

Supplemental material

Cardiovascular risk factors and differential transcriptomic profile of the subcutaneous and visceral adipose tissue and their resident stem cells

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Supplemental Table S1: TaqMan gene expression assay probes used in the custom array.

| Gene symbol | Gene name | TaqMan Assay reference | Localization |
|--------------------|--|-------------------------------|--------------------------------|
| AGER | Advanced glycosilation end product-specific receptor | Hs00153957_m1 | Plasma membrane |
| ANGPT1 | Angiopoietin 1 | Hs00181613_m1 | Extracellular |
| ANGPT2 | Angiopoietin 2 | Hs00169867_m1 | Extracellular |
| CAPG | Macrophage-capping protein | Hs00156249_m1 | Cytoplasm, Nucleus, Secreted |
| CD34 | Hematopoietic progenitor dell antigen | Hs00156373_m1 | Plasma membrane |
| CD68 | Macrosialin | Hs00154355_m1 | Plasma membrane |
| COL18A1 | Collagen alpha-1(XVIII) chain | Hs00181017_m1 | Extracellular |
| DLK1 | Protein delta homolog 1 | Hs00171584_m1 | Plasma membrane |
| DLL4 | Delta like protein-4 | Hs00184092_m1 | Plasma membrane |
| ENG | Endoglin | Hs00923996_m1 | Plasma membrane |
| IFI30 | Gamma-intertefon-inducible lysosomal thiol reductase | Hs00173838_m1 | Lysosome, extracellulas region |
| NOTCH3 | Neurogenic locus notch homolog protein3 | Hs01128541_m1 | Plasma membrane |
| PECAM1 | Platelet endothelial cell adhesion molecule | Hs00169777_m1 | Plasma membrane |
| PGF | Placenta growth factor | Hs00182176_m1 | Extracellular |
| PLIN2 | Perlpin2 (PLIN2) | Hs00605340_m1 | Membrane, lipid droplets |
| PNPLA2 | Patatin-like phospholipase domain-containing protein 2 | Hs00386101_m1 | Membrane, lipid droplets |
| PTX3 | Pentraxin-related protein PTX3 | Hs00173615_m1 | Extracellular |
| SERPINF1 | Pigment epithelium-derived factor | Hs01106937_m1 | Extracellular |
| SPP1 | Osteopontin | Hs00959010_m1 | Extracellular |
| TEK | Angiopoietin-1 receptor | Hs00945155_m1 | Plasma membrane, extracellular |
| TNC | Tenascin | Hs01115665_m1 | Extracellular |
| VEGFA | Vascular endothelial growth factor A | Hs00900055_m1 | Extracellular |

Supplemental Table S2: Antibodies used to perform the experiments.

| | | |
|--|----------------------|---------------|
| CD105 PE anti-Human | BD | 560839 |
| CD44 PE anti-Human | BD PHARMINGEN | 555479 |
| CD29 PE Mouse Anti-Human | BD | 555443 |
| CD90 FITC anti-Human | BD PHARMINGEN | 555595 |
| CD73 FITC anti-Human | BD PHARMINGEN | 561254 |
| CD45 PE-Cy5 anti-Human | BD PHARMINGEN | 555484 |
| CD14 FITC anti-Human | IMMUNOSTEP | 14F-100T |
| CFBlue Annexin V | IMMUNOSTEP | ANXVCFB |
| FITC-Conjugated Monoclonal Antibody Against Human Tissue Factor | CIENTISOL | 4508CJ |
| TF anti-goat | CIENTISOL | 4501 |
| anti beta-actin anti-mouse | ABCAM | ab8226 |
| VEGF Receptor2 (55B11) | CELL SIGNALING | 2479 |
| VEGFA | ABCAM | ab214424 |

Supplemental Table S3: Gene expression profile of the subcutaneous and visceral adipose tissue of patients and controls.

