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Bioprinting Foods for the Future: A STEAM Lesson and Design Thinking Approach

Resource Guide

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Key Journal Articles

Pusch, K., Hinton, T. J., & Feinberg, A. W. (2018). Large volume syringe pump extruder for desktop 3D printers. *HardwareX*, *3*, 49-61. https://doi.org/10.1016/j.ohx.2018.02.001

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Hardware Article

Large volume syringe pump extruder for desktop 3D printers

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ABSTRACT

Syringe pump extruders are required for a wide range of 3D printing applications, including bioprinting, embedded printing, and food printing. However, the mass of the syringe becomes a major challenge for most printing platforms, requiring compromises in speed, resolution and/or volume. To address these issues, we have designed a syringe pump large volume extruder (LVE) that is compatible with low-cost, open source 3D printers, and herein demonstrate its performance on a PrintrBot Simple Metal. Key aspects of the LVE include: (1) it is open source and compatible with open source hardware and software, making it inexpensive and widely accessible to the 3D printing community, (2) it utilizes a standard 60 mL syringe as its ink reservoir, effectively increasing print volume of the average bioprinter, (3) it is capable of retraction and high speed movements, and (4) it can print fluids using nozzle diameters as small as 100 µm, enabling the printing of complex shapes/objects when used in conjunction with the freeform reversible embedding of suspended hydrogels (FRESH) 3D printing method. Printing performance of the LVE is demonstrated by utilizing alginate as a model biomaterial ink to fabricate parametric CAD models and standard calibration objects.

Notes

This paper was used to support the creation of the 3D-printed syringe pump that moves the printing material through the extruder, as well as for preparing the printing material and support bath. It also has a bill of materials for what is needed to build the pump. It does not provide a detailed guide on how to do the firmware edits. We had to experiment with this to get it right. A saved version of the edited firmware is attached to this document (see Useful Links & Files).

To create the gelatin support bath, we used Mckenzies Gelatin sourced from Woolworths. We formed 250 mL gels by heating the gelatin in 0.1 M calcium chloride in the microwave. The gels were set in the fridge overnight. The next day, they were blended using a standard stick blender while the mixing vessel was placed over an ice bath. The ice bath is needed to prevent the gelatin from dissolving during blending from the heat generated by the friction of the stick blender blades. The challenge here is to obtain small enough particles of undissolved gel. The gelatin needs to be the consistency of purée. Following this, a centrifuge was used to spin down 50 mL aliquots of the gelatin slurry. The supernatant was discarded, and fresh 0.1 M calcium chloride was used to resuspend the gelatin particles. This process was repeated for each 50 mL aliquot. These resuspended aliquots were stored in the fridge until needed. Prior to printing, they underwent a final spin down with supernatant discarded. The gelatin mixture was then transferred to a 100 mL beaker ready for printing.

