Temperature Controlled 3D Bio-printing Using Bio-polymeric Ink

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Abstract

Bio-Printing 3D organs, tissues & scaffolds is advancing everyday, with various methods being introduces to mimic a perfect working organ or tissue which can be functional at the same time. We have developed a method to print bio-polymer in liquid state using them as bio-inks to prints scaffolds, while maintaining their temperatures. The design is cheap and efficient to print 3d scaffolds. Our Bio-Printer is also aided with a robotic arm to induce any bio-active ingredient in the 3d structures once it is printed without deforming or altering the structure of the scaffold. The main printer can be paused to allow the robotic arm to continue embedding and make repairs to the scaffold from any angle.

Introduction

3d Bio-printing is playing an important role in tissue engineering by printing 3d bio-structures for research purposes. To design and develop an ECM with a bio-active element (for cell growth) and to maintain the structure properties after printing are all achieved by the right combination of Bio-inks used with the best possible extrusion technique in 3d printing.

Our design introduces a syringe pump with temperature controlled assembly to print bio-polymers like PLA, PCL, PVA, hydrogel etc. in liquid state. The robotic arms provides flexibility in injecting bio-materials at different angles without interfering with the primary structure . We have produced great structural support for the scaffolds by viscous biocompatible polymer.

Software

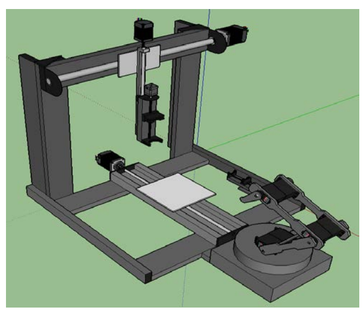
3d model of biostructures or any other object can be easily modelled on any CAD software like PROE, SOLIDWORKS, AutoCAD. We have used Fusion 360 Cad software to design our models due to its feature of directly integrating with the 3dprinter and saving the (.stl) files in cloud, to be easily accessed from anywhere.

The second software we have used is Reptier Host which has slicing software integrated in it with printer control settings. Cura and Slicer software are already present in Reptier Host which slice the (.stl) 3d model file in different layers and show visual representation of each layer during printing.

Device Design

Bio-printer

A.



B.

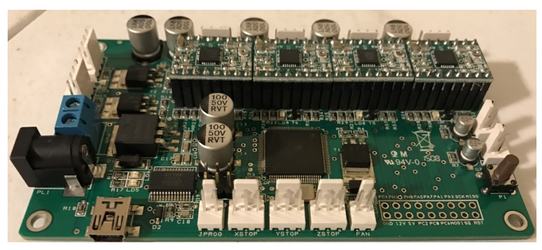
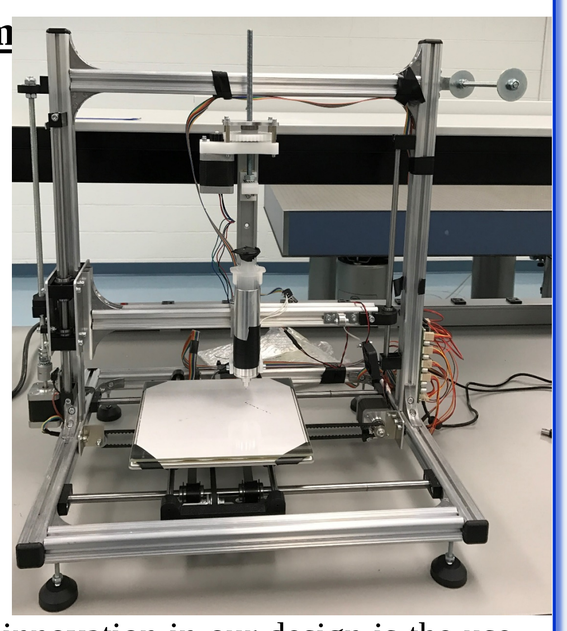


Figure 1. (A) Full bio-printer design. (B) 3D printer control system, featuring stepper motor control chips, Velleman board, and Arduino Mega Micro Controller.

The printer is a modified version of simple 3 axis 3d printer with the regular extruder replaced by a syringe extruder with a temperature controlled assembly. The printer is controlled by Velleman board which has an Atmega 2560 microprocessor and motor circuit bridge embedded to control the motor and define their movements. The extrusion needle is currently 0.9 mm (900um) which will be reduced to achieve more fine prints.

3 Axis Printer & Robotic Arm



▲Figure 3. **3D Bio-Printer**

Three axis printer prototype of the bio-printer, featuring six mechanical end stops, a syringe pump with temperature controlled assembly. The syringe contains a 30 gauge needle, which when combined with 0.9º stepper motors and three M5 fine pitched threaded rods, allows for as low as 10µm precision in all dimensions and extrusion.