| | Obese | | Non-obese |
|-----------------|----------------------------------|----------------------------------|----------------------------------|
| | SAT (2 ^{-ΔCT})*1000 | VAT (2 ^{-ΔCT})*1000 | SAT (2 ^{-ΔCT})*1000 |
| AGER | 7.05 ± 0.64 | 8.30 ± 0.48 | 4.03 ± 0.53 |
| ANGPT1 | 89.58 ± 9.17 | 52.7 ± 5.16 | 68.47 ± 5.30 |
| ANGPT2 | 15.35 ± 1.93 | 8.89 ± 0.92 | 11.01 ± 1.01 |
| CAPG | 297.94 ± 24.63 | 254.41 ± 13.81 | 224.31 ± 16.15 |
| CD34 | 349.26 ± 28.80 | 320.7 ± 22.89 | 250.38 ± 27.62 |
| CD68 | 635.13 ± 41.73 | 515.71 ± 27.11 | 298.78 ± 49.20 |
| COL18A1 | 244.05 ± 17.61 | 220.96 ± 11.44 | 262.04 ± 17.95 |
| DLL4 | 36.47 ± 2.98 | 28.043 ± 2.52 | 59.22 ± 6.77 |
| ENG | 354.15 ± 31.40 | 295.37 ± 13.72 | 382.82 ± 30.05 |
| IFI30 | 288.80 ± 28.28 | 246.45 ± 22.96 | 113.32 ± 24.85 |
| NOTCH3 | 337.38 ± 36.22 | 84.68 ± 6.18 | 198.90 ± 18.99 |
| PECAM1 | 497.34 ± 49.34 | 327.96 ± 21.81 | 562.18 ± 71.80 |
| PGF | 6.82 ± 0.76 | 8.26 ± 0.48 | 6.67 ± 1.10 |
| PLIN2 | 297.58 ± 27.13 | 257.68 ± 14.46 | 442.55 ± 29.19 |
| PNPLA2 | 1084 ± 62.10 | 831.19 ± 70.16 | 1747.56 ± 11017 |
| PTX3 | 6.02 ± 2.37 | 5.44 ± 0.58 | 5.99 ± 1.39 |
| SERPINF1 | 2562.60 ± 170.68 | 1705.26 ± 126.53 | 2459.77 ± 184.53 |
| SPP1 | 238.56 ± 42.88 | 82.34 ± 12.96 | 50.08 ± 16.78 |
| TEK | 65.82 ± 4.17 | 53.02 ± 3.24 | 79.95 ± 8.54 |
| TNC | 38.79 ± 7.41 | 12.82 ± 1.11 | 34.56 ± 11.77 |
| VEGFA | 198.17 ± 20.14 | 231.32 ± 13.68 | 426.72 ± 54.49 |

Supplemental Table S4. Gene expression of SAT and VAT from non diabetic (non-DM) and diabetic (DM) obese patients. Significance between non-DM versus DM.

