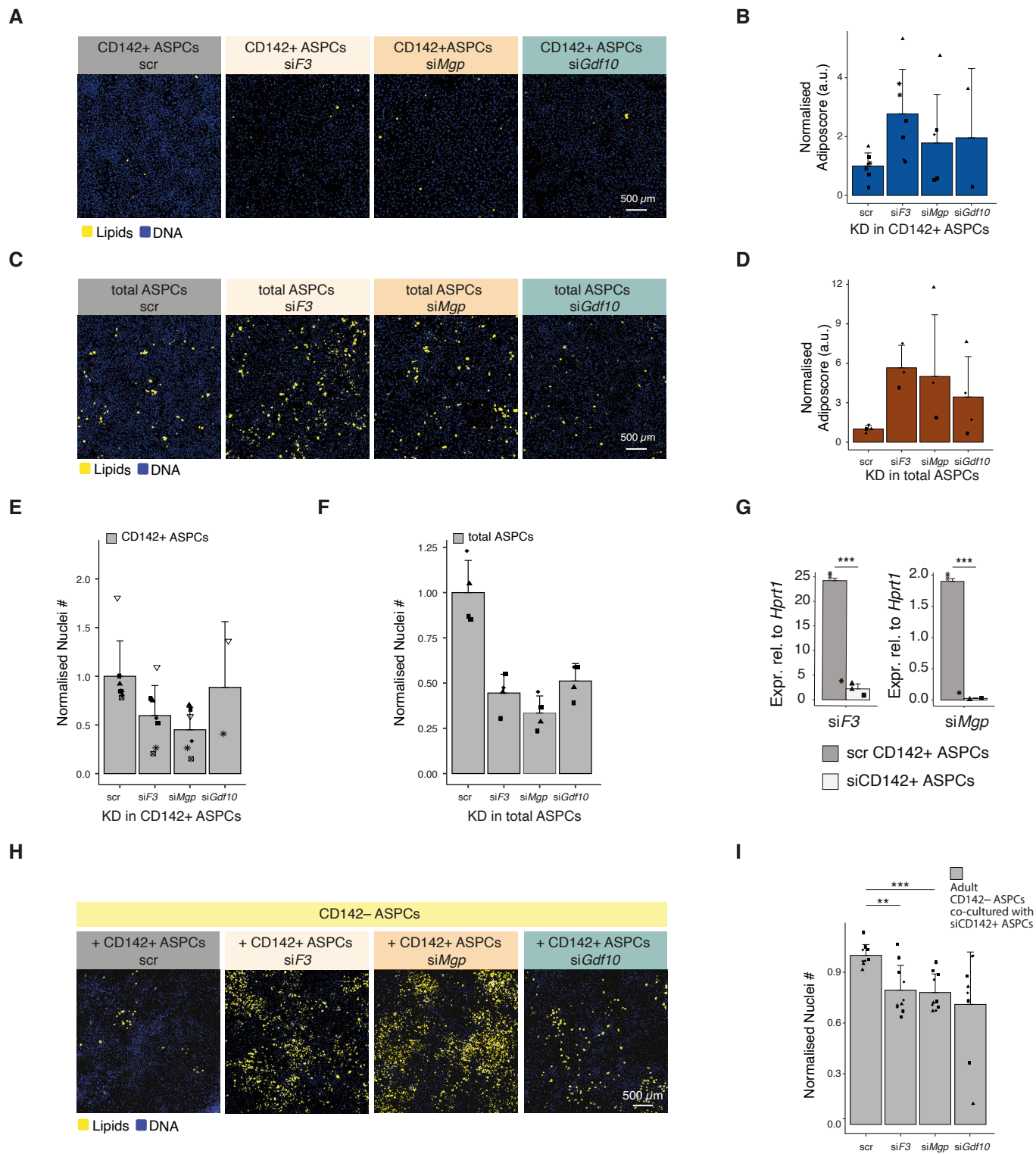
In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μm , bar colours: CD142–ASPCs treated with the indicated recombinant protein of interest — yellow, nuclei (**B**) or recombinant BSA treatment (negative control) (**C-I**) — light grey; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S20. Adipogenic inhibition of P0 mouse-derived CD142– ASPCs with recombinant proteins

- (A)** Representative fluorescence microscopy images of P0 mouse-derived CD142– ASPCs after *in vitro* adipogenic differentiation; the induction cocktail was supplemented with recombinant proteins corresponding to controls and to the selected CD142+ ASPC (Areg)-specific candidates: BSA (negative control), EGF (positive control), CD142, MGP, GDF10, CLEC11A, CPE and BGN at 100 ng/ml; the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**); scale bars 500 μ m (**top**), 100 μ m (**bottom**);
- (B)** Fraction of differentiated CD142– ASPC cells, as quantified by the “adiposcore”, treated with the indicated recombinant proteins shown in **A**; marker shapes correspond to different biological replicates, n=8-13, 1-3 biological replicates, 2-4 independent wells for each;
- (C)** Bar plots showing the nuclei numbers for the cellular fractions shown in **A**; marker shapes correspond to different biological replicates, n=11-59, 2-10 biological replicates, 3-9 independent wells for each;

In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); bar colours: CD142– ASPCs treated with the indicated recombinant protein of interest — yellow, recombinant BSA treatment (negative control) (**B**) or nuclei (**C**) — light grey, EGF treatment (positive control) (**B**) — dark grey; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.

