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Transcriptomic comparisons between cultured human adipose tissue-derived pericytes and mesenchymal stromal cells

Lindolfo da Silva Meirelles [a,b,*], Tathiane Maistro Malta [b], Rodrigo Alexandre Panepucci [c], Wilson Araújo da Silva Jr. [b,d]

[a] Laboratory for Stem Cells and Tissue Engineering, PPGBioSaúde, Lutheran University of Brazil, Av. Farroupilha 8001, 92425-900 Canoas, RS, Brazil
[b] Center for Cell-Based Therapy (CEPID/FAPESP), Regional Center for Hemotherapy of Ribeirão Preto, University of São Paulo, Rua Tenente Catão Roxo 2501, 14051-140 Ribeirão Preto, SP, Brazil
[c] Laboratory of Large-Scale Functional Biology (LLSFBio), Regional Center for Hemotherapy of Ribeirão Preto, University of São Paulo, Rua Tenente Catão Roxo 2501, 14051-140 Ribeirão Preto, SP, Brazil
[d] Department of Genetics, School of Medicine of Ribeirão Preto, University of São Paulo, Av. Bandeirantes 3900, 14049-900 Ribeirão Preto, SP, Brazil

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ABSTRACT

Mesenchymal stromal cells (MSCs), sometimes called mesenchymal stem cells, are cultured cells able to give rise to mature mesenchymal cells such as adipocytes, osteoblasts, and chondrocytes, and to secrete a wide range of trophic and immunomodulatory molecules. Evidence indicates that pericytes, cells that surround and maintain physical connections with endothelial cells in blood vessels, can give rise to MSCs (da Silva Meirelles et al., 2008 [1]; Caplan and Correa, 2011 [2]). We have compared the transcriptomes of highly purified, human adipose tissue pericytes subjected to culture-expansion in pericyte medium or MSC medium, with that of human adipose tissue MSCs isolated with traditional methods to test the hypothesis that their transcriptomes are similar (da Silva Meirelles et al., 2015 [3]). Here, we provide further information and analyses of microarray data from three pericyte populations cultured in pericyte medium, three pericyte populations cultured in MSC medium, and three adipose tissue MSC populations deposited in the Gene Expression Omnibus under accession number GSE67747.

Specifications	
Organism/cell line/tissue	*Homo sapiens*
Sex	*Female*
Sequencer or array type	*Whole Human Genome Oligo Microarray chips (Agilent, G4112F and G4845A; design IDs 014,850 and 026,652, respectively)*
Data format	*Raw: TXT; normalized data: TXT, SOFT, MINiML*
Experimental factors	*Human adipose tissue mesenchymal stromal cells vs. human adipose tissue pericytes cultured under mesenchymal stromal cell conditions; human adipose tissue pericytes vs. human adipose tissue pericytes cultured under mesenchymal stromal cell conditions; human adipose tissue mesenchymal stromal cells vs. human adipose tissue pericytes cultured in pericyte medium.*
Experimental features	*The transcriptomes of highly purified, human adipose tissue-derived pericytes subjected to culture-expansion in pericyte medium ($n = 3$) were compared to those of human adipose tissue mesenchymal stromal cells ($n = 3$) and human adipose tissue pericytes cultured under mesenchymal stromal cell conditions ($n = 3$) to detect differentially expressed transcripts.*
Consent	*Informed consent was obtained from all tissue donors enrolled in the study.*
Sample source location	*Ribeirão Preto, São Paulo, Brazil*

1. Direct link to deposited data

http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE67747

2. Experimental design, materials and methods

2.1. Samples and sample donors

Adipose tissue was obtained from patients undergoing elective plastic surgery at the University Hospital of the School of Medicine of Ribeirão Preto, University of São Paulo at Ribeirão Preto, Brazil. All patients provided informed consent for the use of their biological material in this study. This study was approved by the Brazilian National Commission on Ethics in Research (CAAE 0054.0.004.000-08).

Adipose tissue pericytes were isolated from donors 1, 2 and 3 (and named cAT3G5Cs 1, 2, and 3, respectively), and adipose tissue mesenchymal stromal cells were isolated from donors 16, 17 and 18 (and named ATMSCs 16, 17, and 18, respectively). Adipose tissue pericytes from donors 1, 2, and 3 were also cultured under mesenchymal stromal cell conditions prior to transcriptomic analyses, and named cAT3G5Cs 1 DME10, cAT3G5Cs 2 DME10, and cAT3G5Cs 3 DME10, respectively. The samples and corresponding data used here were obtained in a previously published study [3]. All tissue donors were females. Tissue

* Corresponding author at: Laboratory for Stem Cells and Tissue Engineering, PPGBioSaúde, Lutheran University of Brazil, Av. Farroupilha 8001, 92425-900 Canoas, RS, Brazil.
E-mail addresses: lindolfomeirelles@gmail.com, lindolfomeirelles@ulbra.edu.br (L. da Silva Meirelles).

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samples used to isolate the cells consisted of liposuction material with exception of the sample obtained from donor 17, which was a tissue fragment removed during dermolipectomy.

2.2. Microarray hybridization and scanning

RNA was extracted using TRIzol LS reagent (Life Technologies do Brasil Ltda, São Paulo, SP, Brazil), and cleaned up using the RNeasy mini kit (QIAGEN Biotecnologia Brasil Ltda, São Paulo, SP, Brazil) following the manufacturers' instructions. RNA was quantified using a NanoDrop 1000 spectrophotometer (ThermoScientific, Wilmington, DE).

