



Data in Brief

Transcriptome analysis of bone marrow mesenchymal stromal cells from patients with primary myelofibrosis



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ABSTRACT

Primary myelofibrosis (PMF) is a clonal myeloproliferative neoplasm whose severity and treatment complexity are attributed to the presence of bone marrow (BM) fibrosis and alterations of stroma impairing the production of normal blood cells. Despite the recently discovered mutations including the JAK2V617F mutation in about half of patients, the primitive event responsible for the clonal proliferation is still unknown. In the highly inflammatory context of PMF, the presence of fibrosis associated with a neoangiogenesis and an osteosclerosis concomitant to the myeloproliferation and to the increase number of circulating hematopoietic progenitors suggests that the crosstalk between hematopoietic and stromal cells is deregulated in the PMF BM microenvironmental niches. Within these niches, mesenchymal stromal cells (BM-MSC) play a hematopoietic supportive role in the production of growth factors and extracellular matrix which regulate the proliferation, differentiation, adhesion and migration of hematopoietic stem/progenitor cells. A transcriptome analysis of BM-MSC in PMF patients will help to characterize their molecular alterations and to understand their involvement in the hematopoietic stem/progenitor cell deregulation that features PMF.

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Specifications

Organism/cell line/tissue	Primary mesenchymal stromal cells from human bone marrow
Sex	Pathology sex independent
Sequencer or array type	Agilent single color Oligo Microarray 4 × 44k
Data format	Agilent normalized matrix
Experimental factors	Bone marrow from primary myelofibrosis patients versus healthy donors (similar age)
Experimental features	Adult (60–80 years old) primary BM-MSCs amplified in vitro for 3 to 5 passages – GSE44426
Consent	Helsinki consent
Sample source location	France and Italia

1. Direct link to deposited data

<http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE44426>.

1.1. Experimental design, materials and methods

The transcriptome of medullary stromal cells of patients suffering from primary myelofibrosis was studied by Agilent Oligo microarray technology. The primary myelofibrosis is a chronic myeloproliferative syndrome. To invest the role of bone marrow stroma in the pathophysiology of this disease, we isolated primary cultured of bone marrow stromal cells from these patients. The osteo-medullary biopsies for the diagnosis of the disease were implanted in DMEM medium with 10% fetal calf serum. Stromal cells during their proliferation adhere to plastic and they were trypsinized between each passage (3–5 passages) when cultures came to confluence. These steps were performed to eliminate

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the remaining hematopoietic cell fraction in these cultures. A cytometric control was carried out on the cells prior to performing molecular biology experiments. CD105, CD73 and CD90 marker positivity was verified to validate the mesenchymal cell phenotype. The negativity of the CD45 marker was also carried out to prove the absence of residual hematopoietic cells in culture. Each culture of mesenchymal stromal cell is isolated from the bone marrow of an individual. In total, the bone marrow samples were studied individually from 6 healthy donors (6 controls: GSM1084994, GSM1084995, GSM1084996, GSM1084997, GSM1084998, GSM1084999) and from 6 patients with primary myelofibrosis (6 PMF: GSM1085000, GSM1085001, GSM1085002, GSM1085003, GSM1085004, GSM1085005). Concerning the enrollment of control samples: subjects are negative for alcohol abuse and HCV, HBV and HIV virus infections. The choice of control subjects was conditioned by access to bone marrow from subjects having a hip prosthesis surgery: indeed the subjects have an average age similar to that of patients with primary myelofibrosis (between 60 and 80 years). Each sample was treated individually for the extraction of nucleic acids and the achievement of microarrays. RNA was isolated using RNA extraction protocols (NucleoSpin RNA II, Macherey-Nagel) on the Miltenyi platform. RNA samples were quality-checked via the Agilent 2100 Bioanalyzer (Agilent Technologies). Total RNA sample (1 µg) was used for linear T7-based amplification. RNA samples were amplified and labeled using the Agilent Quick Amp Labeling Kit/Low RNA Input Linear Amp Kit (Agilent Technologies). The hybridization procedure was performed using Agilent Gene Expression Hybridization Kit (Agilent Technologies). Briefly, 1.65 µg Cy3-labeled fragmented cRNA in hybridization buffer was hybridized

overnight (17 h, 65 °C) to Agilent Whole Human Genome Oligo Microarrays 4 * 44k using Agilent's in hybridization chamber. The fluorescence signals were detected using Agilent's Microarray Scanner System (Agilent Technologies). The Agilent Feature Extraction Software (FES) v9.1 was used to read out and process the microarray image files. The software determines feature intensities including background subtraction. The signal intensities from single experiment raw data lists are normalized by dividing the intensities values by their median. Normalized data were accessible on public database: (GEO submission number GSE44426, <http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=GSE44426>), online on Jan. 10, 2015) [1,2].

Appendix A Supplementary data

Supplementary data to this article can be found online at <http://dx.doi.org/10.1016/j.gdata.2015.04.017>.

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