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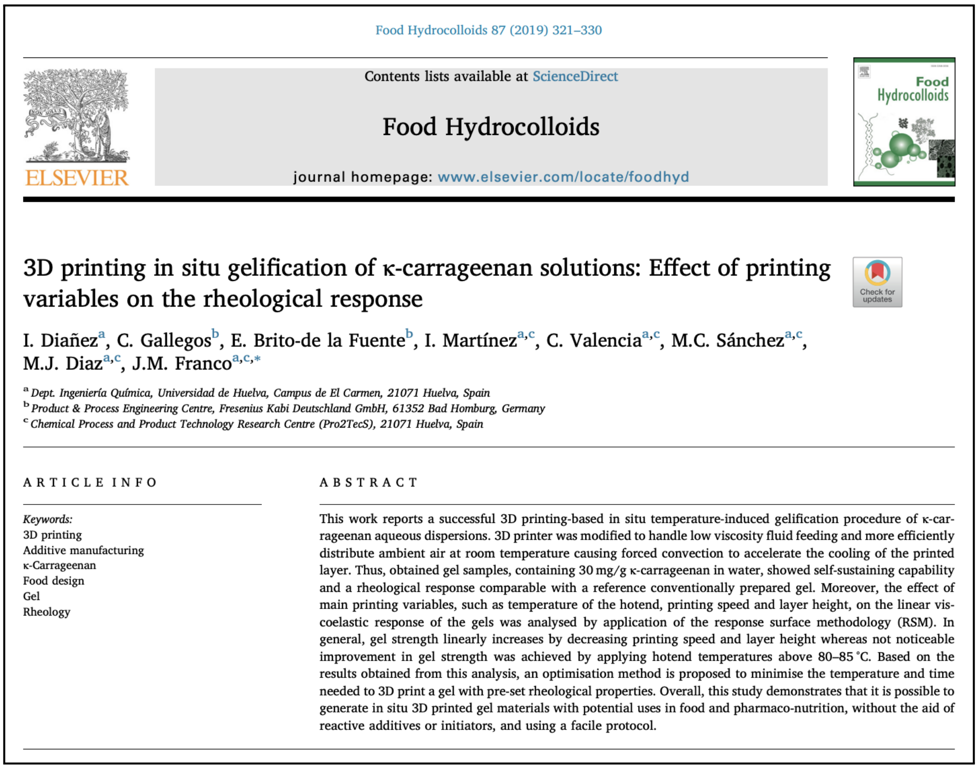
Diañez, I., Gallegos, C., Brito-de la Fuente, E., Martínez, I., Valencia, C., Sánchez, M. C., Diaz, M. J., & Franco, J. M. (2019). 3D printing in situ gelification of κ-carrageenan solutions: Effect of printing variables on the rheological response. *Food Hydrocolloids*, *87*, 321–330. https://doi.org/10.1016/j.foodhyd.2018.08.010


Food Hydrocolloids 87 (2019) 321–330

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3D printing in situ gelification of κ-carrageenan solutions: Effect of printing variables on the rheological response

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ABSTRACT

This work reports a successful 3D printing-based in situ temperature-induced gelification procedure of κ-carrageenan aqueous dispersions. 3D printer was modified to handle low viscosity fluid feeding and more efficiently distribute ambient air at room temperature causing forced convection to accelerate the cooling of the printed layer. Thus, obtained gel samples, containing 30 mg/g κ-carrageenan in water, showed self-sustaining capability and a rheological response comparable with a reference conventionally prepared gel. Moreover, the effect of main printing variables, such as temperature of the hotend, printing speed and layer height, on the linear viscoelastic response of the gels was analysed by application of the response surface methodology (RSM). In general, gel strength linearly increases by decreasing printing speed and layer height whereas not noticeable improvement in gel strength was achieved by applying hotend temperatures above 80–85 °C. Based on the results obtained from this analysis, an optimisation method is proposed to minimise the temperature and time needed to 3D print a gel with pre-set rheological properties. Overall, this study demonstrates that it is possible to generate in situ 3D printed gel materials with potential uses in food and pharmaco-nutrition, without the aid of reactive additives or initiators, and using a facile protocol.

Notes

This paper covers the use of k-carrageenan as a printing material. While we used the pump from the previous paper in combination with this pump, we were unsuccessful in achieving fast gelification. This issue could be due to the k-carrageenan that was sourced, inadequate cooling, or other inconsistencies in the tap water used between the authors and Monash Tech School.

To implement the changes of this paper to be used with the pump from the previous device, we didn't need to adjust any of the firmware on the printer. Instead, changes were made directly to the slicing software. These slicing profiles have been linked below to allow you to experiment with them. In addition, we needed to swap the hot end assembly back in and remove our extrusion needle from the bioprinters from the previous setup for the sodium alginate/gelatine method.

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Materials List & Pricing

Materials Needed for Large Volume Syringe Pump Extruder

All materials can be easily sourced locally except for those listed by McMaster-Carr. While the total price list may come to $150 AUD, this figure is for individual parts. Generally, you will have to buy parts in packs (e.g., ball bearings, M8 nuts) to get the minimum number needed - so prices may differ.

