Computer Aided Tissue Engineering for Modeling and Design of Novel Tissue Scaffolds

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ABSTRACT

Computer-aided tissue engineering (CATE) integrates advances of multi-disciplinary fields of Biology, Biomedical Engineering, Information Technology, and modern Design and Manufacturing. Application of CATE to the design and fabrication of tissue scaffolds can facilitate the exploration of many novel ideas of incorporating biomimetic and biological features into the scaffold design. This paper presents some of the salient applications of CATE, particularly in the modeling and design of scaffolds with controlled internal and external architecture; with vascular channels of different sizes; with modular and interconnecting subunits; with multi-layered heterogeneous dense and compact regions; and the scaffolds with designed artificial chambers for drug delivery, embedded growth factors and other sophisticated features.

Keywords: Computer-aided tissue engineering, Tissue scaffold, Solid freeform fabrication

1. INTRODUCTION

Tissue Engineering, the science and engineering of creating functional tissues and organs for transplantation, integrates a variety of scientific and engineering disciplines to produce physiologic "replacement parts" for the development of viable substitutes which restore, maintain or improve the function of human tissues [1, 2]. In the success of tissue engineering, three-dimensional (3D) scaffolds play important roles as extra-cellular matrices onto which cells can attach, grow, and form new tissues. Modeling, design and fabrication of tissue scaffolds to meet multiple biological and biophysical requirements is always a difficult task in the regenerative tissue engineering. This is particularly true when design load bearing scaffolds for bone and cartilage tissue application. In general, this type of scaffolds usually have intricate architecture, porosity, pore size and shape, and interconnectivity in order to provide the needed structural integrity, strength, transport, and ideal micro-environment for cell and tissue ingrowth [3-5]. In addition, thus designed scaffolds often can only be fabricated through advanced manufacturing techniques, such as solid freeform fabrication (SFF) to manufacture complex structural architectures [6-7]. Among available SFF techniques, the precision extruding deposition seems to be the most promising one for advanced scaffold fabrication due to its versatility of using different scaffolding materials, possibility of manufacturing scaffolds in a cell-friendly environment, and feasibility of controlled drop-on-demand high precision deposition [8-10].

Computer-aided tissue engineering (CATE) advances modeling, design and fabrication of tissue scaffolds [11]. For example, CATE can apply biomimetic design approach to introduce multiple biological and biophysical requirements into the scaffold design [12]. CATE can also integrate both biomimetic and non-biomimetic features into the scaffold modeling database to form high fidelity and smart scaffolds. Biomimetic features can be based upon real anatomical data regenerated from CT/MRI images, or can be created purely within a CAD environment – for example, channels and porous structures. Non-biomimetic feature do not imitate nature but can be designed as drug storage chambers, mechanical elements, and attachment interfaces for tubes, sensors, electronics, and other devices. The objective of this paper is to present our recent study on applying CATE for advanced tissue scaffold modeling and design, including an overview on recent development of CATE, a CATE based modeling for representation of heterogeneous biological tissue structure, and the use of CATE to introduce various design intents of internal and external architecture, porosity, interconnectivity, mechanical properties, vascularization, and drug/growth factor delivery into the scaffold design.

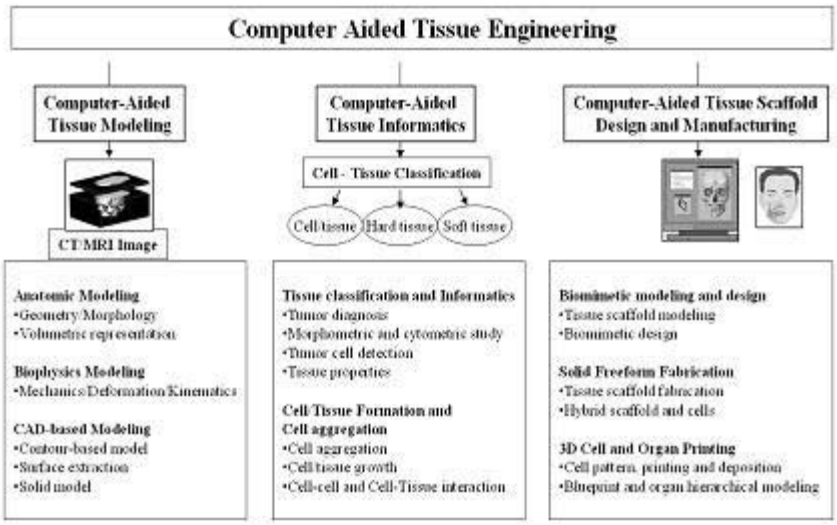


Fig. 1. Overview of Computer Aided Tissue Engineering [11]

