

# Laboratory Listing

Contact Name: Lydia Masako Ferreira

Laboratory Name: Cell Culture and Stem Cell Laboratory of Plastic Surgery Division- Federal University of São Paulo

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Animal Facilities (Yes/No): Yes

Type(s) of Animal(s): Rats and Rabbits

Type of Research: Experimental research (in vitro analyses and in vivo)

**Funding:** CNPQ (Conselho Nacional de Desenvolvimento Científico e Tecnologico) 312356/2009-9

## Research Fellowship Available: (Yes/No): Yes

## **Primary Investigator's Name:**

- 1. Lydia Masako Ferreira
- 2. Antonio Carlos Aloise

#### **Project Title:**

1. AngiotensinII in human adipose-derived stem cell (ASCs)

2. Expression of receptor activator NF-kB ligand (RANKL), alkaline phosphatase (ALP) and osteocalcin (OC) in human dermal fibroblasts and preadipocytes cultivated in osteogenic medium.

## **Brief Project Description:**

1. There is a crescent scientific interest about the plasticity and therapeutic potential of adipose-derived stem cells (ASCs), multipotent and abundant cells in adipose tissue, which are

able to differentiate in multiple cellular lineages in vitro, including adipocytes, chondrocytes, osteoblasts, neural cells, endothelial and cardiomyocytes. The aim of this study is to verify the effect of AII on ASCs. Human adipose tissue precursor cells will be obtained of subcutaneous abdominal tissue, separated by density centrifugation gradient, cultivated and stimulated with different culture media and then, analyzed. Oil Red O will be used for morphological characterization by fluorescence microscopy. Cell cycle phases, immunophenotypic characterization, size and complexity will be determined by flow cytometry.

2. The bone tissue engineering (BTE) using scaffolds and cell therapy, has established itself as an alternative for the treatment of critical bone defects with decreased morbidity compared to the standard procedure of graft. The objective of this study is to evaluate the gene expression of the receptor-activating NF-kB (RANKL), alkaline phosphatase and osteocalcin in dermal fibroblasts and human preadipocytes cultured in osteogenic medium. Will be held the primary culture of dermal fibroblasts and preadipocytes from discards of abdominoplasty procedures performed by plastic surgery division of Federal University of São Paulo. The cells are cultured and expanded and after the fourth passage will be frozen until used in experiments. Evaluations will performed in the second, seventh, tenth to fourth, twenty-first and twenty-eighth days of culture, where the cells are grown in culture medium to sort itself Cellular and also in osteogenic medium, using plates of 12 wells. At each evaluation point of the study will be performed fixation with 4% formaldehyde and performs the following ratings: Immunocytochemistry for osteocalcin and alkaline phosphatase in all experiments; flow cytometry to characterize the populations of adult stem cells in the two populations cells (preadipocytes and fibroblasts); trypan blue for cell viability in all experiments; ELISA for alkaline phosphatase and osteocalcin to evaluate the culture medium in all experiments; and gene expression for RANKL mRNA (RT-PCR) in all experiments and detection of mineralized nodules by alizarin red.

# Opportunity for student degree program (Yes/No): Yes

# Funding available from supervisor – if yes, include details: Yes

 CNPq Proc.: 312356/2009-14. Coordinator: Lydia Masako Ferreira
This project received a institucional grant from PNPD (Post Doctorate National Program) -CAPES (Coordination of Improvement of Higher Education Personel) number: 007243/2011-16 as well as a scholarship for post doctorate for a period of 36 months from December 2011. Supervisor: Lydia Masako Ferreira Post doctorate student: Antonio Carlos Aloise