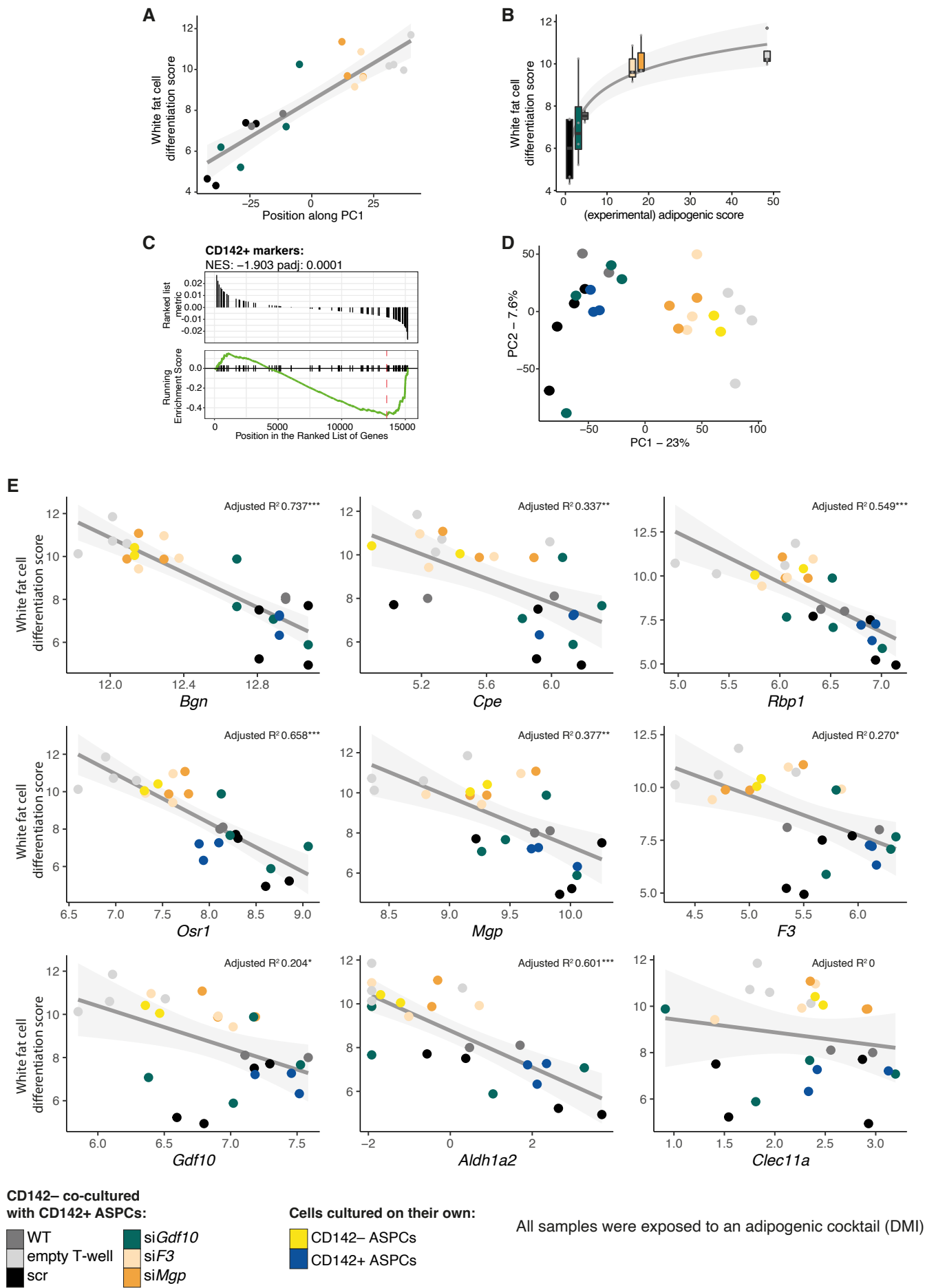


Appendix Figure S21 | See next page for caption

Appendix Figure S21. siRNA mediated knockdown of CD142+ ASPC (Areg) candidates

- (A) Representative fluorescence microscopy images of CD142+ ASPCs (Aregs) after *in vitro* adipogenic differentiation subjected to siRNA control (scrambled (scr)) or siRNA-mediated knockdowns of selected CD142+ ASPC (Areg)-specific candidate genes: *F3*, *Mgp*, and *Gdf10*;
- (B) Fraction of differentiated CD142+ ASPCs (Aregs), as quantified by the “adiposcore”, for each siRNA knockdown assay shown in **A**; marker shapes correspond to different biological replicates, n=2-7, 2-5 biological replicates;
- (C) Representative fluorescence microscopy images of total ASPCs after *in vitro* adipogenic differentiation subjected to control (scrambled (scr)) or siRNA-mediated knockdowns of selected CD142+ ASPC (Areg)-specific candidate genes: *F3*, *Mgp*, and *Gdf10*;
- (D) Fraction of differentiated total ASPCs, as quantified by the “adiposcore”, for each siRNA knockdown assay shown in **C**; marker shapes correspond to different biological replicates, n=4, 3 biological replicates;
- (E) Bar plot showing the nuclei numbers for the cellular fractions shown in **A**; marker shapes correspond to different biological replicates, n=2-7, 2-5 biological replicates;
- (F) Bar plot showing the nuclei numbers for the cellular fractions shown in **C**; marker shapes correspond to different biological replicates, n=4, 3 biological replicates;
- (G) qPCR-measured gene expression of *F3* and *Mgp* in CD142+ ASPCs (Aregs) subjected to control (scrambled (scr)) or siRNA-mediated knockdown of the indicated gene normalised to *Hprt1* expression, 1-2 biological replicates, 3 independent wells for each;
- (H) Representative fluorescence microscopy images of CD142- ASPCs, co-cultured with CD142+ ASPCs (Aregs) subjected to control (scrambled (scr)) or siRNA-mediated knockdowns of selected CD142+ ASPCs (Areg)-specific candidate genes: *F3*, *Mgp*, and *Gdf10* after adipogenic differentiation; the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**);
- (I) Bar plot showing the nuclei numbers for the cellular fractions shown in **H**; marker shapes correspond to different biological replicates, n=10, 4 biological replicates.

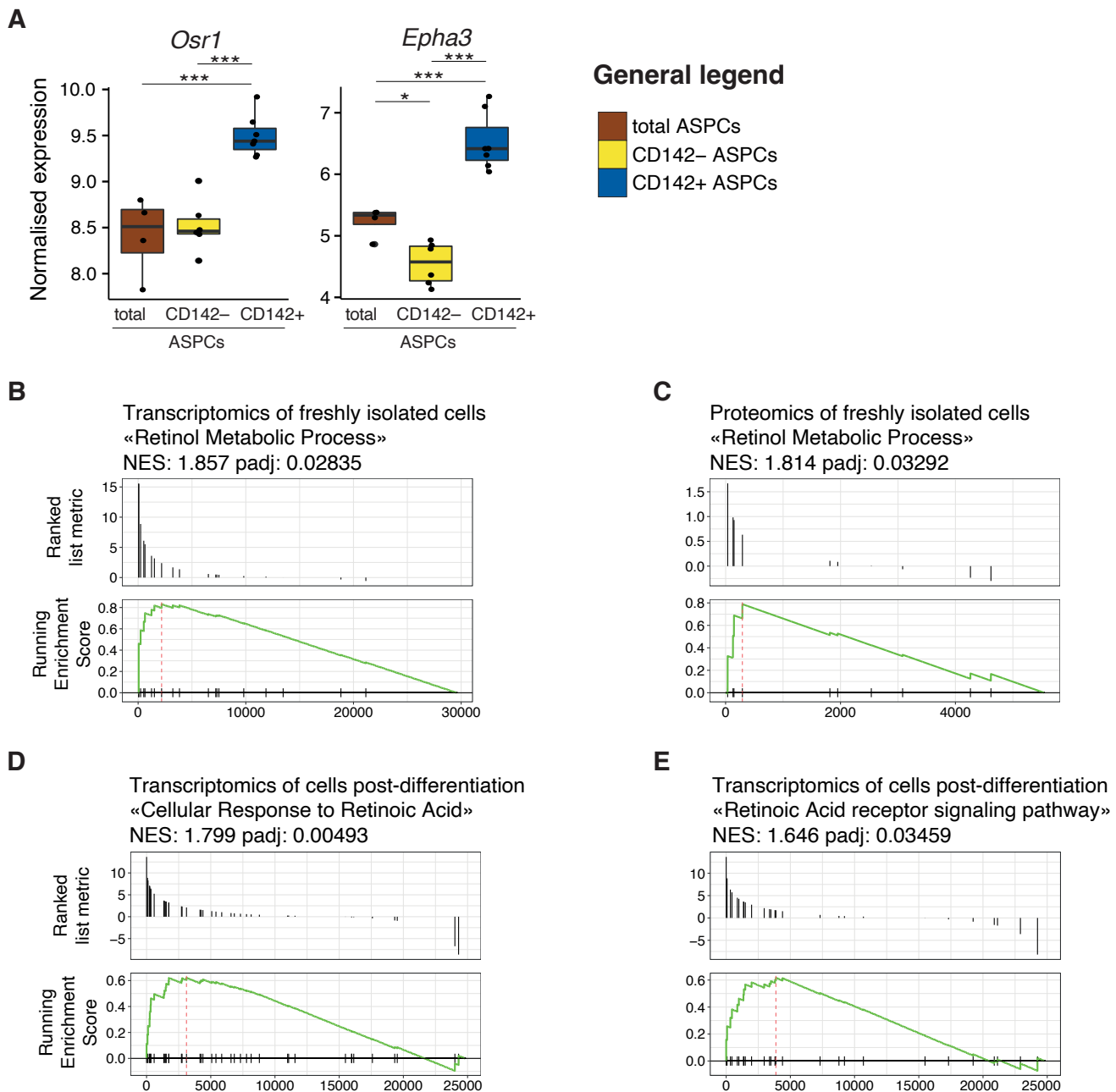
In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500µm, bar colours: total ASPCs — brown, CD142+ ASPCs — blue, nuclei (**E-I**) - grey; *P ≤ 0.05, **P ≤ 0.01, ***P ≤ 0.001, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S22 | See next page for caption

Appendix Figure S22. CD142– ASPCs exposed to the secretome of “active Aregs” (CD142+ ASPCs) exhibit upregulation of CD142+ ASPC (Areg) markers and retinoic acid-related genes

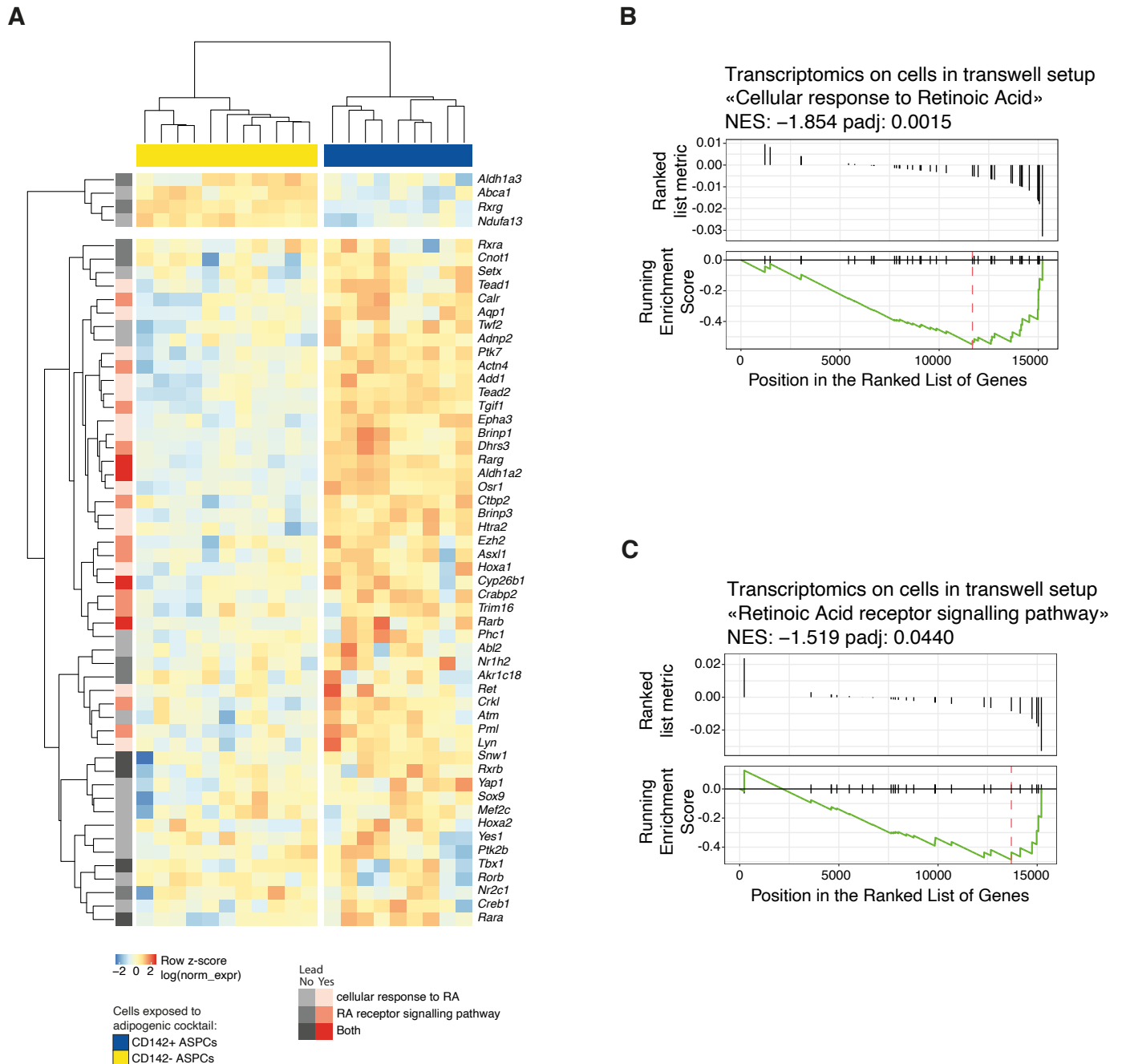
- (A) Correlation of the position on the PC1 of the PCA shown in **Fig. 4C** and the “white fat cell differentiation score” (GO:0050872) (**Methods**); the colour legend is listed at the bottom of the figure;
- (B) Correlation of the experimental adipogenic score *versus* the “white fat cell differentiation score” (GO:0050872) (**Methods**);
- (C) GSEA results of the top CD142+ ASPC (Aregs) markers (**Appendix Table S1**) performed on the genes driving PC1 of the PCA shown in **Fig. 4C**;
- (D) PCA of the bulk RNA-seq of CD142– ASPCs that are exposed (*via* transwell (T-well)) to distinct types of CD142+ ASPCs (knockdown and control), together with CD142+ ASPCs (Aregs) and CD142– ASPCs post-differentiation samples;
- (E) Correlation of the normalised gene expression of specific CD142+ ASPC (Areg) candidates *versus* the “white fat cell differentiation score”.