| Gene | SUBCUTANEOUS | | | VISCERAL | | |
|-----------------|-------------------------------------|---------------------------------|----------------------------|-------------------------------------|---------------------------------|----------------------------|
| | non-DM (2 ^{-ΔCT})*1000 | DM (2 ^{-ΔCT})*1000 | non-DM vs DM P-value | non-DM (2 ^{-ΔCT})*1000 | DM (2 ^{-ΔCT})*1000 | non-DM vs DM P value |
| AGER | 7.12 ± 0.74 | 6.80 ± 1.37 | 0.906 | 8.31 ± 0.51 | 8.26 ± 1.36 | 0.969 |
| ANGPT1 | 97.66 ± 10.79 | 62.12 ± 10.71 | 0.072 | 60.38 ± 6.46 | 46.66 ± 3.18 | 0.256 |
| ANGPT2 | 16.52 ± 2.31 | 11.36 ± 2.91 | 0.127 | 8.89 ± 1.18 | 8.88 ± 0.70 | 0.411 |
| CAPG | 292.25 ± 26.21 | 317.3067.46 | 0.724 | 250.95 ± 14.53 | 266.19 ± 38.60 | 0.368 |
| CD34 | 365.63 ± 32.28 | 293.59 ± 63.12 | 0.292 | 303.5 ± 21.35 | 379.19 ± 69.25 | 0.368 |
| CD68 | 625.10 ± 44.51 | 669.24 ± 113.62 | 0.505 | 504.91 ± 28.68 | 552.45 ± 73.19 | 0.724 |
| COL18A1 | 238.25 ± 17.62 | 263.75 ± 53.09 | 0.457 | 219.32 ± 12.09 | 226.53 ± 32.03 | 0.969 |
| DLL4 | 38.23 ± 3.44 | 30.49 ± 5.73 | 0.255 | 27.26 ± 3.05 | 30.70 ± 4.28 | 0.368 |
| ENG | 347.79 ± 36.59 | 375.8 ± 66.43 | 0.092 | 283.80 ± 15.45 | 334.73 ± 24.48 | 0.092 |
| IFI30 | 273.79 ± 26.54 | 339.84 ± 89.71 | 0.505 | 243.49 ± 26.54 | 256.51 ± 50.60 | 0.784 |
| NOTCH3 | 329.73 ± 44.53 | 363.38 ± 55.81 | 0.327 | 84.78 ± 7.92 | 84.34 ± 5.46 | 0.906 |
| PECAM1 | 500.98 ± 57.45 | 484.94 ± 106.32 | 0.906 | 320.66 ± 26.01 | 352.78 ± 39.80 | 0.365 |
| PGF | 6.62 ± 0.72 | 7.51 ± 2.49 | 0.906 | 8.10 ± 0.54 | 8.80 ± 1.13 | 0.457 |
| PLIN2 | 280.11 ± 25.91 | 356.97 ± 82.03 | 0.457 | 247.77 ± 13.05 | 291.36 ± 46.30 | 0.417 |
| PNPLA2 | 1120.28 ± 54.25 | 964.90 ± 210.66 | 0.557 | 867.77 ± 86.72 | 706.82 ± 80.22 | 0.457 |
| PTX3 | 6.47 ± 3.05 | 4.50 ± 1.78 | 0.611 | 5.32 ± 0.71 | 5.86 ± 0.88 | 0.411 |
| SERPINF1 | 2715.24 ± 196.51 | 2043.64 ± 246.24 | 0.092 | 1782.30 ± 158.90 | 1443.35 ± 65.09 | 0.368 |
| SPP1 | 195.23 ± 31.68 | 385.85 ± 147.71 | 0.290 | 74.20 ± 15.87 | 110.02 ± 14.47 | 0.055 |
| TEK | 64.84 ± 4.79 | 69.15 ± 14.05 | 0.611 | 51.69 ± 4.09 | 57.54 ± 2.86 | 0.365 |
| TNC | 38.66 ± 9.10 | 39.25 ± 11.94 | 0.784 | 11.84 ± 1.08 | 16.14 ± 3.01 | 0.256 |
| VEGFA | 205.44 ± 23.19 | 173.46 ± 43.11 | 0.667 | 222.89 ± 14.09 | 259.96 ± 36.80 | 0.290 |

