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

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; identify an aberrant X chromosome both *in vitro* and *in vivo* in one out of the 3 hESC line, we detected 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.

Biography

Jose Inzunza is an associate professor and senior researcher at the Department of Biosciences and Nutrition, Karolinska Institutet (KI). He received his doctorate in obstetrics and gynecology at Karolinska University Hospital, KI. With a specialization in cytogenetics, he worked on his doctoral work with a project in clinical application of preimplantation genetic diagnosis (PGD). He has also been involved in the development and implementation of the laboratory for derivation and differentiation of human embryonic stem cell research at KI. This was the first bank of human embryo stem cells in Sweden and Scandinavia. Jose has also worked with cellular re-programming. Today, Jose's line of research is in stem cells and tumorigenesis and genetic stability of these cells during the differentiation process.

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