Appendix Figure S23. Retinoic acid-related terms are enriched in CD142+ ASPCs (Aregs) compared to their negative counterparts, or in CD142- ASPCs when exposed to “active” Aregs’ signalling

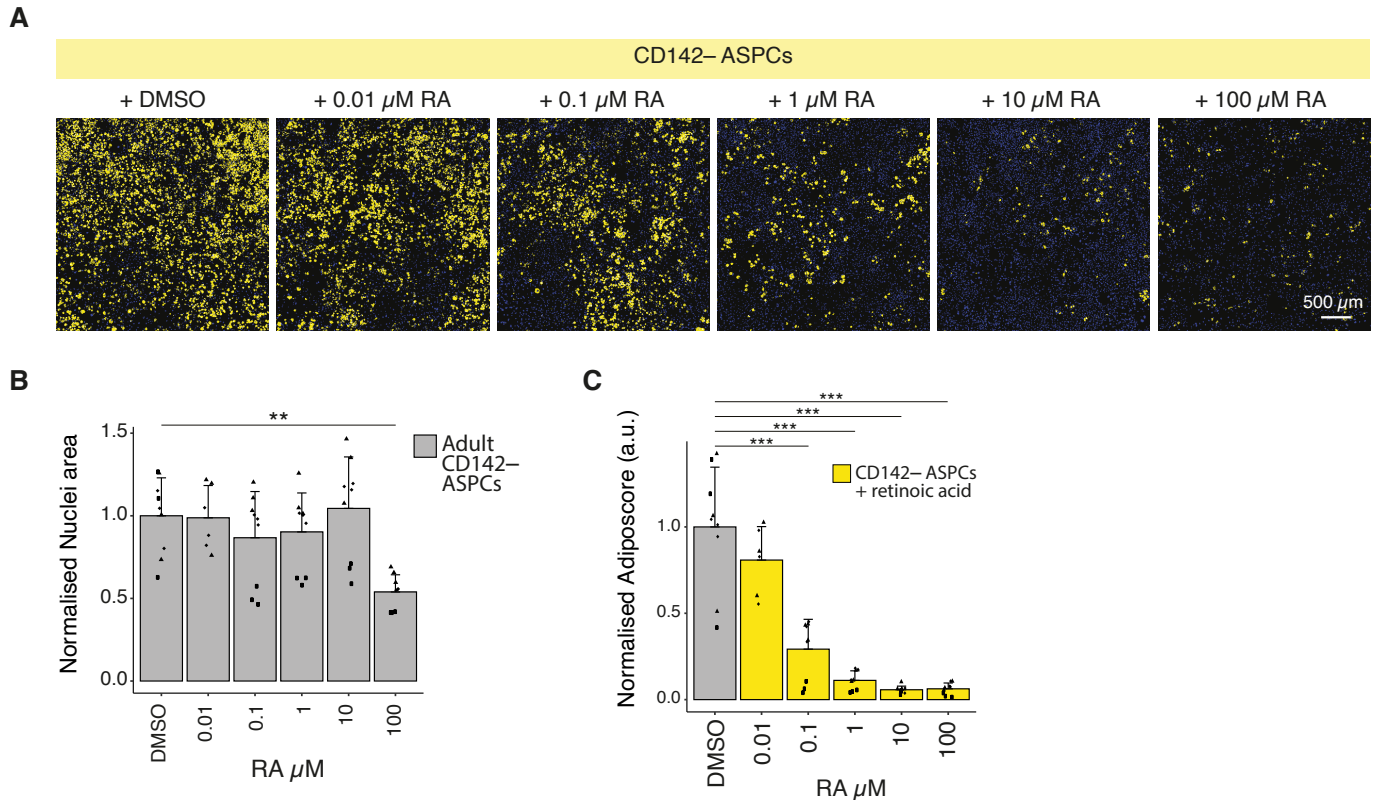
- (A) Bulk RNA-seq-derived expression plots of *Osr1* (Odd-skipped related transcription factor) and *Epha3* (Ephrin type-A receptor 3) in freshly isolated total, CD142- and CD142+ ASPCs;
- (B) The term “retinol metabolic process” (GO:0042572) was identified as significantly enriched in freshly isolated CD142+ *versus* CD142- ASPCs by GSEA when performed on differential expression data based on bulk RNA-seq of freshly sorted ASPCs;
- (C) The term “retinol metabolic process” (GO:0042572) was identified as significantly enriched in freshly isolated CD142+ *versus* CD142- ASPCs when performed on differential expression data based on mass spectrometry of freshly sorted ASPCs;
- (D) The term “cellular response to retinoic acid (RA)” (GO:0071300) was identified as significantly enriched in CD142+ *versus* CD142- ASPCs after adipogenic differentiation when performed on differential expression data based on bulk RNA-seq of ASPCs after adipogenic differentiation;
- (E) The term “RA receptor signalling pathway” (GO:0048384) was identified as significantly enriched in CD142+ *versus* CD142- ASPCs after adipogenic differentiation when performed on differential expression data based on bulk RNA-seq of ASPCs after adipogenic differentiation.

* $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, one-way ANOVA and Tukey HSD *post hoc* test (A), for statistical details see **Methods**.



Appendix Figure S24. Expression of genes linked to RA-associated terms in CD142– and CD142+ ASPCs (Aregs) post-differentiation and in CD142– ASPCs co-cultured with “active” and “dysfunctional” Aregs