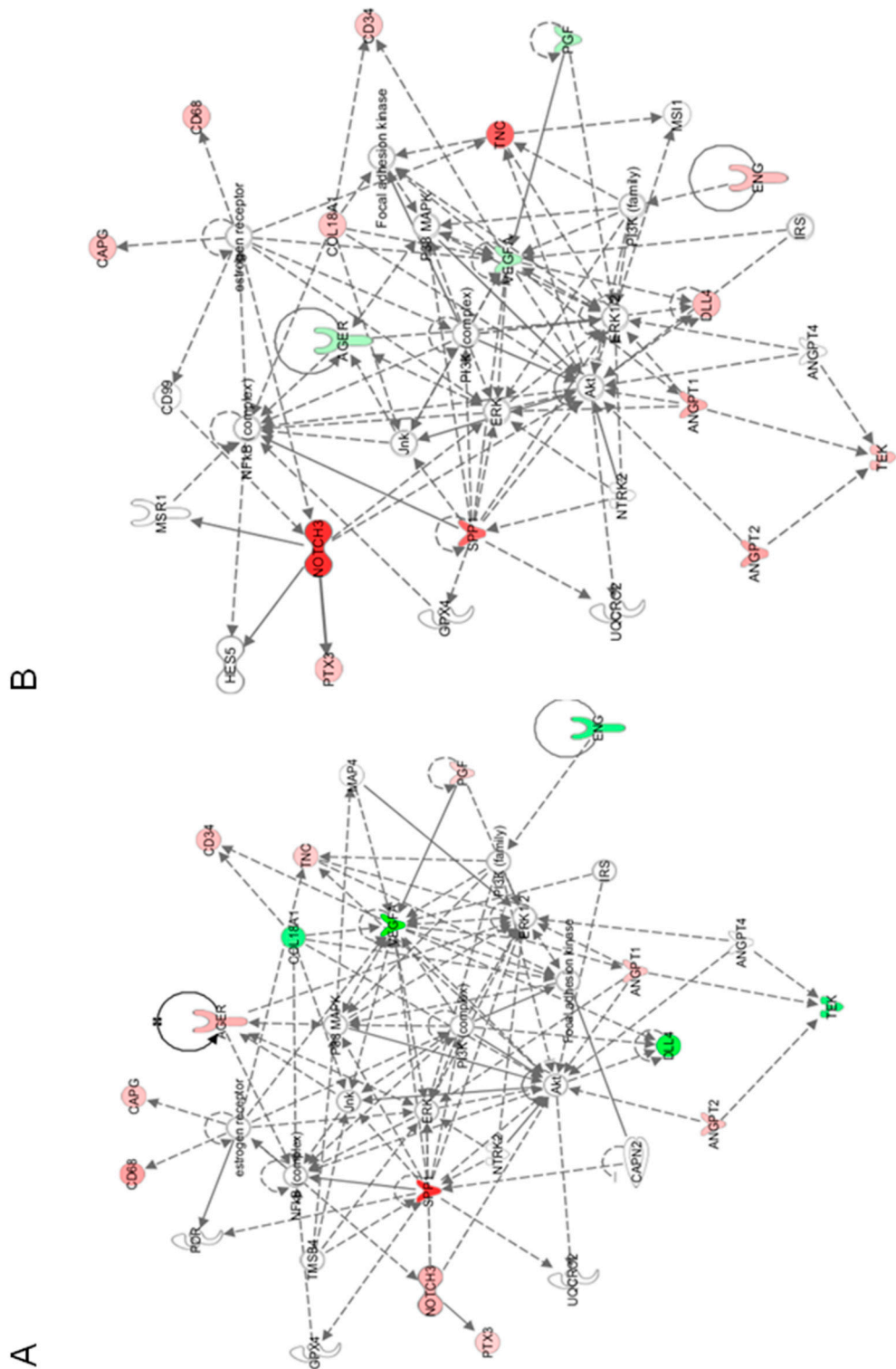
Supplemental Table S5. Gene expression of SAT and VAT from non dyslipemic (non-DLP) and dyslipemic (DLP) obese patients. Significance between non-DLP versus DLP.