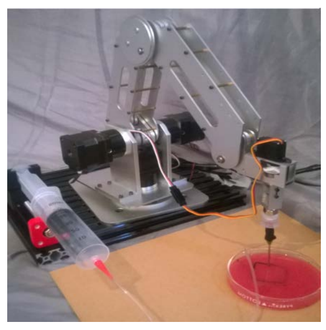


Figure 4. Precision printing arm.

The key innovation in our design is the use of a high precision robotic arm in union with a rotating printing stage. This allows for simultaneous injection of bio-ink, growth factors or hydrogels from 64800 different angels while the main structure is being constructed or after for structural repairs.

Printing Materials

Polylactic acid:

Polymer melts are too viscous to print via inkjet and therefore, either a dilute solution is used or reactive components are printed and cured in a post-processing step. For polylactic acid (PLA) grades which are processed via FDM, 1,4- dioxane proved to be a good solvent and simple surface finishing by dipping or polishing has been explored by the Reprap community. Also, 1,4-dioxane is often used as solvent when creating PLA phase-separated scaffolds. Its physical properties are very convenient for inkjet printing due to its high boiling point of 101 ˚C and its compatibility with the used cartridge and print head. In our study, we prepared PLA solvent inks with different concentrations by dissolving PLA in 1,4-dioxane followed by incorporation of silver(Ag) nanoparticles(Nps) and investigated the printability of this material and scaffold generated for its Bio-medical application[1].

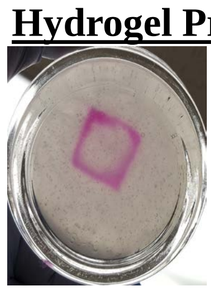
Scaffolding and Bioink:

Biocompatible polymer based materials were used to form the scaffold and bioink using the following procedure:

- Slices of the PLA filament were dissolved in 1,4-dioxane at different polymer concentrations.
- The polymer solution was stirred for 2 h at 65 ˚C and then was passed through a syringe filter (pore size 5 µm) to eliminate insoluble particles.
- Ag Nps were added to this polymeric solution and scaffolds were printed with and without the Nps.
- The degradation rate of the produced scaffold is 3 months.[2]

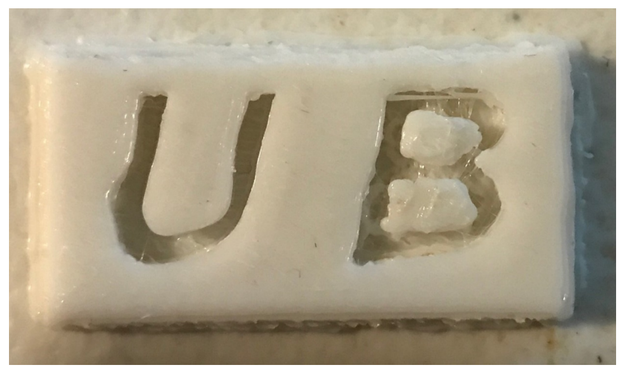
Results

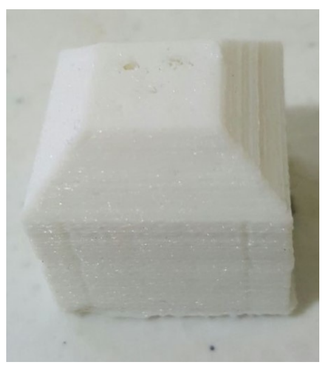
Hydrogel Printing



▲Figure 5. (A) 3D printed square using stained 10% PCL. (B) Final solidified product.[1]

PLA Printing:





▲Figure 6. (A) PLA printed UB Logo. (B) PLA printed Cube

Conclusion

We were able to manufacture a 3D bio-printer capable of printing synthetic prototypes of biological structures with bio-inks under a controlled temperature. In the future, we plan to expand the testing capabilities and methods to produce more elaborate and complex microstructural scaffolds; such as highly fragile vascular networks for the tissue engineering industry. Furthermore, we plan to use different biocompatible materials that can be directly embedded making a more favorable media for the cellularization of the fabricated scaffolds. We also want to extend the extruding capabilities to extrude multiple inks at same time to induce certain enzymes and growth factors at the printing time.

References

[1] Kimberly A. Homan et.al Bioprinting of 3D Convoluted Renal Proximal Tubules on Perfusable Chips. Sci. Reports 6:34845 DOI: 10.1038/srep34845.

[2] Bual, G. S.; Kumar, P. Manuf. Sci. Technol. 2014, 2, 51

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