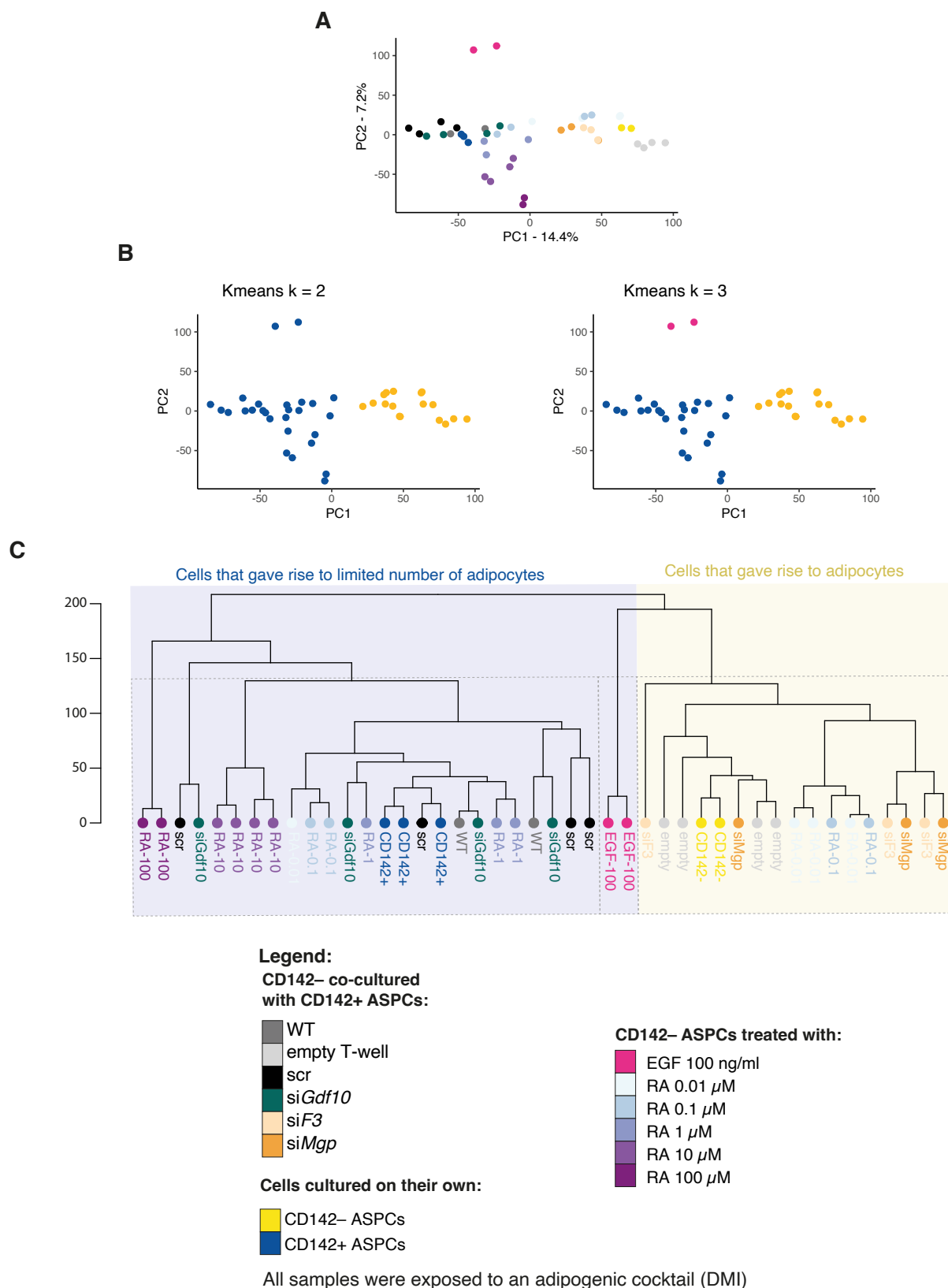
- (A) Expression heatmap of genes linked to “Cellular response to RA” (GO:0071300) and “RA receptor signalling pathway” (GO:0048384) across bulk RNA-seq samples of CD142– and CD142+ ASPCs (Aregs) post-differentiation; genes identified as lead by GSEA are indicated in red (**Methods**); log normalized expression scaled by row;
- (B) GSEA results of “cellular response to retinoic acid (RA)” (GO:0071300) performed on the genes driving the PC1 of the PCA shown in **Fig. 4C**. CD142– ASPCs were exposed to “dysfunctional” (positive NES) or “functional” (negative NES) Aregs;
- (C) GSEA results of “RA receptor signalling pathway” (GO:0048384) performed on the genes driving PC1 of the PCA shown in **Fig. 4C**. CD142– ASPCs were exposed to “dysfunctional” (positive NES) or “functional” (negative NES) Aregs.



Appendix Figure S25. Retinoic acid treatment inhibits CD142–ASPCs differentiation into adipocytes

- (A)** Representative fluorescence microscopy images of CD142–ASPCs after *in vitro* adipogenic differentiation with the differentiation cocktail supplemented with DMSO (RA carrier) and RA at the indicated concentrations; the presented images correspond to 25 tiled and thresholded 20x images containing 7-8 z-stacks in order to capture most of the well surface (**Methods**);
- (B)** Bar plots showing the nuclei numbers for the cellular fractions shown in **A**; marker shapes correspond to different biological replicates, n=9, 3 biological replicates, 3 independent wells for each;
- (C)** Fraction of differentiated adult-derived CD142–ASPCs, as quantified by the “adiposcore”, after *in vitro* adipogenic differentiation with the differentiation cocktail supplemented with DMSO (RA carrier) and RA at the indicated concentrations as shown in **A**; marker shapes correspond to different biological replicates, n=9, 3 biological replicates, 3 independent wells for each.

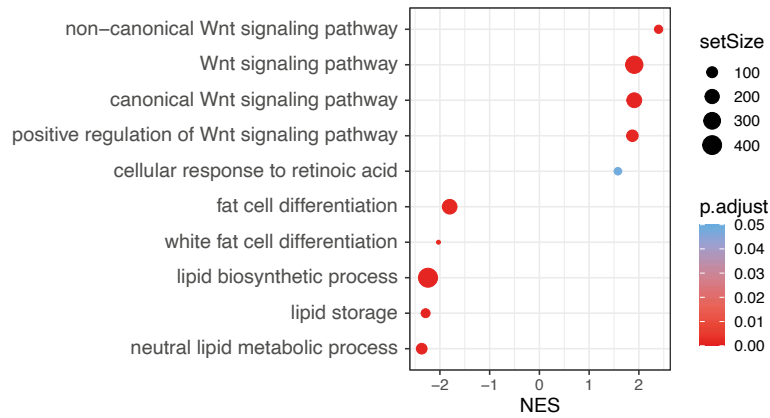
In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μ m; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



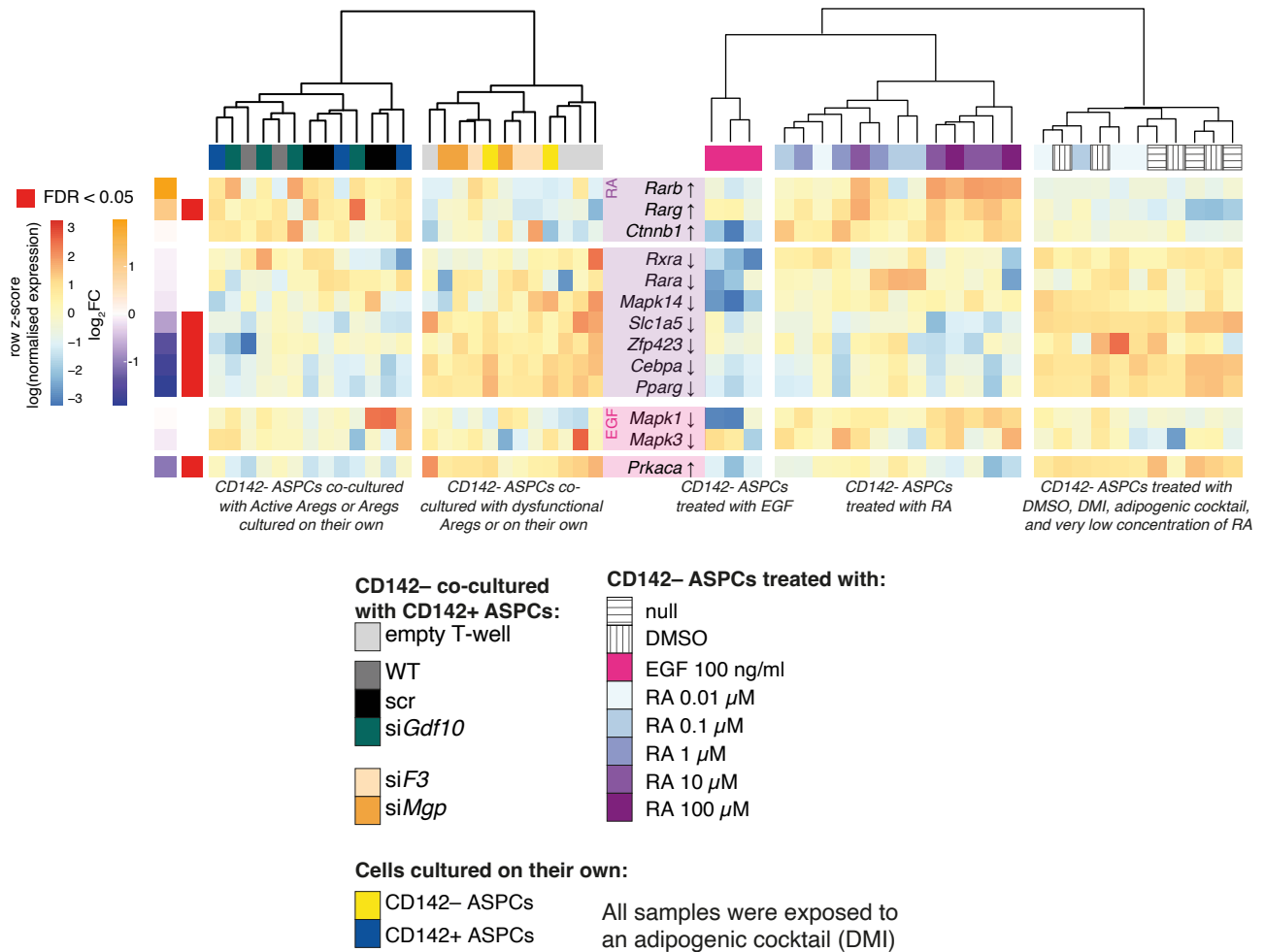
Appendix Figure S26. CD142- ASCs co-cultured with “active Aregs” CD142+ ASCs are transcriptomically similar to CD142- ASCs treated with RA

- (A)** PCA of the bulk RNA-seq of CD142- ASCs that were exposed (via transwell (T-well)) to the secretome of distinct CD142+ ASC (Areg) types (knockdown or control (scrambled (scr))) as listed in the colour legend, together with CD142+ (Aregs) and CD142- ASCs having been exposed to differentiation medium on their own as well as CD142- ASCs treated with EGF or different concentrations of RA;
- (B)** k-means clustering results performed on the first five principal components of the PCA shown in **A**, for $k = 2$ (left) or $k = 3$ (right);
- (C)** Hierarchical clustering tree based on bulk RNA-seq samples described in **A**.