Item Name	Material Type	Source	No. of Units
Double Shielded Ball Bearings 608-ZZ	Steel	RS Components	2
M8 × 2.4 mm Steel Hex Nuts	Steel	RS Components	4
M8 Brass Hex Nuts	Brass	RS Components	2
M8 – 1.25 × 1 m, Threaded Rod, Stainless Steel, 304	Steel	RS Components	1
M3 × 10 mm Socket Head Screws (i.e. Hex Bolts)	Steel	RS Components	10
M3 × 16 mm Socket Head Screws (i.e. Hex Bolts)	Steel	RS Components	4
M3 × 8 mm Socket Head Screws (i.e. Hex Bolts)	Steel	RS Components	2
M3 × 45 mm Partially-Threaded Socket Head Screws (i.e. Hex Bolts)	Steel	RS Components	2
M3 Hex Nuts	Steel	RS Components	16
60 mL Luer-Lock BD Syringe	Polypropylene	Pacific Labs	1
Standard Polyurethane Tubing, 1/8″ OD, 1/16″ ID	Polyurethane	RS Components	1
18-Gauge, 4″ Long Blunt-Tip Needle With Female Luer-Lock Connection	Stainless Steel	McMaster-Carr	1
Male Luer to 1/16″ Tubing Barb	Polypropylene	McMaster-Carr	1
Steel Reinforced Epoxy	Epoxy	Bunnings	1
17-Gauge Luer-Lock Needle	Stainless Steel	McMaster-Carr	1
26-Gauge Luer-Lock Needle	Stainless Steel	McMaster-Carr	1
Loctite 0.2 fl. oz. Threadlocker Blue 242	Glue/Epoxy	Bunnings	1

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Useful Links & Files

For Both Setups/Methods (Sodium Alginate/Gelatine & k-Carrageenan)
Video guides detailing assembly of 3D-printed syringe pump and Slic3r software setup https://www.youtube.com/channel/UCEwYrE9pROR0kinbDDSCFWg/videos
Article detailing 3D bioprinter hardware/software conversion processes and algae bioink preparation https://www.hackster.io/dynamic-ink/open-affordable-3d-bioprinting-14d68b
Firmware file for Creality Ender 3 conversion to Ender 3 bioprinter https://drive.google.com/file/d/1OAlez-e3D07s51viyic42J-5L5JFuXCv/view?usp=sharing
Pronterface program for controlling bioprinter directly from computer while connected via USB https://www.pronterface.com/
Index of G-code commands https://marlinfw.org/meta/gcode/

For Bioprinting with Sodium Alginate/Gelatine (Method 1)
Slic3r program for slicing STL files https://slic3r.org/
Video guide detailing setup of Slic3r program for printing with sodium alginate https://www.youtube.com/watch?v=ASdhlE9vMF8

For Bioprinting with k-Carrageenan (Method 2)
PrusaSlicer program for slicing STL files https://www.prusa3d.com/prusaslicer/
Bioprinting slicer settings and profile (for PrusaSlicer program only) https://drive.google.com/file/d/1lY_bw88ZMrYNcU2hjIl48kRYmLyw_OwE/view?usp=sharing

Other Links
List of commercially available food 3D printers https://all3dp.com/1/best-3d-food-printer/
Scents that can be purchased for design thinking activity https://www.muaustralia.com/fragrance/demeter-fragrance-library/

Uploading Your Firmware to Your Ender 3 3D Printer

1. Download the firmware file for Creality Ender 3 from the link above
2. Copy the BIN file
3. Plug in your mainboard's Micro SD card into your device
4. Clear (delete or move) everything on it
5. Paste the BIN file onto the Micro SD card
6. Insert the Micro SD card into the mainboard's Micro SD card slot
7. Make sure your Micro SD card is securely plugged in and oriented the right way
8. Make sure all cables and electrical configurations are connected and secured
9. Power on the printer by plugging the power cable back into your printer and turning the power switch back on
10. Wait until you see the home screen on the LCD, then navigate the user interface to find and click the "Restore Defaults" button

Watch this video from the set time point for a walk-through of the instructions. You do not need to edit any firmware; this has been done, so you will only need to put it on a Micro SD card and upload it to your printer.