

Happy Printing!

Get ready to print
living tissue

www.cellink.eu

www.roosterbio.com

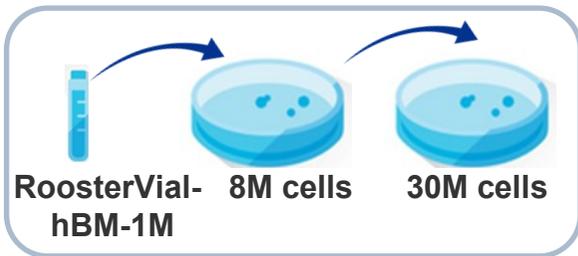


APPLICATION NOTE



CELLINK™ can be used both as a bioink as well as a suspension 3D cell culture solution. The mixture of hBM-MSCs with CELLINK™, can be directly bioprinted to your desired structure, or dispensed manually onto well-plates and cross-linked with the proprietary *Crosslinking solution*. A 5 min incubation is sufficient for fixing the bioprinted structures in a well-plate. Once crosslinked, aspirate the *Crosslinking Solution* using a pipette and replace with culture or differentiation media.

RoosterBio hBM-MSC Expansion



- Using aseptic technique, thaw RoosterVial-hBM-1M and resuspend cells in RoosterBio hMSC High Performance Media
- Seed cells onto one T225 flask and culture for 4 days
- Passage 8M cells into 4-5 T225 flasks and culture for 3-4 days
- Harvest ~30M cells and pellet by centrifuging at 1000 rpm for 5 min
- Reconstitute cells to final volume of 0.3 ml to obtain final concentration of 100M cells/ml. This is the Cell Suspension.

Mixing of CELLINK™ with hBM-MSC.

Using CELLMIXER

The CELLMIXER is used at 1:10 ratio.

For 1 mL of CELLINK™, one should prepare a corresponding cell suspension of 10 million cells in 0.1 mL. The current application note is explained for 3 mL of CELLINK™ to be mixed with 30 million RoosterBio hBM-MSCs. Simply maintain the CELLINK:cell ratio for different volumes of CELLINK.

Follow this simple 5 step procedure:

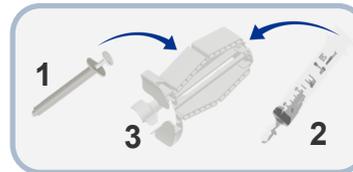
Watch the online instructional video: cellink.weebly.com/store/p4/CELLMIXER.html

STEP 1



Fill 1 with the 0.3 mL of Cell Suspension from 5. of *RoosterBio hBM-MSC Expansion*. A needle with a large diameter for low shear stress can be used to fill in the syringe.

STEP 2



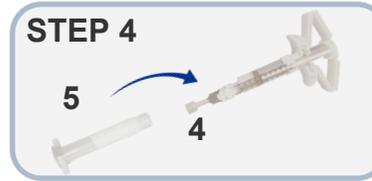
Clip 2 onto 3
Then 1 onto 3

Make sure to clip the collar of the syringe in the bottom part of the dispensing unit and the top part of the plunger in the top part of the dispensing unit.



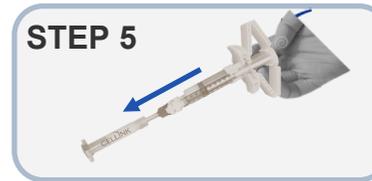
STEP 3

Screw 4 onto 1 and 2



STEP 4

Screw 5 onto 4



STEP 5

Apply gentle pressure onto the dispensing unit to mix the content of both syringes into the empty cartridge

You are now ready to print!

Printing procedure

- Insert the cartridge (5) into the printer
- Set your parameters:

-For inkjet bioprinting, we advise:

- A pressure of around 35 kPa
- A dosing distance of about 0.05 mm
- A valve-opening time of 1200 µs

- Print

Right after printing:

- Pour the *crosslinking solution* onto the final printed structure. Make sure to cover the entire structure and wait 5 min (10 min for ≥ 5 cm structures).
- Remove the crosslinking solution and replace it with the appropriate medium. Exchange medium with fresh medium after approximately 30 min.

Your tissue is now growing!