

Spheroid cell culture of human skin-derived precursors

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SCOPE OF THE METHOD

Alternative method relates to	Human health
Alternative method is situated in	Basic Research, Translational - Applied Research
Type of alternative method	In vitro - Ex vivo
This method makes use of	Human derived cells / tissues / organs
Specify the type of cells/tissues/organs	human skin stem cells

DESCRIPTION

Method keywords

tree-dimensional spheroid culture
hepatic differentiation

Scientific area keywords

spheroids
skin stem cells
differentiation

Method description

For the generation of the human skin-derived precursors (hSKP) spheroids, ultra-low attachment 96-well plates, cell-repellent (Greiner) are used. hSKP cultured on tissue

culture plates are detached with trypsin (5 minutes) and collected in a tube. After centrifugation, hSKP are counted at the Burker chamber and seeded at 2,000 cells per well. The 96-well plates are placed on an orbital shaker, shaking at 40 rpm overnight in a humidified incubator. The outer wells are filled with PBS to reduce evaporation of cell culture medium. The day after, hSKP aggregates of approximately 200-250 μm diameter size are formed. Culture medium is refreshed twice in a day every 3 days to allow complete medium replenishment.

Lab equipment

Cell culture laboratory

Laminar air flow

Ultra-low attachment 96-well plates

Shaker

Method status

Still in development

PROS, CONS & FUTURE POTENTIAL

Advantages

Emulation of 3D cell organization, simple method, applicable to many cell types. Spheroid formation has been shown to enhance anti-inflammatory effects, augment tissue regenerative and reparative effects, facilitate differentiation potentials of multiple lineages, improve stemness properties of adult stem cells.

Challenges

Spheroid handling and transfer step (e.g. spheroids may be easily lost by aspiration with pipette during refreshment of cell culture medium).

Presence of a necrotic core inside the sphere that could lead to poor cell viability in long-term culture.

Future & Other applications

Since spheroid generated from mesenchymal stem cells exert strong anti-inflammatory and immuno-modulatory effects, they could be employed in

regenerative medicine and autoimmune diseases.

REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

References

Cesarz Z, Tamama K (2016) Spheroid Culture of Mesenchymal Stem Cells. *Stem Cells Int* 2016:. doi: 10.1155/2016/9176357

Tsai A-C, Liu Y, Yuan X, Ma T (2015) Compaction, Fusion, and Functional Activation of Three-Dimensional Human Mesenchymal Stem Cell Aggregate. *Tissue Eng Part A* 21:1705–1719. doi: 10.1089/ten.tea.2014.0314

Associated documents

PARTNERS AND COLLABORATIONS

Organisation

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