

Manufacturing of fiber scaffolds and cell seeding

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SCOPE OF THE METHOD

The Method relates to	Human health
The Method is situated in	Basic Research
Type of method	In vitro - Ex vivo
This method makes use of	Human derived cells / tissues / organs
Species from which cells/tissues/organs are derived	human skin stem cells

DESCRIPTION

Method keywords

fiber scaffolds

electrospinning

polycaprolactone

electrospun fibers

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

M. Rampichová, M. Buzgo, J. Chvojka, E. Prosecká, O. Kofronová, and E. Amler, "Cell penetration to nanofibrous scaffolds: Forcespinning®, an alternative approach for fabricating 3D nanofibers," *Cell Adhes. Migr.*, vol. 8, no. 1, pp. 36–41, 2014

Associated documents

PARTNERS AND COLLABORATIONS

Organisation

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