

3D BIOPRINTING OF VASCULAR CONSTRUCTS

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Introduction



In the additive manufacturing (AM) used for the production of tissues and organs, more commonly known as 3D printing, various types of materials can be used for the bio-ink depending on the type of tissue and different cell types. Several properties of bioink influence the printability including gelation, viscosity, nozzle gauge, shear stress, network properties, and fabrication time. In this project, the construction of vascular structures is aimed by printing of alginate laden cells into a Pluronic® F-127 supporting bath. This work mainly focused on the optimization of the conditions for Pluronic® F-127 for bioprinting. In addition, a model and algorithm to successfully and efficiently print an artery structure with Rhino 3D was developed. Based on optimization studies, it can be said that the storage modulus of the Pluronic® F-127, and the sol-gel transition temperature were found to depend on the concentration of polymer.

Appropriate Conditions for 3D Bioprinting of Vascular Constructs

2.1. Materials and methods

2.1.1. Selection of hydrogel for supporting bath

Two different hydrogels were investigated. One of them is gelatin. Gelatin is liquid in room temperature and gel in the cold temperature. Thus, second hydrogel which is called Pluronic® F-127, was examined. Pluronic® F-127 is liquid in cold temperature and gel in the room temperature.

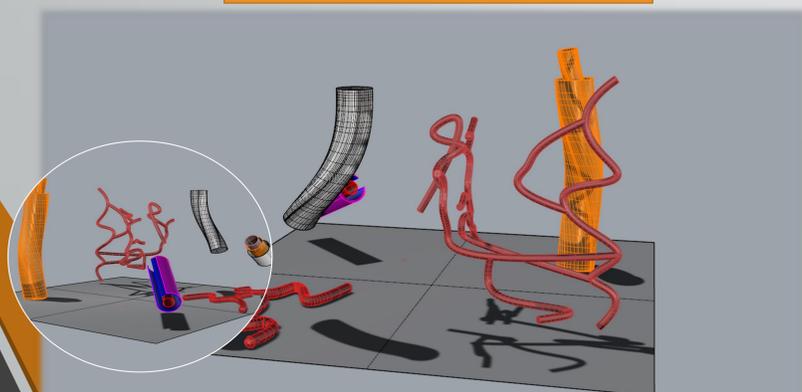
2.1.2 Preparation and testing of different concentrations of Pluronic® F-127 hydrogels

Concentration of Pluronic® F-127 (%)	Volume of CaCl ₂ (ml)	Quantity of Pluronic® F-127 (g)
•16	•50	•8
•18	•50	•9
•20	•50	•10
•25	•50	•12,5
•40	•50	•20

2.1.3. Rheology experiment of 16% and 25% concentrations of Pluronic® F-127

The gel was waited 5 minutes at 4 C°. After that temperature was increased as 5 degrees per minute. Then, gel was waited 30 minutes at 37 C°.

2.1.4. Create a model



We used a program called Rhino 3D to sculpt and model structures that can be used as placeholders for artery structures. After the modelling we moved on to a built-in coding system called Rhino Script that allowed us to take 3D models as inputs and transit them to the 3D printer for the required process. To achieve this we started to design an algorithm to effectively build a bridge between Rhino 3D and the 3D printer.

2.1.5 Forming an algorithm

We started with a structure that checks X/Y plane to find intersections. While designing this we assumed the shape of the artery to be cylinder and detected certain intersection points that we named as inner and outer circles.

Then we took the saved data and compared them within each other to determine the upper and lower ends of the inner/outer circles. Using these recorded points we determine the WIDTH, RADIUS and CENTER of the 3d structure. In the last and recurring step we determine the inputs we get from the user and the implication of this data.

Next was the incorporation of the 3D printer attributes, such as the radius of syringe and rate of printing. Both are taken as variables. The radius of the syringe is used alongside the width, we divide it in a way such that it prints in spirals and starts/ stops R/2 units away from the designed coordinates.

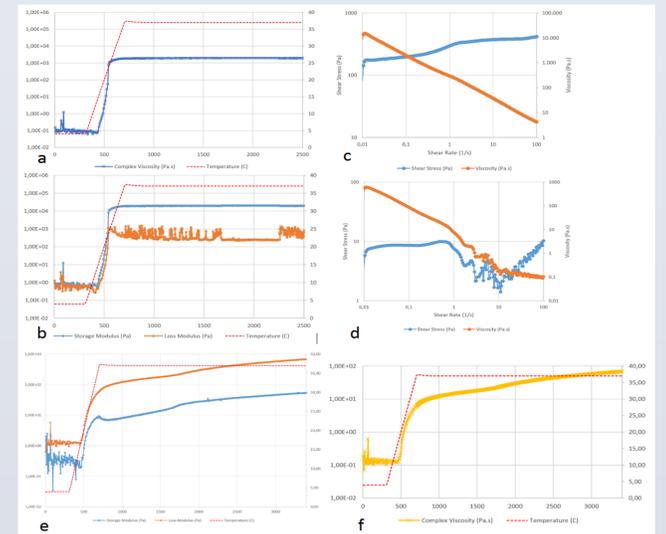
As we are trying to make the algorithm work properly and effectively, we are also trying to find ways to make the process smoother and less demanding on the hardware we are using.



Results and Conclusions

Rheological analysis results of 16% and 25% concentrations of Pluronic® F-127.

(a, b) Viscosity and storage modulus of 25% Pluronic® F-127 as a function of temperature. (c) Shear stress and viscosity of 25% Pluronic® F-127 as a function of shear rate. (e, f) Viscosity and storage modulus of 16% Pluronic® F-127 as a function of temperature. (d) Shear stress and viscosity of 16% Pluronic® F-127 as a function of shear rate.



The result of 18% concentration.



The result of 16% concentration.

In the consequence, the rheological properties of Pluronic with 16% and 25% concentrations were tested and observed to see physiological changes. According to shear stress-shear rate graph, the concentration of 25% causes a linear behavior in the graph and that line breaks under shear stress which indicates that this concentration is not suitable for using. We can assume that the interactions between the formed micelles are too weak and do not contribute in forming a network showing yield behavior for 16%. Based on both previous works and this experiment, it can be said that the storage modulus of the F127 solution, and the sol-gel transition temperature were found to depend on the concentration of polymer (Jiang et al., 2008).

Chosen concentrations were tested and observed by external responses such as observing any occurrence of break-through of the gel or fiber structures. Pluronic F127 with 16% and 18% concentrations were tested. The resulted mixture with 16% concentration wasn't stiff enough to hold the alginate in it as where as 18% showed a stiffer structure while it was in gel form. The subsequent testing concentrations were 20%, 25%, and 40%, as all of them resulted in having tearing of the gel when the alginate was printed in; and the wanted fiber-like structure hadn't occurred. Latterly, testing of the Pluronic with 21% and 22% concentrations were decided on, of which results showed that fiber-like structures couldn't be obtained due to the tearing of the gel during printing. Moving from that conclusions, it was decided to keep trying lower concentrations, but by changing other parameters such as temperature. It seemed to have been reached its gel form as where as it regained fluidic structure when it's kept in room temperature due to its thermo-responsivity; which resulted in a decision of printing in Pluronic with concentrations of 16% and 18% while they were kept at 37°C with a heater around them by using the printer other than manually.

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