| Gene | SUBCUTANEOUS | | | VISCERAL | | |
|----------|--------------------------------------|----------------------------------|------------------------------|--------------------------------------|----------------------------------|------------------------------|
| | non-DLP (2 ^{-ΔCT})*1000 | DLP (2 ^{-ΔCT})*1000 | non-DLP vs DLP P-value | non-DLP (2 ^{-ΔCT})*1000 | DLP (2 ^{-ΔCT})*1000 | non-DLP vs DLP P value |
| AGER | 6.38 ± 0.56 | 7.72 ± 1.14 | 0.375 | 8.58 ± 0.67 | 8.02 ± 0.71 | 0.718 |
| ANGPT1 | 99.31 ± 10.92 | 79.85 ± 14.66 | 0.076 | 63.65 ± 9.4 | 50.87 ± 3.9 | 0.279 |
| ANGPT2 | 12.22 ± 1.51 | 18.48 ± 3.39 | 0.178 | 9.71 ± 1.68 | 8.07 ± 0.75 | 0.622 |
| CAPG | 292.66 ± 33.87 | 303.23 ± 37.35 | 0.87 | 237.78 ± 19.61 | 271.04 ± 19.01 | 0.200 |
| CD34 | 321.84 ± 30.3 | 376.67 ± 49.15 | 0.412 | 310.62 ± 37.44 | 330.78 ± 27.89 | 0.412 |
| CD68 | 628.63 ± 54.92 | 641.63 ± 65.49 | 0.922 | 559.19 ± 45.88 | 472.23 ± 24.58 | 0.178 |
| COL18A1 | 210.01 ± 12.87 | 278.09 ± 30.09 | 0.033 | 227 ± 17.77 | 214.93 ± 15.05 | 0.718 |
| DLL4 | 31.37 ± 4.11 | 41.57 ± 3.9 | 0.061 | 26.63 ± 4.61 | 29.46 ± 2.24 | 0.158 |
| ENG | 309.31 ± 15.67 | 399 ± 59.1 | 0.123 | 284.18 ± 23.59 | 306.56 ± 14.45 | 0.450 |
| IFI30 | 281.41±33.01 | 296.18 ± 47.51 | 0.768 | 271.26 ± 36.59 | 221.64 ± 27.42 | 0.309 |
| NOTCH3 | 264.52 ± 23.1 | 410.23 ± 62.56 | 0.071 | 81.19 ± 9.96 | 88.18 ± 7.67 | 0.622 |
| PECAM1 | 439.63 ± 27.2 | 555.04 ± 93.92 | 0.450 | 340.81 ± 38.92 | 315.11 ± 21.21 | 0.818 |
| PGF | 5.54 ± 0.61 | 8.1 ± 1.32 | 0.123 | 8.1 ± 0.69 | 8.42 ± 0.7 | 0.670 |
| PLIN2 | 316.24 ± 39.11 | 278.91 ± 38.62 | 0.870 | 245.07 ± 20.76 | 270.29 ± 20.37 | 0.279 |
| PNPLA2 | 1165.41 ± 81.6 | 1004.53 ± 90.79 | 0.341 | 843.06 ± 118.89 | 819.32 ± 80.7 | 0.718 |
| PTX3 | 2.66 ± 0.67 | 9.38 ± 4.58 | 0.622 | 5.47 ± 0.96 | 5.42 ± 0.7 | 0.870 |
| SERPINF1 | 2573.44 ± 235.73 | 2551.76 ± 258.39 | 0.974 | 1747.51 ± 231.96 | 1663.02 ± 114.38 | 0.533 |
| SPP1 | 190.05 ± 37.93 | 287.06 ± 76.25 | 0.622 | 74.67 ± 19.25 | 90.01 ± 17.98 | 0.341 |
| TEK | 62.71 ± 5.92 | 68.93 ± 7.51 | 0.450 | 53.57 ± 5.72 | 52.47 ± 3.34 | 0.870 |
| TNC | 30.96 ± 6.72 | 46.62 ± 13.16 | 0.412 | 11.83 ± 1.11 | 13.8 ± 1.94 | 0.768 |
| VEGFA | 194.1 ± 21.6 | 202.24 ± 35.12 | 0.768 | 230.42 ± 24.19 | 232.22 ± 14.15 | 0.670 |

Supplemental Table S6. Gene expression of SAT and VAT from non hypertens (non-HP) and hypertens (HP) obese patients. Significance between non-HP versus HP.