A



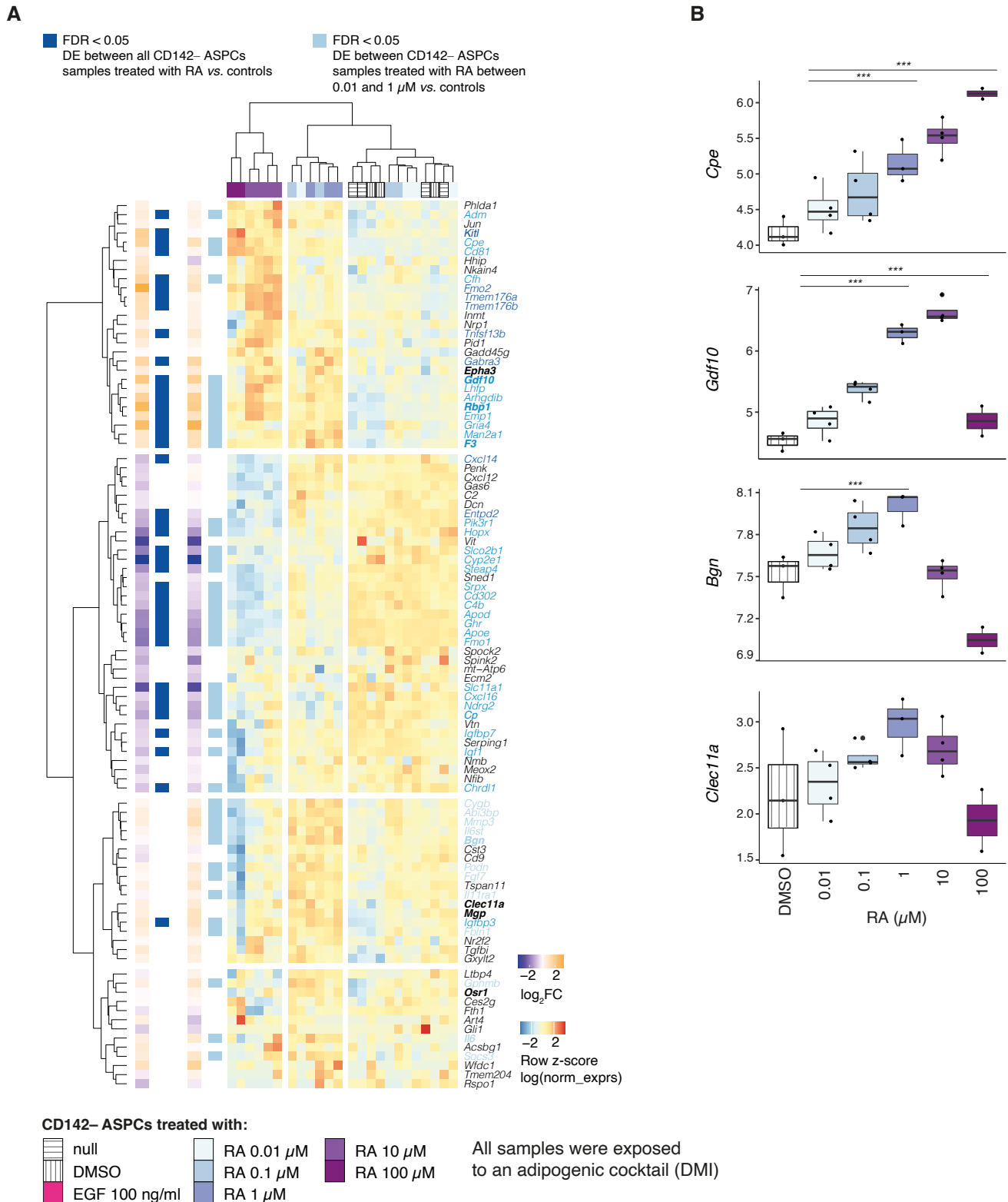
B



Appendix Figure S27. CD142- ASPCs exposed to the secretome of “active Aregs” (CD142+ ASPCs) exhibit upregulation of genes known to be involved in RA-mediated inhibition of adipogenesis

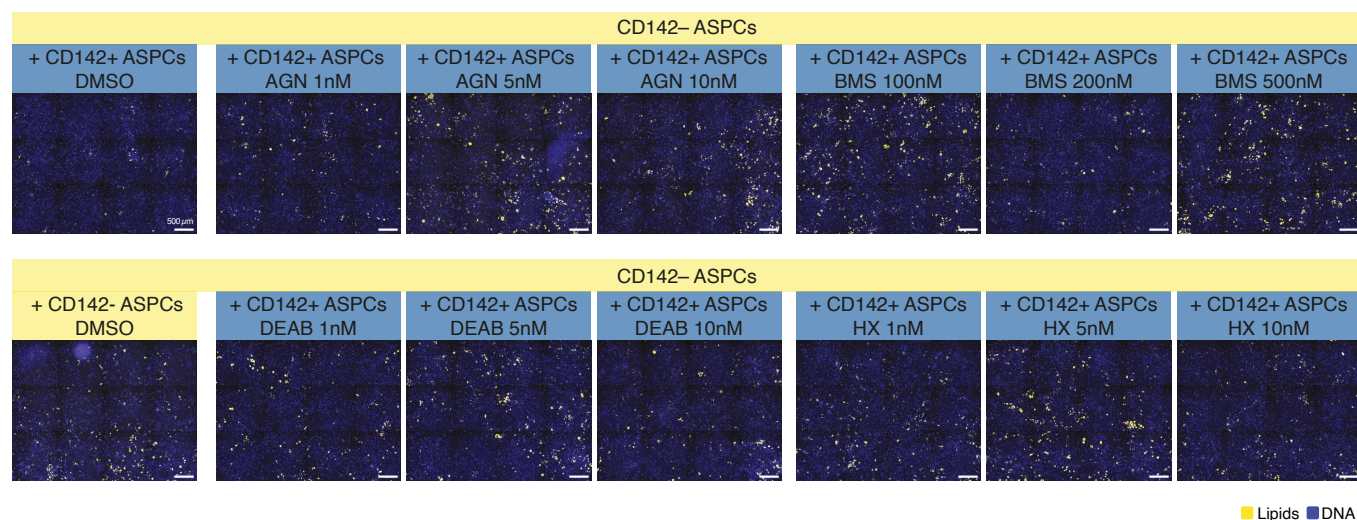
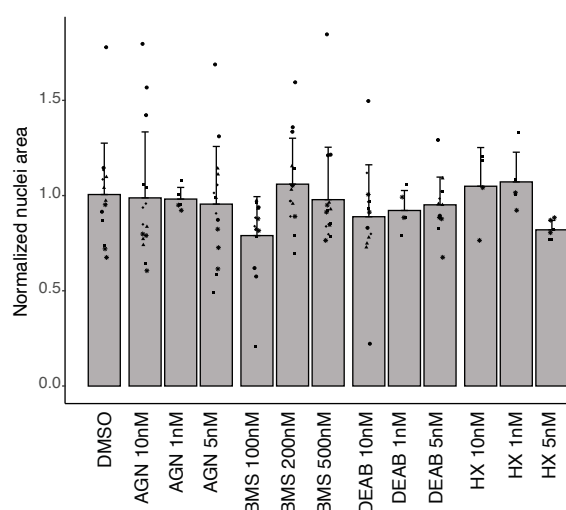
- (A) GSEA results of selected significant terms performed on the genes driving PC1 of the PCA shown in **Fig. 4C**;
- (B) Expression heatmap of genes identified as involved in the RA- or EGF-mediated inhibition of adipogenesis, based on manual curation of the literature. The arrow next to each gene indicates the reported expression response to either RA or EGF. The heatmap on the **left** shows the log normalized expression by row across bulk RNA-seq samples of CD142- ASPCs co-cultured (*via* transwell (T-well)) with “active Aregs” (wild-type (WT), scrambled (scr), si*Gdf10*) and standard CD142+ ASPCs (Aregs) post-differentiation (left cluster) and CD142- ASPCs co-cultured with “dysfunctional Aregs” (si*F3*, si*Mgp*) and CD142- ASPCs post-differentiation (right cluster); the heatmap on the **right** shows the normalized expression by row across bulk RNA-seq samples of CD142- ASPCs treated with EGF (left cluster), with RA (middle cluster) or with DMI with or without DMSO (RA carrier) (right cluster); for the two gene categories (involved in RA (purple) or EGF (pink)), the genes are ordered from top to bottom by the log₂FC of CD142- ASPCs co-cultured with “active Aregs” over CD142- ASPCs co-cultured with “dysfunctional Aregs”; significantly differentially expressed genes (FDR < 0.05) are indicated by the red vertical bar.

*P ≤ 0.05, **P ≤ 0.01, ***P ≤ 0.001 one-way ANOVA and Tukey HSD *post hoc* test (**B**), for statistical details, see **Methods**.



Appendix Figure S28. Key CD142+ ASPC (Areg) markers are upregulated in CD142- ASPCs treated with RA

- (A)** Expression heatmap of the top CD142+ ASPC (Areg) markers (**Appendix Table S1**) across bulk RNA-seq of CD142- ASPCs exposed to adipogenic cocktail (null) together with DMSO (RA carrier), EGF or different RA concentrations. The $\log_2$ FC and significance are based on the differential expression analysis between (**left**, dark blue) CD142- ASPCs treated with all RA concentrations or (**right**, light blue) treated with RA concentrations ranging between 0.01 and 1 μ M *versus* controls (treatment with DMI with or without DMSO);
- (B)** Boxplots showing the distribution of the log normalized expression (y axis) of *Cpe*, *Gdf10*, *Bgn*, and *Clec11a* across transcriptomic data from CD142- ASPCs treated with DMSO or different RA concentrations (0.01 to 100 μ M); the indicated significance is based on the differential expression analysis result between all RA treated samples or samples treated with RA at a concentration between 0.01 and 1 μ M *versus* controls (treatment with DMI with or without DMSO).

