J Cell Sci Ther 2017, 8:1 (Suppl)
http://dx.doi.org/10.4172/2157-7013.C1.039

Genetic changes at chromosomal and DNA level during long term cultivation of hES cells

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Human embryonic stem cells (hESCs) are important research tools in studies of the physiology of early tissue differentiation. In addition these cells are regarded as a promising approach to generate transplantable cells for the treatment of several diseases, and therefore offer an immense potential as a source of cells for regenerative medicine. However the possible ability of these cells to produce tumors *in vivo* presents a major impediment for this achievement. hESCs can obtain growth advantages *in vitro* by acquired mutations. The mechanisms that may influence chromosome modification in hESCs are not well known. We have performed a comparative *in vitro* and *in vivo* study on hESC lines produced in our laboratory to see if there are changes also during *in vivo* growth. *In vivo* differentiated cells and *in vitro* cultured hESCs were analyzed by using first comparative genome hybridization (CGH) and second a high-resolution Affymetrix SNP 6.0 array revealing DNA copy number variations. We were able, for the first time, to identify an aberrant X chromosome both *in vitro* and *in vivo* in one out of the 3 hESC line, we decteted an amplification of the whole X chromosome, possibly due to mosaicism of XY and XX cells. In the other hESC line, array results showed small amplifications and gains. The third hESC line was less altered, but contained also a new gain verified by fluorescent in situ hybridization in a teratoma in 21% of the cells. These results indicate that mutations occur during the *in vivo* differentiation process as well as *in vitro*. The potential of precancerous mutations in in-vivo conditions is important to consider for safety measures, and underlines the necessity to remove all pluripotent stem cells from the differentiated cell population that will be transplanted.

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Cutting edge concepts in the use of stem cell and PRP injections in an office setting

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The presentation concerns PRP and stem cell (both bone marrow and adipose) injections for musculoskeletal conditions in an office setting. Indications are given as to which type of cell and technique to use to accomplish repair. Stem cells, both bone marrow derived (BMAC) and adipose, are used for the more difficult problems. PRP injections are utilized for the less severe problems. Indications are given when to use stem cells verses PRP and when to use both. The newest concepts in stem cell science are presented. These concepts include the clinical use of MUSE cells, exosomes, and blastomere like stem cells. Basic science of both PRP and stem cells are discussed. This presentation defines what constitutes an effective PRP preparation. Myths concerning stem cells are dispelled. One myth is that mesenchymal stem cells are the most important stem cell. This was the initial interpretation of Dr. Arnold Caplan the Father of Mesenchymal Stem Cell Science. Dr. Caplan now feels that MSCs have an immunomodulation capacity which may have a more profound and immediate effect on joint chemistry and biology. We now learn in the talk that the hematopoietic stem cells are the drivers of tissue regeneration. Also, discussed are adjuncts used which enhance the results. These therapies include supplements, LED therapy, lasers, electrical stimulation, and cytokine therapy. The scientific rationale is presented for each of these entities as to how they have a direct on stem cells.

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