HLA-G expression in human preimplantation embryos and embryonic stem cells

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Introduction

Successful implantation in the human is dependent on the early embryo avoiding immune recognition and destruction by the maternal immune system. Human leukocyte antigen G (HLA-G), a non-classical HLA class I antigen, is thought to play a key role in this by modulating cytokine secretion in order to control trophoblast invasion and to maintain a local immunotolerance. HLA-G is secreted in culture medium of cleavage stage IVF embryos but later on in development it is predominantly expressed in the invasive extravillous trophoblast of the placenta and in hematopoietic precursors originated from the extraembryonic yolk sac rather than in the fetus. We investigated the expression of HLA-G throughout human preimplantation development in order to determine the up- and down-regulation of HLA-G in the early embryo.

Materials and methods

Fifty normally fertilized human preimplantation embryos and oocytes donated for research and six human embryonic stem cell lines (hESC) were subjected to indirect immunocytochemistry. Staining was analyzed by confocal microscopy. FACS analysis and immunogold-silver staining were also performed on hESC lines. In each experiment control reactions for non-specific binding of the HLA-G antibody were included. The human choriocarcinoma cell line JEG-3 was used as positive control.

Results

Expression of HLA-G could be detected in the membrane and the cytoplasm of nearly all oocytes, zygotes, cleavage stage and compacting embryos. The proportion of HLA-G expressing embryos and the intensity of the signal increased with developmental stage. At the blastocyst stage, the first stage at which differentiation into trophectoderm (TE) and inner cell mass (ICM) is detectable, HLA-G protein was observed in the TE as well as in all the cells of the ICM. When the blastocyst started to hatch, the TE and the outer cells of the ICM remained positive whereas the staining in the inner cells of the ICM disappeared. Because hESC are undifferentiated pluripotent cells derived from the ICM of blastocysts, we examined six hESC lines by three different staining methods. They all stained positive for HLA-G.

Discussion

HLA-G is expressed in unfertilized oocytes and throughout preimplantation development. In expanded blastocysts, HLA-G is expressed in the TE as well as in all the cells of the ICM. In hatching blastocysts, HLA-G expression is restricted to the TE and to the hypoblast (outer cells of the differentiated ICM). This is in agreement with expression found later during pregnancy in the trophoblast and the yolk sac respectively

Since HLA-G is expressed by hESC but not by epiblast cells (inner cells of the differentiated ICM), we hypothesize that hESC are not derived from the epiblast, as suggested by others, but from earlier undifferentiated ICM cells co-expressing stemness factors (e.g. NANOG) and HLA-G.