2. OVERVIEW OF COMPUTER AIDED TISSUE ENGINEERING

Utilization of computer-aided technologies in tissue engineering research and development has evolved a development of a new field of Computer-Aided Tissue Engineering (CATE). CATE integrates advances in Biology, Biomedical Engineering, Information Technology, and modern Design and Manufacturing to Tissue Engineering application. Specifically, it applies enabling computer-aided technologies, including computer-aided design (CAD), medical image processing, computer-aided manufacturing (CAM), and solid freeform fabrication (SFF) for multi-scale biological modeling, biophysical analysis and simulation, and design and manufacturing of tissue and organ substitutes. In a broad definition, CATE embraces three major categories in tissue engineering: 1) computer-aided tissue modeling, including 3D anatomic visualization, 3D reconstruction and CAD-based tissue modeling; 2) computer-aided tissue informatics, including computer-aided tissue classification and application for tissue identification and characterization at different tissue hierarchical levels; and 3) computer-aided tissue scaffold design and manufacturing, including scaffold modeling and design, solid freeform fabrication of tissue scaffolds, bio-blueprint modeling for 3D cell and organ printing. An outline of CATE is presented in Figure 1. Detail presentation on major issues in each category can be found in authors' early reports [11, 12].

3. TISSUE FEATURE PRIMITIVES

3.1 FEATURE PRIMITIVE BASED MODELING

To model a heterogeneous tissue in a CAD environment, we developed a feature-primitive based Reasoning Boolean Operation (RBO) [13]. In contrast to conventional Boolean operations (for homogeneous solid objects), RBO manipulates multi-volume feature primitives through two distinctive operations: 1) merging; and 2) extracting. The feature primitives are defined on a design-by-features paradigm which is commonly used by most CAD systems, with tissue morphological, anatomical and biological characteristics being introduced as material, structural, or biological attributes. Once the individual tissue primitive is selected, the merging operation identifies the attributes assigned to the feature primitives and compares them to decide whether they are identical or need to be merged. Extracting then follows to generate the needed intersecting surfaces, edges, and/or the splitting of volumes for merged primitives to form heterogeneous tissue model according to the distinctive attribute identification. A schematic illustration for constructing a heterogeneous object consisting of two-volume feature primitives A(M_A) and B(M_B) is shown in Figure 2.

Figure 2 schematically shows how one can apply a series of reasoning Boolean operation to construct a small heterogeneous structure model, called a unit cell model, based on feature primitives A (matrix) and B (plain weave pattern). By analogous composite applications, we consider a unit cell as a small volumetric element which characterizes tissue heterogeneity, feature primitives, and effective tissue structural properties. Unit cell representative method has been widely used in composite design and analysis. However, in contrast to conventional

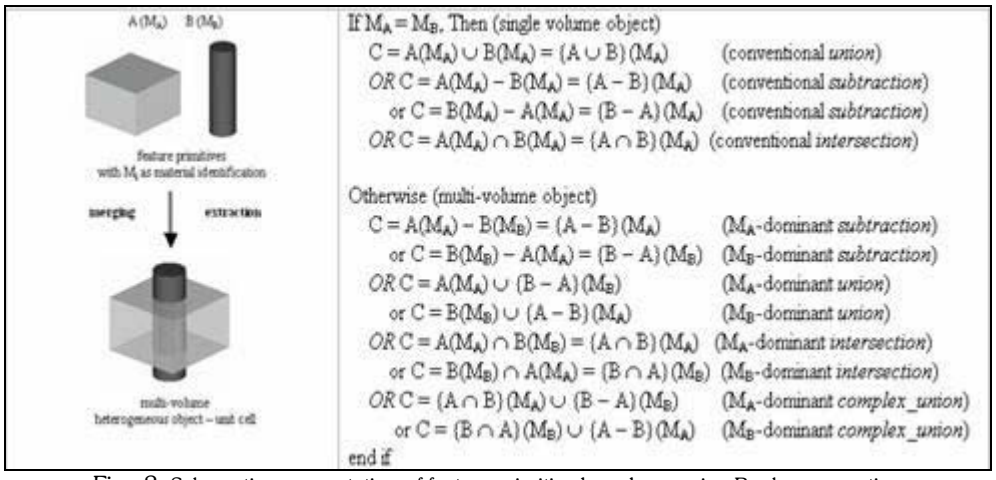
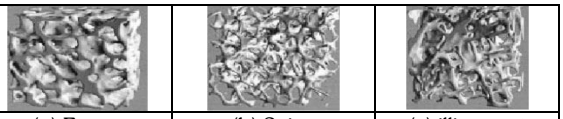