Oligonucleotide microarrays from two 4 × 44K Whole Human Genome Microarray Kits, (G4112F, and G4845A; design IDs 014850, and 026652, respectively; Agilent Technologies, Santa Clara, CA), which contain probes for more than 41,000 gene transcripts, were used to analyze gene expression of the samples. A predetermined amount of control bacterial RNA from the One Color RNA Spike-In Kit (Agilent, 5188–5282) was added to total RNA prior to synthesis of complementary RNA (cRNA) and labeling with cyanine 3 (Cy3) using the One Color Quick Amp Labeling Kit (Agilent, 5190–0442). RNA was reverse-transcribed using oligo (dT) containing a promoter for RNA T7 polymerase. The resultant cDNA was purified, fragmented, and used as template for cRNA in vitro transcription using T7 RNA polymerase and nucleotides, which included Cy3-CTP for labeling. The cDNA obtained was purified using the Illustra RNAspin mini Kit (25-0500-71; GE Healthcare Life Sciences, Logan, UT). cDNA quantitation and labeling efficiency were determined using a NanoDrop 1000 spectrophotometer (ThermoScientific). Labeled cRNA was hybridized with microarray slides using the Gene Expression Hybridization Kit (Agilent, 5188–5242) in SureHyb hybridization chambers (Agilent, G2534A) for 17 h at 65 °C at 10 RPM in a hybridization oven (Agilent, G2545A). After hybridization, microarray slides were washed and dried. The slides were then scanned at 535 nm with a resolution of 5 μm/pixel using a DNA Microarray Scanner with Sure Scan High-Resolution Technology (Agilent). Expression data were extracted using Agilent's Feature Extraction software versions 8.5 or 11.5.

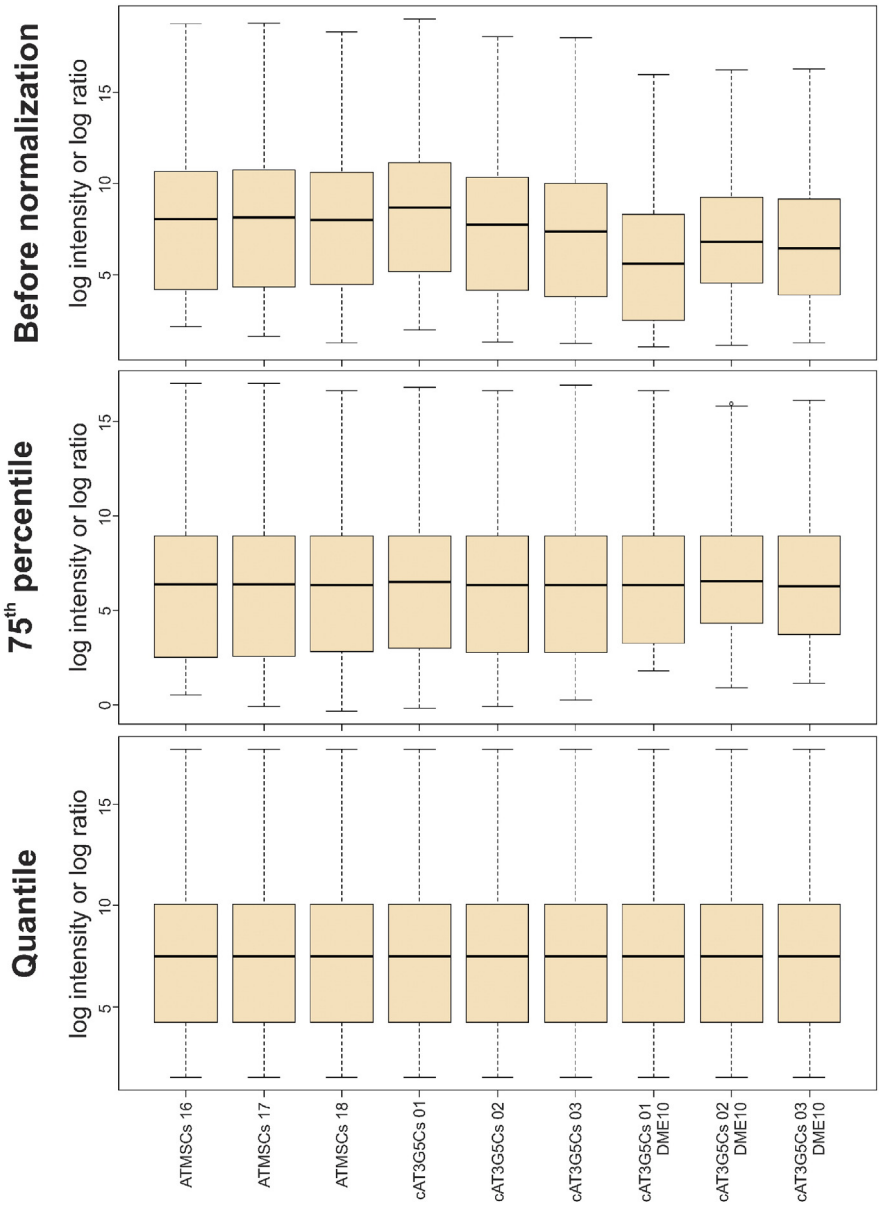

Fig. 1. Boxplots showing the distribution of expression values of non-control probes in each consolidated microarray dataset before normalization (top panel), after normalization to the 75th percentile (middle panel), and after quantile normalization (bottom panel).