Isolation and phenotypical characterization of mesenchymal stem cells from the Wharton's jelly of preterm human umbilical cord

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Extraembryonic tissues such as umbilical cord are considered a promising source of stem cells, potentially useful in therapy. Pre-term umbilical cords may be also a useful source of mesenchymal populations, given also that pre-term birth infants may develop diseases during childhood which may be reverted by a cell therapy approach. Pre-term cords can be also made available from therapeutic abortions, providing a further cell source to obtain high numbers of WJ-MSCs. Little is known about the phenotype and differentiative potential of these cells. Preterm UC were obtained following therapeutic abortions after mothers' informed consent and processed within 12 hours from tissue collection. Characterization of cells was performed at P2 and P5, by both flow cytometry (FC) and ICC, to detect of classical MSC markers, immunomodulatory molecules, tissue-specific markers.

The isolation protocol allowed to successfully derive WJ-MSCs which showed the typical morphology, and were routinely passaged up to passage 10. Multi-color flow cytometric analyses showed that isolated cells were positive for classical MSCs markers (CD29, CD44, CD73, CD90, CD105) and. negative (or weakly positive) for typical hematopoietic and endothelial markers (CD45, CD34, CD14, CD68, CD39 and CD31). In addition, we demonstrated the expression of tissue specific markers, both endodermal (CK18, CK19, alpha-fetoprotein and albumin) and neuro-ectodermal (nestin). This may indicate the potential of preterm WJ-MSC to undergo multiple differentiation pathways, as demonstrated for cells isolated from term UC. Preterm WJ-MSCs showed MHC class I expression (but not class II), suggesting hypoimmunogenic properties for these cells. Moreover, B7H3 expression should favor immune tolerance by the host following cellular transplantation. ICC allowed confirming part of the FC data and was used to assess the expression of further antigens. Present data demonstrate that preterm WJ-MSCs can be isolated and expanded, with high reproducibility. these cells do express classical MSC markers and immunomodulatory molecules, independently from the underlying pathology which led to abortion.

Keywords

Umbilical cord, perinatal stem cells, mesenchymal stem cells, immune modulation.