

## PRINTING PROTOCOL

# CELLINK START X

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

### Protocol aim

The aim of this protocol is to provide instructions for printing of CELLINK START X using the INKREDIBLE, INKREDIBLE+, BIO X, or BIO X6. Changing the parameters in the protocol might change printing pressure required. CELLINK START X can be photocrosslinked and is therefore used for long lasting structural support, to evaluate construct geometry and for training purposes.

### Materials needed

- CELLINK START X\*
- Conical bioprinting nozzles, 22-27G recommended\*
- BIO X\*, BIO X6\* or INKREDIBLE-series\* 3D bioprinter
- Well plate or Petri dish\*

\*The product can be purchased in the CELLINK shop at [www.cellink.com/shop](http://www.cellink.com/shop).

KEEP THE INK PROTECTED FROM LIGHT IF TRANSFERRED FROM THE ORANGE UV PROTECTED CARTRIDGES TO AVOID CROSSLINKING BEFORE PRINTING. WORK WITH 3D PRINTERS IN DARK MODE. THE PHOTOINITIATOR IS SENSITIVE TO REPEATED OR PROLONGED EXPOSURE TO HEAT.

## Protocol

This protocol can be performed with printheads and print bed at room temperature, where room temperature is between 20-25°C. CELLINK START X is liquid at temperatures below 12°C.

# 1. Preparing the ink

### MATERIAL

CELLINK START X

### DESCRIPTION

- Use CELLINK START X at room temperature. It is recommended to not use CELLINK START X below 20°C or above 30°C as the printability decrease.

Note: It is not recommended to blend cells with CELLINK START X as it is intended for use as a support or training material.

# 2. Printing

### MATERIAL

Conical bioprinting nozzles, 22-27G recommended

BIO X, BIO X6 or INKREDIBLE series bioprinter

Well plate or Petri dish

### DESCRIPTION

- Cap the cartridge with a printing nozzle of choice and place in the printhead. Connect the cartridge to the air adapter.
- For optimal printability use a layer height equal to the nozzle inner diameter at room temperature. See Figure 1 for suggested printing pressures for corresponding nozzle size and printing speed.
- Print structures onto a well plate or Petri dish. If printability is not as desired, adjust the pressure up/down by 1 kPa to extrude more/less material.

Note: If waiting more than 15 between extrusions the ink can dry in the nozzle causing it to clog. If this occurs, replace with new nozzle.

Note: Test the flow of the ink after the calibration is performed and start with a low pressure and increase stepwise.

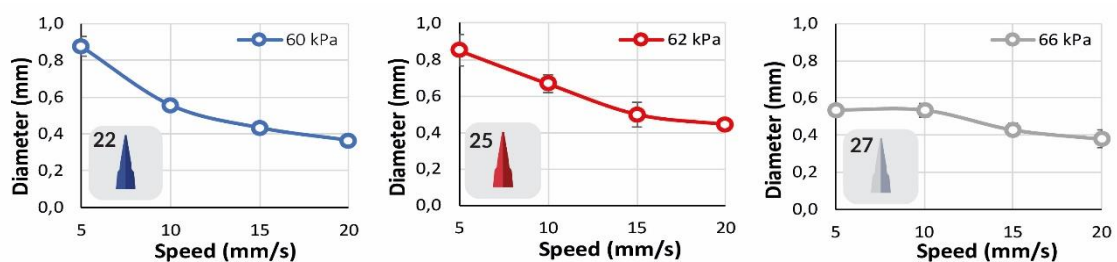


Figure 1. Resulting filament diameter when printing at different nozzle sizes, printing pressures and printing speeds.

# 3. Crosslinking

## MATERIAL

405 nm LED modules for photocuring

## DESCRIPTION

- CELLINK START X can be crosslinked with the 405 nm LED module.
- Either layer by layer or, after printing, expose CELLINK START X to 405 nm light for approximately 10-30 seconds at a layer height of 3 cm.

Note: If crosslinking is unsure add 37°C media to one printed well to validate that it doesn't dissolve.