

MIXING CELLS WITH CELLINK SKIN+

This is a suggested procedure, please adjust according to your experimental needs.

Aim of the protocol:

To demonstrate how to mix cells with **CELLINK SKIN+** bioink.

Materials needed:

- Primary human keratinocytes, HEK
- Primary human fibroblasts, HDF
- **CELLINK SKIN+** bioink
- Syringes with luer lock connections
- [CELLMIXER](#)
- [Female/Female luer lock adaptor](#)
- [Empty Cartridges with End and Tip cap, 3cc](#)
- Pipette
- Pipette tips
- Culture medium

Protocol:

Step n°	Title	Material	Description
1	Prepare cell suspension	<ul style="list-style-type: none"> ▪ Cells ▪ Culture medium 	<ul style="list-style-type: none"> ❖ Resuspend cells in cell culture medium <p>NOTE! Cells should be mixed with CELLINK SKIN+ at a 1:10 ratio to reach the final cell concentration of 5×10^6 cells/ml, 10×10^6 cells/ml or 20×10^6 cells/ml depending on part of construct.</p>
2	Mix the CELLINK SKIN+ bioink with the cells	<ul style="list-style-type: none"> ▪ CELLINK SKIN+ bioink ▪ Cell suspensions ▪ CELLMIXER 	<ul style="list-style-type: none"> ❖ Add cell suspension to a syringe with a pipette (see video) ❖ Add CELLINK SKIN+ to another syringe using a Female/Female luer lock adaptor ❖ The cell suspension is carefully mixed with the CELLMIXER: <p>Please watch the video in this link for a detailed illustration on how to do the mixing process:</p>

CELLINK AB

Arvid Wallgrens Backe 20
41346 Gothenburg,
SWEDEN


675 W Kendall St,
Cambridge, MA 02142
USA

Yoshida-honmachi,
Sakyo-ku, Kyoto 606-8501

JAPAN


CIN No.: 559050-5052

			https://www.youtube.com/watch?v=CmSYL1-oltI
3	Transfer mixed cells and ink to cartridges	<ul style="list-style-type: none"> ▪ Empty Cartridges with End and Tip cap, 3cc ▪ Female/Female luer lock adaptor ▪ Conical tips, 410µm ID (22 GA) 	<ul style="list-style-type: none"> ❖ Transfer the mixed cells and CELLINK SKIN+ bioink with the female/female luer lock adaptor to an Empty Cartridge with End and Tip cap, 3cc ❖ Put a Conical tip, 410µm ID (22 GA) on the cartridge and mount in the printer. ❖ Repeat for all three cartridges.

 Want to see our talented Biologist proceed to this protocol? Feel free to find the video here:
<https://www.youtube.com/...>

Applications:

- ➔ Link to Videos of some applications
- ➔ photos of some applications

 Want to see our existing tissue model?
Just go to <http://bioverse.co/> and discover a whole library of CAD files especially created for sharing 3D Bioprinting models.

References:

Markstedt K *et al.* 3D Bioprinting Human Chondrocytes with Nanocellulose-Alginate Bioink for Cartilage Tissue Engineering Applications. *Biomacromolecules*. 2015;16;1489-96
Martínez H *et al.* 3D Bioprinting of Human Chondrocyte-laden Nanocellulose Hydrogels for Patient-specific Auricular Cartilage Regeneration. *Bioprinting*. 2016;1;22-35

- This protocol is optimized based on CELLINK™ Bioink, and may need further optimization for other bioinks. For more information, please contact: bioinkteam@cellink.com

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