Fig. 2. Schematic representation of feature primitive based reasoning Boolean operation

composite materials, we need many different customized unit cells in designing a tissue substitute in order to adopt the true tissue anatomical and structural heterogeneity. In addition, the internal topologies for most unit cells in tissue scaffold application are contiguous with open cell porosity, in contrast to commonly used continuous fiber-reinforced composites.

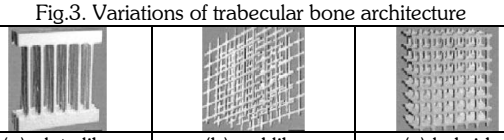
3.2 REPRESENTATION

Biological tissues are inherently heterogeneous structures. At the macrostructure level, tissue exhibits both morphological and mechanical heterogeneity and varies greatly at different anatomical and structural levels. For example, Figure 3 [14] shows three different types of trabecular architectures as found at different anatomical sites in the human skeleton. In using feature primitive based modeling approach, these architectures can be analogized by three different types of feature primitives: plate-like primitive (for femur), rod-like primitive (for spine), and hybrid primitive (for iliac crest). These analogies are shown in Figure 4.



(a) Femur	(b) Spine	(c) illiac crest

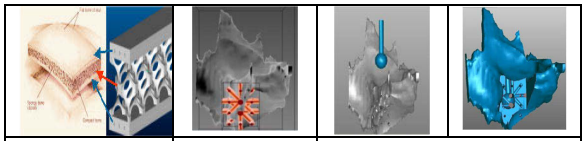
Fig.3. Variations of trabecular bone architecture



(a) plate like	(b) rod like	(c) hybrid

Fig. 4. Associated feature primitives

To biomimic the natural morphologies of bone through the use of CT and CT imagery, 3D reconstruction, and modeling techniques, one can further design customized feature primitives [15] for specific tissue structures, morphologies, and functional requirements. For example, a vascular tree system and non-biological drug delivery primitive as shown in Figure 5 can all be designed and incorporated into the scaffold system. A set of feature primitives that are represented by the CAD and parametric based model structures can be generated according to different tissue internal architectures, designed topologies, pore size, shape, porosity volume faction, and vascular/drug delivery network. The process of image-based 3D reconstruction from CT and MRI, the reverse engineering to develop NURBS based bio-CAD model, and the reasoning Boolean algebra for heterogeneous primitive operations defined in the CATE paradigm has laid a critically important foundation for integrating both biological tissue and non-biological artificial elements, such as syringes, drugs, tubes, sensors, electronics, and nano- or micro-scale bio-devices (represented by the feature primitives) for next generation “smart” and “functional” scaffolds.



(a) Customized feature primitives for cranial tissue	(b) scaffold with vascular tree primitives	(c) Scaffold with drug delivery primitives

Fig. 5. Representation of scaffolds with different feature primitives

Using feature primitive approach, each primitive discrete volume can be represented by a specific design feature, such as different internal architecture patterns used in common tissue scaffold design, for example, the standard weave, braided, and knit geometric feature of textile fiber patterns can be used as scaffold architectures or muscular pattern in soft tissues. Enabling computer-aided technology can then be applied to develop a CAD-based model based on information provided from the feature primitives, such as desirable feature patterns and architectures, desirable pores, pore sizes and shapes, and its distribution in the scaffold internal structure so that the required biophysical and biological design constraints can be met. Thus heterogeneous scaffold modeling database could serve as a central repository for scaffold design, analysis, and fabrication. Samples of thus generated CAD-based unit cells based on different feature primitive patterns are presented in Figure 6. Each unit cell in the figure is intended to be designed with different characteristics based on using different scaffolding material, feature primitive pattern, and the spatial distribution of scaffolding material to form unit cell internal architecture for porosity and pore interconnectivity considerations. With an appropriate selection of unit cells, one can design a customized heterogeneous tissue scaffold by tailoring unit cell properties, for example, using different feature patterns to design a specific porosity geometry (for pore size and shape), arranging feature patterns in a specific 3D architecture to form a preferable pore distribution and interconnectivity (for cell growing and proliferation), and analyzing or simulating to verify if the designed model meets the scaffold strength and stability requirements.