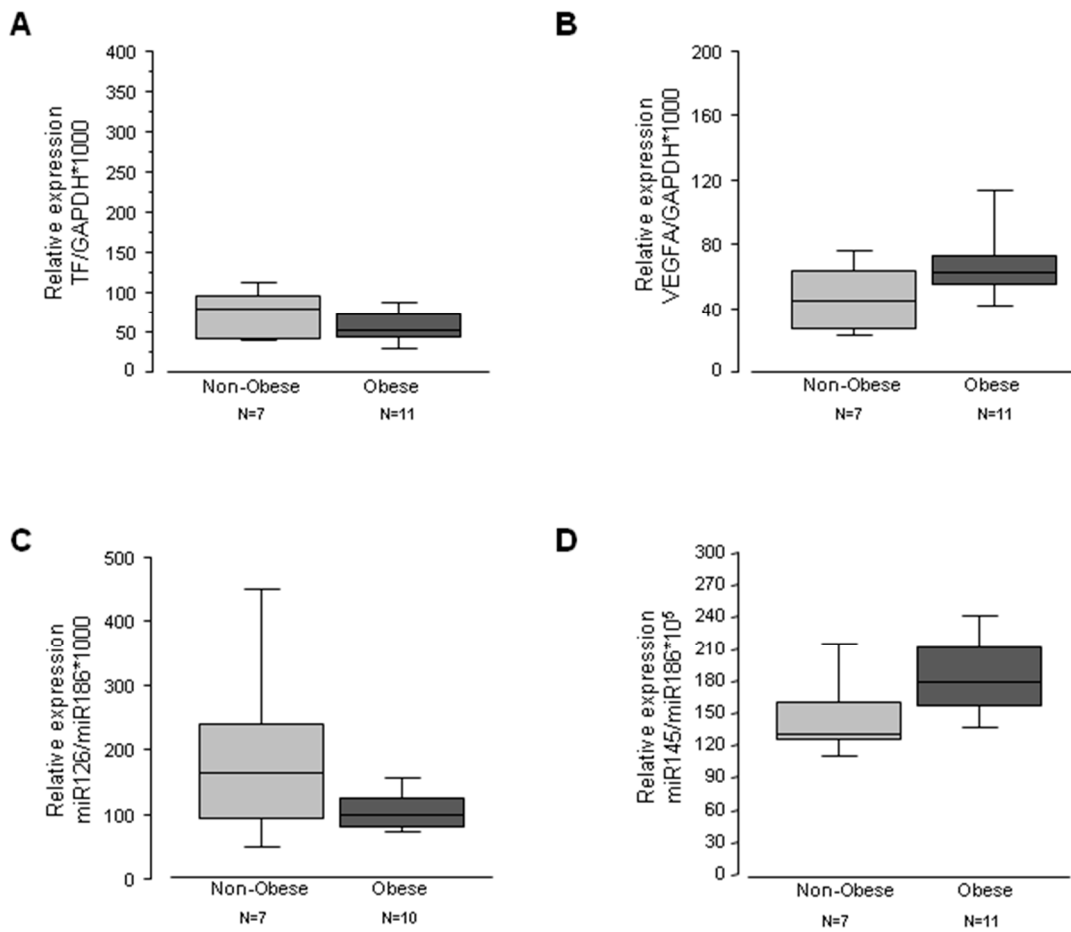
| Gene | SUBCUTANEOUS | | | VISCERAL | | |
|----------|--------------------------------------|----------------------------------|------------------------------|--------------------------------------|----------------------------------|------------------------------|
| | non-DLP (2 ^{-ΔCT})*1000 | DLP (2 ^{-ΔCT})*1000 | non-DLP vs DLP P-value | non-DLP (2 ^{-ΔCT})*1000 | DLP (2 ^{-ΔCT})*1000 | non-DLP vs DLP P value |
| AGER | 6.89±0.63 | 7.4±1.57 | 0.860 | 7.99±0.6 | 8.61±0.77 | 0.698 |
| ANGPT1 | 101.59±9.99 | 77.58±14.98 | 0.007 | 62.55±9.53 | 51.96±3.91 | 0.972 |
| ANGPT2 | 13.22±1.71 | 17.48±3.44 | 0.459 | 9.02±1.55 | 8.76±1.07 | 0.459 |
| CAPG | 303.05±33.52 | 292.84±37.67 | 0.597 | 253.76±19.66 | 255.06±20.36 | 0.459 |
| CD34 | 330.51±28.51 | 368±51 | 0.805 | 321.63±35.65 | 319.77±30.48 | 0.062 |
| CD68 | 626.47±55.72 | 643.79±64.76 | 0.647 | 573.5±41.87 | 457.92±25.82 | 0.217 |
| COL18A1 | 220.47±17.25 | 267.62±29.9 | 0.459 | 229.86±18.38 | 212.06±14 | 0.549 |
| DLL4 | 33.57±4.23 | 39.37±4.22 | 0.805 | 22.81±2.82 | 33.28±3.64 | 0.045 |
| ENG | 312.04±16.8 | 396.27±59.19 | 0.148 | 268.59±18.91 | 322.16±17.01 | 0.053 |
| IFI30 | 279.69±32.93 | 297.91±47.51 | 0.418 | 287.02±34.88 | 205.87±25.85 | 0.148 |
| NOTCH3 | 254.68±22.97 | 420.08±60.11 | 0.113 | 73.13±8.24 | 96.24±8.12 | 0.459 |
| PECAM1 | 465.39±34.97 | 529.29±93.8 | 0.751 | 322.51±35.39 | 333.4±27.18 | 0.307 |