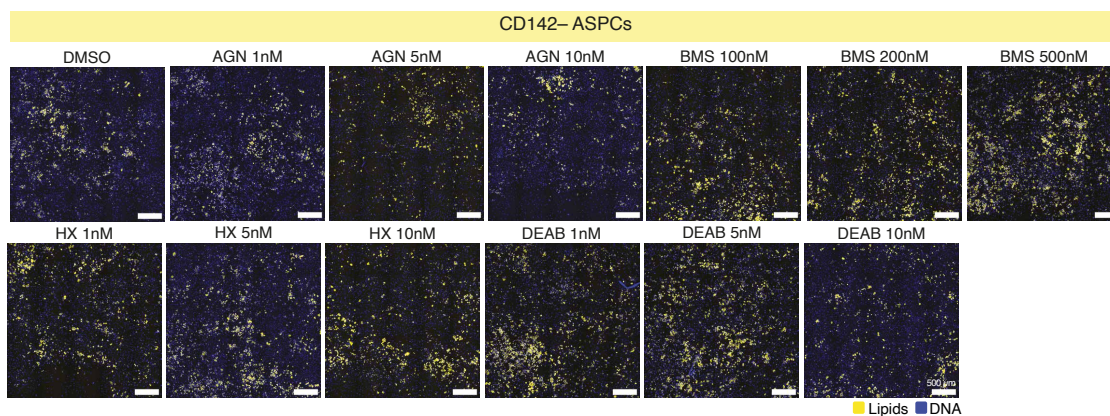
A**B**

Appendix Figure S29. Treatment with RA antagonists dampens the anti-adipogenic effect of Aregs on CD142- ASPCs

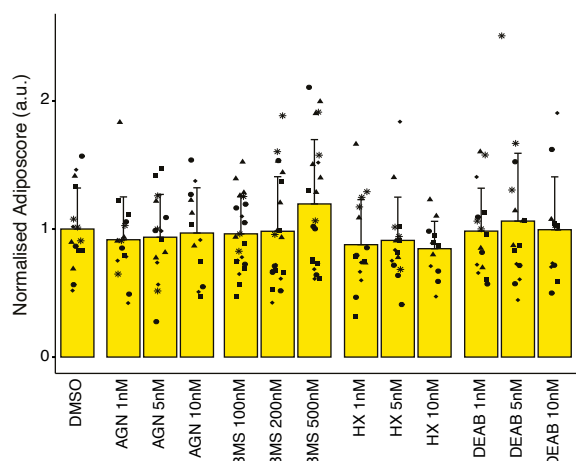
- (A)** Representative fluorescence microscopy images of CD142- ASPCs co-cultured with CD142+ ASPCs (or CD142- ASPCs with DMSO as a representative control) treated with the indicated RA antagonists at the indicated concentration after *in vitro* adipogenic differentiation; the RA antagonists were added to the top wells (containing the CD142+ ASPCs) of the transwell set-up;
- (B)** Bar plots showing the nuclei numbers for the cellular fractions shown in **A**; marker shapes correspond to different biological replicates, n=4-15, 2-5 biological replicates, 2-3 independent wells for each.

In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μ m; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.

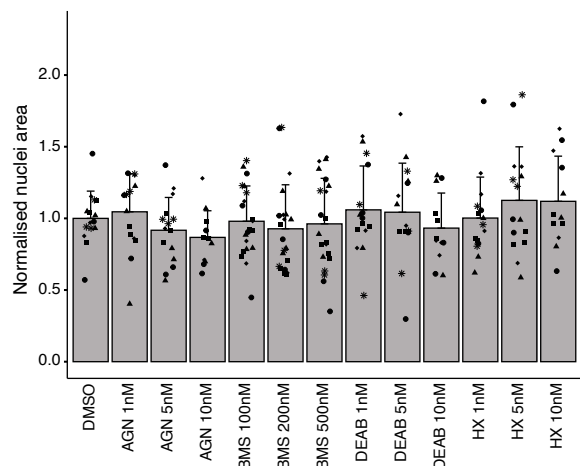
A



B



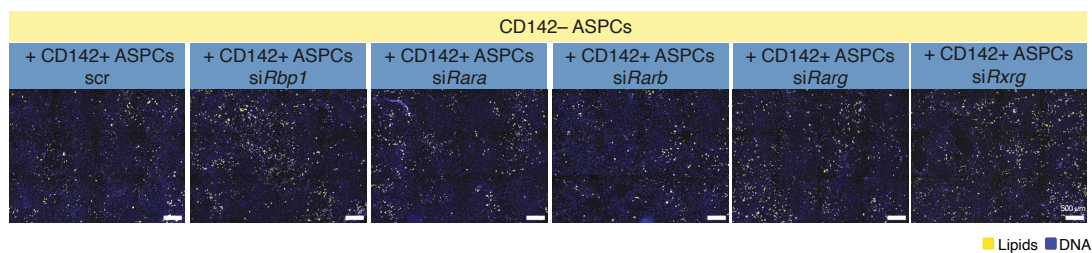
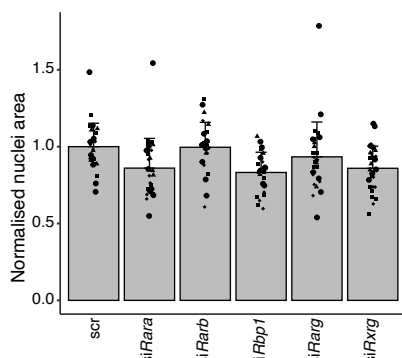
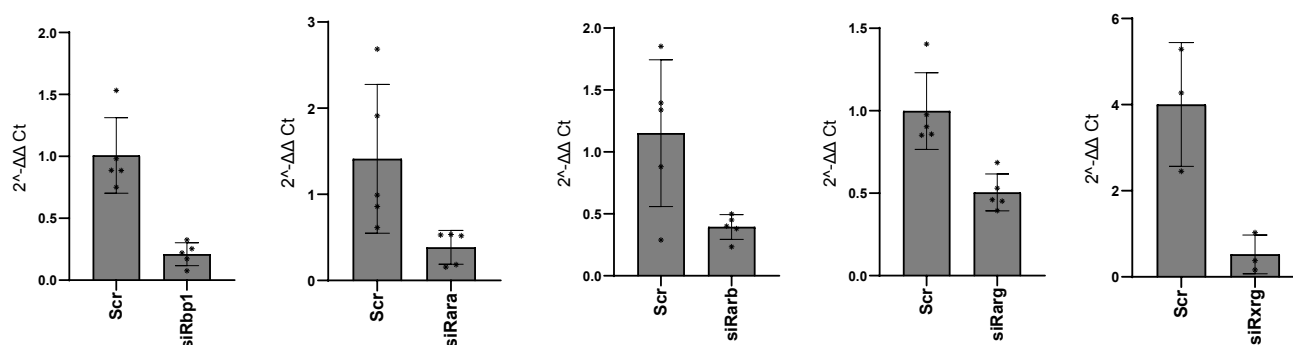
C



Appendix Figure S30. RA antagonists have negligible impact on CD142- ASPC differentiation

- (A) Representative fluorescence microscopy images of CD142- ASPCs after *in vitro* adipogenic differentiation; the induction cocktail was supplemented with recombinant proteins of RA antagonists at the indicated concentrations;
- (B) Bar plots showing the fraction of differentiated adult mouse-derived CD142- ASPCs, as quantified by the “adiposcore”, treated with the indicated RA antagonists at concentrations shown in **A**; marker shapes correspond to different biological replicates, $n=12-21$, 4-5 biological replicates, 3 independent wells for each;
- (C) Bar plots showing the normalized nuclei area for the cellular fractions shown in **A**; marker shapes correspond to different biological replicates, $n=12-21$, 4-5 biological replicates, 3 independent wells for each.

