

ADIPOSE TISSUE AS A NOVEL RESOURCE FOR BONE REGENERATION: ANALYSIS OF OSTEOGENIC POTENTIAL OF ADIPOSE-DERIVED MESENCHYMAL CELLS

S. Pagani^{1,2}, M. Sartori², E. Della Bella¹, F. Veronesi¹, G. Giavaresi^{1,3}, L. Negosanti⁴, V. Pinto⁴, M. Fini^{1,2}, P.G. Morselli⁴

¹Laboratory of Preclinical and Surgical Studies, and ²Laboratory of Biocompatibility, Technological Innovations, and Advanced Therapies (BITTA), RIT Department, Rizzoli Orthopaedic Institute, Bologna; ³Laboratory of Tissue Engineering - Innovative Technology Platforms for Tissue Engineering (PON01-00829), Rizzoli Orthopaedic Institute, Palermo; ⁴Division of Plastic and Reconstructive Surgery, S.Orsola-Malpighi Polyclinic Hospital, University of Bologna, Italy.

E-mail: stefania.pagani@ior.it

The novelty in mesenchymal stem cell research has been represented by the adipose tissue as a promising source of mesodermal derived-multipotent cells, called adipose stromal cells (ASCs). Adipose tissue can be harvested with a minimally invasive liposuction, and ASCs could overcome important constraints such as harvest site pain, morbidity and risks of infection. The aim of this study was to analyze the influence of different human adipose harvesting sites on ASC yield, proliferation, stemness, characterization and osteogenic potential when cultured in differentiating condition.

Eighteen specimens were collected by liposuction from 14 subjects (12 females and 2 males) with different age, body weight, height and body mass index: 6 samples were obtained from abdominal area, 5 from trochanteric area and 7 from breast.

Surface characterization showed the typical mesenchymal CD pattern (CD44, CD73, CD90, CD105) and negative expression of endothelial and hematopoietic markers (CD31 and CD45). No significant differences among the harvesting sites were found for ASC characterization, yield and stemness, collagen type I gene activation, alkaline phosphatase synthesis and mineralized nodules formation. Abdomen derived ASCs showed lower proliferation values than breast and trochanteric ASCs and higher values of RUNX2 and TGF 1. Finally, a significant synthesis of VEGF (vascular endothelial growth factor, important for its angiogenic role) was found in ASC cultures.

These preliminary results demonstrate that all sample of ASCs, regardless to harvesting sites and patient characteristics, are able to differentiate into osteoblasts. Mesenchymal cells adipose-derived could be a viable and abundant cell source useful in orthopedic regenerative medicine.

PHENOTYPIC PROFILING OF OSTEOTROPIC BREAST CANCER CELLS

I. Pucci Minafra¹, G. Di Cara¹, R. Musso¹, N.N. Albanese², G. Peri³, B. Valentino³, M. D'Arienzo⁴, D. Martini⁵, M. Raspanti⁶

¹Centro di Oncobiologia Sperimentale (C.OB.S), University of Palermo and La Maddalena Hospital, Palermo, Italy; ²Dipartimento di Scienze e Tecnologie Biologiche Chimiche e Farmaceutiche (STEBICEF), University of Palermo, Italy; ³Dipartimento di Biomedicina Sperimentale e Neuroscienze Cliniche, University of Palermo, Italy; ⁴Dipartimento Discipline Chirurgiche, Oncologiche e Stomatologiche (DICHIRONS), University of Palermo, Italy; ⁵Dipartimento di Scienze Biomediche e Neuromotorie (DIBINEM), sez Anatomia Umana, University of Bologna, Italy; ⁶Dipartimento di Scienze Chirurgiche e Morfologiche, University of Insubria, Varese, Italy.

E-mail: ida.pucci@unipa.it

One of the preferred locations of metastases from breast cancer is the bone tissue. On the other hand, it should be recalled that mammary tumors with equal clinical diagnosis have a different course, and also different metastatic progression. Therefore, it would be helpful to have appropriate markers of osteotropism to test on the surgical cancer tissues, in order to predict the possible propensity of the breast cancer to generate bone metastases and to adequate the therapeutic plan.

We previously reported^{1,2} on the setting-up of an in vitro model for the study of the osteotropic propensity of breast cancer cells and the influences exerted by the bone microenvironment on the cancer cells phenotype.

Viable bone fragments, deriving from surgery on traumatic lesions of young subjects were washed from bio-contaminants under sterile conditions and placed into cell culture capsules with the proper culture medium supplemented with foetal bovine serum, L-glutamine, L-ascorbic acid and antibiotics. The explants were kept for controlled timing in the humidified incubator in order to promote the release of resident cells, and then co-cultured with under-confluent breast cancer cells (SKBR3), until confluence. The bone fragments were then recovered, washed, placed again in culture dishes and monitored daily. The cells released from the bone fragments were then collected and processed for immunological characterization and proteomic profiling. The proteomic profiles of the cells seeded into the bone fragments were compared with the original cell culture, revealing an interesting differential proteomic pattern. The collection of identified proteins on the maps has reached up today the number of 373. Differentially expressed proteins between bone-seeded cells and wild type cells were about 30%. Among the differentially expressed proteins were several proteins belonging to the cytoskeleton remodelling and proteins of the class of calcium-binding cluster. The relevance of these protein clusters is discussed.

The work was co-funded by the Italian 5x1000 to COBS.

1. Pucci Minafra I et al. Eur J Histochem 2013, 57:2.

2. Di Cara G et al. Eur J Histochem 2014, 58:8.