| PGF | 6.1±0.78 | 7.54±1.31 | 0.098 | 8.4±0.81 | 8.12±0.54 | 0.972 |
| PLIN2 | 333.73±39.39 | 261.42±35.75 | 0.698 | 256.82±22.75 | 258.53±18.97 | 0.192 |
| PNPLA2 | 1209.45±77.95 | 960.49±83.78 | 0.698 | 862.61±123.57 | 799.77±72.16 | 0.307 |
| PTX3 | 6±3.45 | 6.05±3.43 | 0.148 | 5.33±0.98 | 5.56±0.67 | 0.698 |
| SERPINF1 | 2671.73±211.38 | 2453.48±274.4 | 0.275 | 1781.31±226.49 | 1629.22±121.6 | 0.698 |
| SPP1 | 205.37±35.63 | 271.74±78.96 | 0.459 | 60.24±12.02 | 104.44±21.53 | 0.140 |
| TEK | 65.64±5.8 | 66±7.72 | 0.751 | 51.64±5.86 | 54.41±3.05 | 0.378 |
| TNC | 29.88±6.21 | 47.7±13.28 | 0.032 | 11.25±1.04 | 14.39±1.9 | 0.418 |
| VEGFA | 206.1±20.72 | 190.24±35.52 | 0.805 | 238.96±23.92 | 223.68±14.22 | 0.916 |

Supplemental Figure S1



(A) IPA network analysis of target genes in subcutaneous adipose tissue from obese and non obese patients. (B) IPA network analysis of target genes in subcutaneous or visceral adipose tissue.

Supplemental Figure S2



Box-plot diagram showing ASC genomic profile. A) Differential protein expression of TF in sASC from obese vs non-obese patients. B) Differential protein expression of VEGFA in sASC from obese vs non-obese patients. C) Differential protein expression of miR126 in sASC from obese vs non-obese patients. D) Differential protein expression of miR145 in sASC from obese vs non-obese patients.