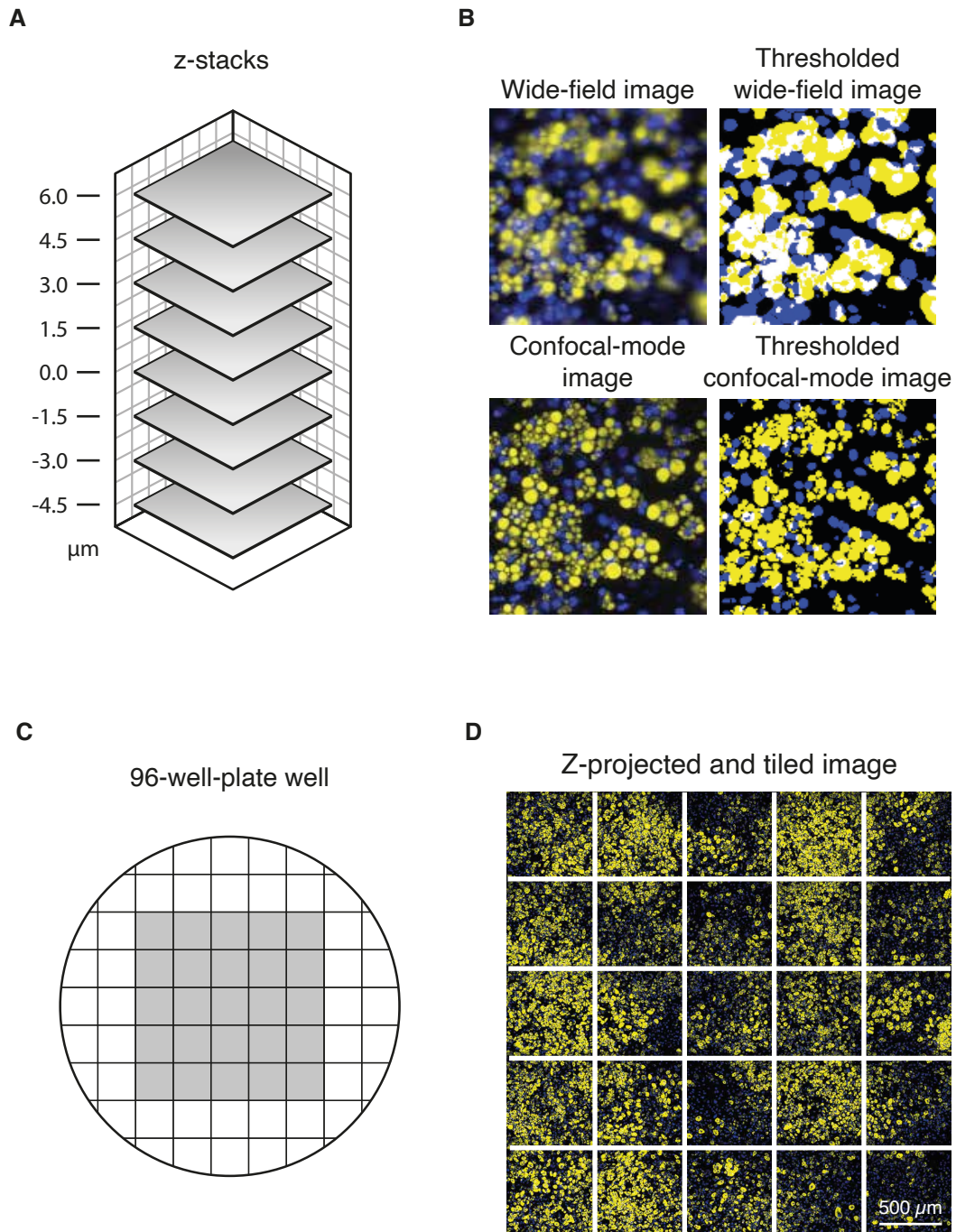
In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μ m; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.

A**B****C**

Appendix Figure S31. siRNA mediated knockdown of RA-related genes in Aregs dampens their anti-adipogenic effect on CD142- ASPCs

- (A) Representative fluorescence microscopy images of CD142- ASPCs co-cultured with CD142+ ASPCs subjected to control siRNA (scrambled (scr)) or siRNA-mediated knockdown of the indicated genes after *in vitro* adipogenic differentiation;
- (B) Bar plots showing the nuclei numbers for the cellular fractions shown in **A**; marker shapes correspond to different biological replicates, n=24-29, 4 biological replicates, 5-8 independent wells for each;
- (C) qPCR-measured gene expression of *Rbp1*, *Rara*, *Rarb*, *Rarg* and *Rxrg* in CD142+ ASPCs (Aregs) subjected to control (scrambled (scr)) or siRNA-mediated knockdown of the indicated genes normalised to the house-keeping gene *Hprt1* expression; 3-5 biological replicates.

In all images, nuclei are stained with Hoechst (blue) and lipids are stained with Bodipy (yellow); scale bars, 500 μ m; * $P \leq 0.05$, ** $P \leq 0.01$, *** $P \leq 0.001$, pairwise two-sided *t*-test, for statistical details see **Methods**.



Appendix Figure S32. Image-based quantification *in vitro* ASPC adipogenesis

- (A)** Schematic representation of z-stacks acquired with the Perkin Elmer Operetta platform upon imaging wells that contain terminally differentiated *in vitro* adipocytes;
- (B)** Representative images resulting from wide-field imaging (**top**) and confocal-mode imaging (**bottom**) and the respective thresholding result;
- (C)** Schematic representation of the 25 images acquired within a 96-well-plate well;
- (D)** A representative image resulting from confocal-mode imaging after tiling 25 acquired and z-projected images (the white grid has been added graphically for indicative purposes).

Appendix Table S1. Top 100 genes specific for CD142+ ASPCs (Aregs) issued from an integration of scRNA-seq (publicly available) and bulk RNA-seq (publicly available and generated in the presented study) datasets.

Aregs 100 Top Markers (1/3)			
gene ID	Single-cell RNA-seq avg logFC	Bulk RNA-seq log₂FC	Mass spectrometry log₂FC
<i>Gas6</i>	1.734934037	2.648	0.573588699
<i>Cxcl12</i>	1.695784366	1.306	NA
<i>Inmt</i>	1.69273622	2.106	2.232207245
<i>Apoe</i>	1.617141956	NA	0.523858909
<i>Gdf10</i>	1.485475157	2.51	1.002882979
<i>Clec11a</i>	1.339330278	3.383	NA
<i>F3</i>	1.306446139	4.364	3.852178637
<i>Mgp</i>	1.211678039	4.412	1.518067937
<i>C2</i>	1.197130218	2.295	0.870029683
<i>Fmo2</i>	1.16670468	4.944	3.451288912
<i>Igfbp7</i>	1.111835725	NA	0.929751786
<i>Igfbp3</i>	1.080026504	NA	NA
<i>Vit</i>	1.016376833	3.256	NA
<i>Bgn</i>	0.991099847	1.424	2.471041469
<i>Steap4</i>	0.986622284	1.661	3.100726969
<i>Gadd45g</i>	0.980075579	NA	NA
<i>Tmem176b</i>	0.972756487	NA	NA
<i>Igf1</i>	0.96280102	NA	NA
<i>Cygb</i>	0.931309354	1.784	2.216748912
<i>Tmem176a</i>	0.908769033	NA	NA
<i>Chrdl1</i>	0.892908836	2.42	NA
<i>Srpx</i>	0.890644606	NA	NA
<i>Nmb</i>	0.883789886	1.416	NA
<i>Abi3bp</i>	0.848341847	1.031	-0.059743746
<i>Penk</i>	0.826923908	NA	NA
<i>Cp</i>	0.825599904	NA	2.109079548
<i>Nrp1</i>	0.8204081	1.223	0.247841773
<i>Vtn</i>	0.795788451	NA	NA
<i>Cxcl14</i>	0.783008446	NA	0.62827134
<i>Cst3</i>	0.77369243	1.874	1.244114617
<i>Mmp3</i>	0.738574153	NA	NA
<i>Il11ra1</i>	0.738383419	NA	NA
<i>Cd9</i>	0.735687305	1.422	NA
<i>C4b</i>	0.731043887	-1.175	-0.331515068
<i>Tgfb1</i>	0.729048091	2.618	NA

Aregs 100 Top Markers (2/3)			
gene ID	Single-cell RNA-seq avg logFC	Bulk RNA-seq log ₂ FC	Mass spectrometry log ₂ FC
<i>Cfh</i>	0.722718972	NA	0.547533656
<i>Socs3</i>	0.709242264	NA	NA
<i>Meox2</i>	0.678224608	3.734	1.593829126
<i>Il6</i>	0.66412741	1.652	NA
<i>Sepp1</i>	0.640493416	1.739	NA
<i>Cpe</i>	0.625167315	2.179	NA
<i>Osr1</i>	0.612115059	1.456	0.199888736
<i>Rbp1</i>	0.610362899	1.805	1.669933451
<i>Gpnmb</i>	0.602833659	1.878	2.115085125
<i>Nfib</i>	0.57788856	NA	0.844491716
<i>Apod</i>	0.575275481	NA	0.685823996
<i>Pid1</i>	0.563071453	2.35	0.313006138
<i>Phlda1</i>	0.562879145	NA	0.036171197
<i>Serping1</i>	0.560522695	NA	0.341085822
<i>Sned1</i>	0.554291693	NA	NA
<i>Fbln1</i>	0.533888358	1.53	NA
<i>Ltbp4</i>	0.518973443	1.06	NA
<i>Ndrp2</i>	0.514044932	3.057	1.805160555
<i>Ecm2</i>	0.484665409	NA	NA
<i>Entpd2</i>	0.483039679	1.163	2.834169075
<i>Podn</i>	0.482052268	1.259	NA
<i>Lhfp</i>	0.478587415	NA	NA
<i>Nr2f2</i>	0.474489959	1.723	0.88969045
<i>Dcn</i>	0.474343194	NA	0.410972407
<i>Fth1</i>	0.473941488	NA	0.444517095
<i>Kitl</i>	0.440142536	1.596	NA
<i>Ghr</i>	0.432869152	1.169	NA
<i>Cd302</i>	0.428252464	NA	NA
<i>Ces2g</i>	0.424280683	NA	NA
<i>Slco2b1</i>	0.410231034	1.509	0.823355932
<i>Al607873</i>	0.400312532	NA	NA
<i>Arhgdib</i>	0.393423297	4.014	2.192112505
<i>Fgf7</i>	0.384580273	1.086	NA
<i>Rspo1</i>	0.383282553	NA	NA
<i>Il6st</i>	0.383014607	1.062	0.77442495
<i>Gxylt2</i>	0.371476281	NA	NA
<i>mt-Atp6</i>	0.36908521	NA	NA
<i>Cd81</i>	0.367270304	NA	0.466254044
<i>Jun</i>	0.365838934	NA	-0.309885662

