CELLINK GelMA High C Kit

Kit Components

Item	Quantity	Storage
CELLINK GelMA High C	3 mL (10 or 50 ml available)	4-8 °C, protected from light
Sterile 12 mL Syringes	2 per 3 mL GelMA High C	Room Temperature
Nozzle Kit	1 per 3 mL GelMA High C	Room Temperature
Sterile 3 cc cartridges	2 per 3 mL GelMA High C	Room Temperature
Sterile luer-lock	1 per 3 mL GelMA High C	Room Temperature

Materials not included

Item	Quantity
Stir Bar – Sterile	1
Irgacure 2959 or LAP	200 mg
PBS	20 mL
15 mL Falcon Tube - Sterile	1
50 mL Falcon Tube - Sterile	1
Sterile 0.22 µm filter	2
Sterile Serological pipets	2

Protocol

Note: The GelMA High C is provided at 20% w/w in PBS at PH of 7.4. This protocol assumes mixing at 1:1 to achieve a 10% GelMA solution. If different final concentrations of GelMA are desired, ensure that the photoinitiator in the diluting buffer is at the right concentration to achieve the desired final concentration required for photocrosslinking.

1. Prepare 3 mL of PBS or your desired diluting buffer.
2. Mix in the desired amount of photoinitiator (PI) to achieve the diluting buffer concentration. Example concentrations for the PI solution can be found below to achieve a 10% GelMA ink and a 5% GelMA ink.

Final PI Concentration in Bioink if mixed 1:1 GelMA:Diluting buffer	Concentration in Diluting Buffer	Mass of PI for 10 ml of Diluting Buffer
0.05% (0.5 mg/mL)	0.10% (1.0 mg/mL)	3 mg
0.10% (1 mg/mL)	0.2% (2.0 mg/mL)	6 mg
0.25% (2.5 mg/mL)	0.5% (5.0 mg/mL)	15 mg

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Final PI Concentration in Bioink if mixed 1:3 GelMA:Diluting buffer	Concentration in Diluting Buffer	Mass of PI for 10 ml of Diluting Buffer
0.05% (0.5 mg/mL)	0.066% (0.66 mg/mL)	2.2 mg
0.10% (1 mg/mL)	0.133% (1.33 mg/mL)	4.4 mg
0.25% (2.5 mg/mL)	0.333% (3.33 mg/mL)	11 mg

3. Sterile filter the photoinitiator solution using the 12 mL syringe and 0.22 µm sterile filter into a sterile falcon tube.
4. Warm the GelMA High C to 37 °C to liquify.
5. Warm the Diluting Buffer to 37 °C.
6. Transfer the desired volume of GelMA High C to a 12 mL syringe using a sterile luer-lock connector.
7. Transfer the necessary volume of diluting buffer into the same syringe to achieve the desired final composition.
8. Attach a second empty syringe and mix the two precursor solutions using a dual-syringe mixing technique a minimum of 25 times back and forth.
9. Transfer the whole volume to one syringe and cap.
10. Lightly centrifuge (500 rpm) to remove air bubbles.
11. Transfer into 3 cc cartridge for bioprinting.
    a. If using GelMA for bioprinting, attach sterile luer-lock to syringe and transfer to printing cartridge. Refer to the bioprinting protocol for future steps.
    b. If using GelMA as a 3D culturing material, pipet the material into a well plate or desired mold with or without cells.
12. To crosslink the GelMA solution, please follow the GelMA crosslinking protocol

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