BIO002

Evaluation of osteogenic effect by different phyestrogens in osteoblasts culture derived from mesenchymal stem cells

Amanda Natalina de Faria[1*]; Pietro Ciancaglini[2]; Ana Maria Sper Simão[2]; Daniela Cervelle Zancanela[2]; Maria José Vieira Fonseca[1]; Maria Regina Torqueti[1]

[1]Faculdade de Ciências Farmacêuticas de Ribeirão Preto - Universidade de São Paulo; [2]Faculdade de Filosofia, Ciências e Letras de Ribeirão Preto - Universidade de São Paulo
[*]amandanf@fcfrp.usp.br

Introduction: Menopause is caused by the failure of ovarian hormones production and can produce unfavorable changes in lipid, glucose and insulin metabolism, in the coagulation/fibrinolysis system, as well as bone loss are some of its consequences. One of the current concerns is the relationship between the decrease in estrogen production and bone mass loss, which is considered a major risk factor for the development of osteoporosis in women. Osteoporosis is currently considered a worldwide public health problem and it is estimated that by 2030, more than one billion and 200 million women will be menopausal. One of the ways to treat menopause symptoms and decrease the chance of someone to develop osteoporosis is the use of the Hormone Replacement Therapy (HRT). However, this therapy has brought some risks to the health of some groups of women, especially those with a history of thromboembolic diseases and breast cancer in their families, and the use of phytoestrogens such as isoflavones is an alternative to the traditional treatment. Isoflavones are found mainly in soybeans (*Glycine max*), red clover (*Trifolium pratense*), *Cimicifuga racemosa* (of American origin) and rye. **Objective:** In this work we compare the ability of two phytoestrogens preparations (one is the soybean extract biotransformed by the fungus *Aspergillus awamori* (ESBF) produced by Faculdade de Ciências Farmacêuticas de Ribeirão Preto - USP, and the other is Menoflavon® 40 mg (Melbrosin International) composed of *Trifolium pratense* isoflavones) to stimulate osteogenesis *in vitro,* using osteoblasts cultures derived from mesenchymal stem cells. **Methods:** Evaluation of cell viability by the MTT method and formation of mineralized matrix. **Results:** a) In the viability assays, ESBF was 106.3% viable at a concentration of 0.5 µg/mL, 93.1% viable at a concentration of 1 µg/mL and 92.4% viable at a concentration of 4 µg/mL. For these concentrations of 0.5 µg/mL, 1 µg/mL and 4 µg/mL ESBF, the main soybean metabolites (daidzein (D) and genistein (G)) were determined by HPLC and we found 1.181 nM D and 0.922 nM G; 2.361 nM D and 1.845 nM G; and 9.445 nM D and 7.379 nM G, respectively. When using Menoflavon® for the viability assays, we found 97.8%, 85.4% and 86.5% of live cells at a concentration of 0.5 µg/mL, 1 µg/mL and 4 µg/mL, respectively, corresponding to 7.5 ng/mL G and 28.75 ng/mL D; 15 ng/mL G and 57.5 ng/mL D; 60 ng/mL G and 230 ng/mL D, respectively. b) The formation of mineralized matrix on day 14 of culture by ESBF was 105.7%, 114.8% and 101.7%, and at day 21 of culture, it was 105.2%, 79.9% and 79.5%, at a concentration of 0.5 µg/mL, 1 µg/mL and 4 µg/mL, respectively. When using Menoflavon®, the formation of mineralized matrix on day 14 of culture was 104.1%, 83.6% and 106.4%, and at day 21 of culture, it was 98.6%, 87.8% and 83.5%, at a concentration of 0.5 µg/mL, 1 µg/mL and 4 µg/mL, respectively. D and G values were the same as presented above (item a). **Conclusion:** With the results obtained, it is not possible to conclude which isoflavones preparation out of the two is the best to induce bone mineralization.

Keywords: Menopause, Isoflavones extract, Biotransformed soy, Menoflavon®, Osteogenesis, Mesenchymal stem cells, Osteoblasts.

Financial support: FAPESP / CNPq / CAPES