Aregs 100 Top Markers (3/3)			
gene ID	Single-cell RNA-seq avg logFC	Bulk RNA-seq log ₂ FC	Mass spectrometry log ₂ FC
<i>Tspan11</i>	0.35906473	4.391	NA
<i>Emp1</i>	0.357989155	NA	NA
<i>Man2a1</i>	0.343316508	NA	-0.163599593
<i>Adm</i>	0.339422608	NA	NA
<i>Cxcl16</i>	0.324837373	NA	NA
<i>Epha3</i>	0.312033992	2.171	1.239960604
<i>Fmo1</i>	0.304309182	1.679	1.319270289
<i>Tnfsf13b</i>	0.301936673	2.169	NA
<i>Gabra3</i>	0.299116342	1.848	0.488352387
<i>Spock2</i>	0.289710678	2.589	NA
<i>Pik3r1</i>	0.265155293	NA	0.003332054
<i>Acsbg1</i>	NA	6.015	0.926057088
<i>Spink2</i>	NA	5.451	NA
<i>Art4</i>	NA	4.883	NA
<i>Hopx</i>	NA	4.794	2.102595259
<i>Tmem204</i>	NA	4.763	NA
<i>Nkain4</i>	NA	4.643	NA
<i>Cldn22</i>	NA	4.32	NA
<i>Wfdc1</i>	NA	4.261	NA
<i>Hhip</i>	NA	4.214	NA
<i>Gria4</i>	NA	4.208	NA
<i>Cyp2e1</i>	NA	4.023	NA
<i>Slc11a1</i>	NA	4.015	NA
<i>lyd</i>	NA	3.983	NA
<i>Gli1</i>	NA	3.964	NA
<i>Cav3</i>	NA	3.921	NA

Appendix Table S2. Summary of bulk RNA-seq and proteomic experiments conducted in the presented study.

TRANSCRIPTOMICS						
Name	Fresh ASPCs	Expanded ASPCs	Post-diff ASPCs	RA & EGF treatment	KD assay	Age Assay
Design	Sequenced were freshly purified murine ASPC cells isolated by FACS using four different anti-CD142 antibodies (Methods)	Sequenced were FACS-isolated murine CD142+ and CD142- ASPCs plated and expanded in culture for 4-5 days	Sequenced were FACS-isolated (with four different anti-CD142 antibodies) murine CD142+ and CD142- ASPCs plated, cultured to confluence and differentiated into adipocytes	Sequenced were CD142- ASPCs differentiated into adipocytes with retinoic acid (RA) or recombinant EGF treatment	Sequenced were CD142- APSCs differentiated into adipocytes in the presence of wild-type CD142+ ASPCs or CD142+ ASPCs with indicated KDs or an empty well	Sequenced were total, CD142- and CD142+ ASPCs isolated from P0, P16, 4wo, 7wo and 11wo mice
Samples	CD142+ CD142- SCA-1+	CD142+ CD142-	CD142+ CD142- SCA-1+	CD142- ASPCs treated with: -0.01 -0.1 -1 -10 -100 μ M of RA -100 ng/ μ L of EGF	CD142- ASPCs co-cultured with CD142+ ASPCs: -scr -si <i>Mgp</i> -si <i>Aldh1a2</i> -si <i>Gdf10</i> -si <i>F3</i> -WT - with an empty well	CD142+ CD142- SCA-1+

PROTEOMICS		
Name	Fresh ASPCs	Expanded ASPCs
samples	CD142+ CD142-	CD142+ CD142-

Appendix Table S3. Sex, age and BMI of all human participants in the present study.

Human research participants		
Sex	Age (years)	BMI (kg/m ²)
Male	75	26
Female	43	32
Male	37	40

Appendix Table S4. List of all antibodies employed in the experiments conducted in the presented study.

Primary antibodies											
Antibody	Clone	Clonality	Host	Reactivity	Conjugate	Application	Company	Ref#	Tested applications	Publication DOI	Dilution
anti-CD31	MEC13.3	monoclonal	Rat	Mouse	AF488	FACS	BioLegend	102514	FC	10.1038/ncomms11302	1:500
anti-CD45	30-F11	monoclonal	Rat	Mouse	AF488	FACS	BioLegend	103122	FC	10.1038/ncomms11533	1:500
anti-CD142	001	monoclonal	Rabbit	Mouse	PE	FACS	SinoBiological	50413-R001-P	FC	10.1038/s41586-018-0226-8	1:50
anti-CD142	001	monoclonal	Rabbit	Mouse	Un-conjugated	FACS	SinoBiological	50413-R001	FC	10.1126/sciadv.aax9605	1:50
anti-CD142	HTF-1	monoclonal	Mouse	Mouse	PE	FACS	BiOrbyt	ORB507485	FC	N/A	1:20
anti-CD142	-	polyclonal	Goat	Mouse	PE	FACS	R&D Systems	FAB3178P	FC	10.3324/haematol.2019.220210	1:20
anti-SCA-1	E13-161.7	monoclonal	Rat	Mouse	PE-Cy7	FACS	BioLegend	122514	FC	10.1371/journal.pbio.1002562	1:500
anti-TER119	TER-119	monoclonal	Rat	Mouse	AF488	FACS	BioLegend	116215	FC	10.1016/j.bbailip.2016.06.010	1:500

Appendix Table S5. Sequences of siRNA probes employed for Areg-related knock down experiments conducted in the presented study.

siRNA name	Sequence
Scrambled	5' /5Phos/rCrUrUrCrCrUrCrUrCrUrUrUrCrUrCrUrCrCrUrUrGrUGA 3' + 5' rUrCrArCrArArGrGrGrArGrArGrArArArGrArGrArGrArArGrGrA 3' -
mm.Ri.F3.13.1	5' rGrGrArUrArUrCrArArCrUrGrArUrUrUrCrArArGrArCrAAT 3' + 5' rArUrUrGrUrCrUrUrGrArArArUrCrArGrUrUrGrArUrArUrCrCrArA 3' -
mm.Ri.F3.13.2	5' rGrUrArArArArGrArCrUrCrArCrUrUrArCrArUrUrArGrUCA 3' + 5' rUrGrArCrUrArArUrGrUrArArGrUrGrArGrUrCrUrUrUrUrArCrArA 3' -
mm.Ri.F3.13.3	5' rArUrGrUrUrArArArUrGrCrArGrGrArUrArUrUrUrCrUrGCT 3' + 5' rArGrCrArGrArArArUrArUrCrCrUrGrCrArUrUrUrArArCrArUrGrU 3' -
mm.Ri.Mgp.13.1	5' rCrUrCrCrUrGrCrArGrUrArGrCrArUrUrArCrUrGrArArGTA 3' + 5' rUrArCrUrUrCrArGrUrArArUrGrCrUrArCrUrGrCrArGrGrArGrArU 3' -
mm.Ri.Mgp.13.2	5' rCrCrUrArUrGrArArArUrCrArGrUrCrCrCrUrUrCrArUrCAA 3' + 5' rUrUrGrArUrGrArArGrGrGrArCrUrGrArUrUrUrCrArUrArGrGrArC 3' -
mm.Ri.Mgp.13.3	5' rGrGrCrArArCrCrCrUrGrUrGrCrUrArCrGrArArUrCrUrCAC 3' + 5' rGrUrGrArGrArUrUrCrGrUrArGrCrArCrArGrGrGrUrUrGrCrCrArC 3' -
mm.Ri.Gdf10.13.1	5' rGrArGrUrArUrUrUrUrCrUrCrUrUrArCrGrUrUrUrCrArCTA 3' + 5' rUrArGrUrGrArArArCrGrUrArArGrArGrArArArUrArCrUrCrArC 3' -
mm.Ri.Gdf10.13.2	5' rUrArArGrArUrGrGrCrUrUrCrArArGrArUrArGrArArGrACA 3' + 5' rUrGrUrCrUrUrCrUrArUrCrUrUrGrArArGrCrCrArUrCrUrUrArCrC 3' -
mm.Ri.Gdf10.13.3	5' rCrGrGrGrUrGrGrArArUrGrArArUrGrGrArUrCrArUrCrUCT 3' + 5' rArGrArGrArUrGrArUrCrCrArUrUrCrArUrUrCrCrArCrCrGrArU 3' -
mm.Ri.Aldh1a2.13.1	5' rGrArGrGrUrUrGrGrArArArGrCrUrUrArUrUrCrArArGrAAG 3' + 5' rCrUrUrCrUrUrGrArArUrArArGrCrUrUrUrCrCrArArCrCrUrCrArG 3' -
mm.Ri.Aldh1a2.13.2	5' rCrArArGrArArArUrUrUrUrGrArGrGrUrUrUrArArGrArCTA 3' + 5' rUrArGrUrCrUrUrArArArCrCrUrCrArArArArUrUrUrCrUrUrGrArA 3' -
mm.Ri.Aldh1a2.13.3	5' rGrGrUrGrArArUrArCrCrGrUrUrUrArUrUrUrArArArArATG 3' + 5' rCrArUrUrUrUrArArArUrArArArCrGrGrUrArUrUrCrArCrCrCrA 3' -