Manufacturing of fiber scaffolds and cell seeding

SCOPE OF THE METHOD

<table>
<thead>
<tr>
<th>The Method relates to</th>
<th>Human health</th>
</tr>
</thead>
<tbody>
<tr>
<td>The Method is situated in</td>
<td>Basic Research</td>
</tr>
<tr>
<td>Type of method</td>
<td>In vitro - Ex vivo</td>
</tr>
<tr>
<td>Species from which cells/tissues/organs are derived</td>
<td>human skin stem cells</td>
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</tbody>
</table>

DESCRIPTION

Method keywords

fiber scaffolds
electrospinning
polycaprolactone
Scientific area keywords

three-dimensional culture systems
in vitro cell culture
tissue engineering
stem cell culture
differentiation
skin stem cells

Method description

Polycaprolactone (PCL) fiber materials are fabricated using an electrospinning method. Molecular weight of PCL is 45,000 Da (Sigma-Aldrich). The electrospinning process is performed using 18wt% PCL solution dissolved in chloroform:ethanol at a ratio of 9:1. Two high-voltage sources are used to generate positive and negative potentials. The positive source is connected to a syringe needle, whereas the negative source is connected to a collector. Voltage applied to the syringe needle is 20 kV, as well as voltage of the collector. Distance between syringe needle and collector was approximately 20 cm. Obtained 3D PCL fiber materials can be stored until use. From the bulk material of electrospun nanofiber mats, small discs with areas of approximately 15 cm² are cut out and placed in 24-well plastic cell culture plates. Scaffolds are sterilized by gamma sterilization in 70% filtrated ethanol for 30 minutes and further left under UV-light for 30 minutes. All scaffolds are incubated in cell culture medium at 37°C in a humidified 5% CO2 incubator for 1 hour prior to cell seeding to facilitate cell attachment onto the nanofiber. Human skin derived precursors (hSKP) are seeded and cultured on the on fiber scaffolds for 7 days.

Lab equipment

Electrospinning machine;
Cell culture laboratory;
Laminar air flow.

Method status

History of use
PROS, CONS & FUTURE POTENTIAL

Advantages
The fiber mesh mimics features of the biological extracellular matrix, leading to potential improvement of cell morphology and functionality. The scaffolds fit any cell culture plate dimensions, it is handy and easy to use and it is applicable to many cell types.

Challenges
Cells might not infiltrate the fiber mesh and grow on the surface of the scaffold.

Future & Other applications
Improvement of stem cells differentiation potential.

REFERENCES, ASSOCIATED DOCUMENTS AND OTHER INFORMATION

References