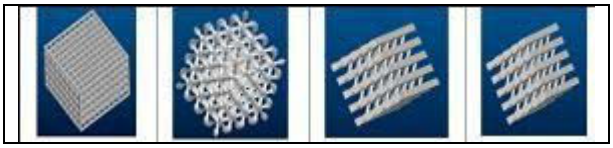


Fig. 6. Samples of unit cells generated from different feature primitives

4. APPLICATIONS

4.1 CONTROLLED INTERNAL AND EXTERNAL ARCHITECTURE

By reverse engineering CT data with medical imaging reconstruction software (MIMICS [16]) and enabling CAD, one can design a tissue implant with controlled external and internal architecture. In the following hypothetical case, we present how a seeded implant would be designed and used for repairing a skull defect after a tumor had been removed.

To create the external structure of the implant we took the CT data, and used the 3D-reconstruction tools in MIMICS to isolate the tumor (Figure 7a). The STL file for the skull and the STL file for the tumor were then exported from MIMICS and imported into Geomagic [17]. Within Geomagic we used a mirroring operation to mirror the tumor from the right side of the skull to the left side, i.e. the healthy side (Figure 7b). We then used the Boolean intersection between the tumor and skull to generate a bone implant structure. We then mirrored the implant back across the plane (Figure 7c, 7d) to complete the final external architecture of the implant (Figure 7e). Through this procedure, we essentially replaced the void created by the removal of the tumor, with a mirrored copy of the normal bone structure. Using this method, we were able to create a complex structure that matched the symmetry of the patient's skull.

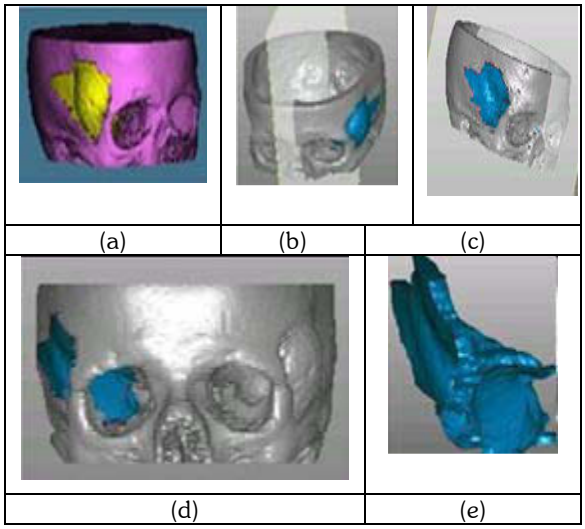
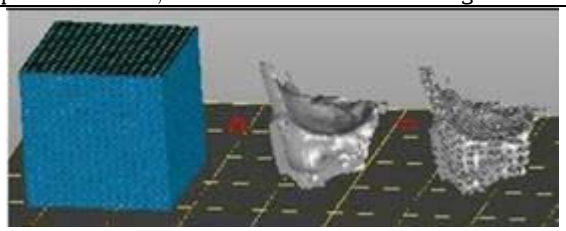


Fig. 7. Steps in creating the external architecture of a tissue implant by using mirroring and Boolean operations to replace the defect site after tumor removal

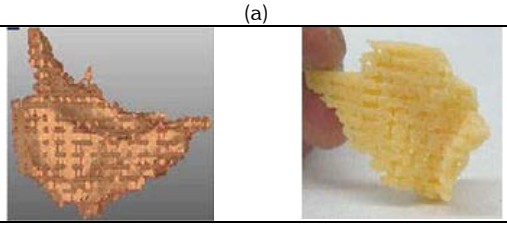
After defining the external shape of the implant, we defined the internal, porous structure. Using Pro/Engineer [18], we designed a scaffold with approximately 70% porosity and 700 μm x 700 μm channels in an 80 mm x 80 mm x 80 mm cube. To create the implant, we used a Boolean intersection between the scaffold and implant structures within Geomagic (Figure 8a) to create the final result (Figure 8b). The finished implant design was then exported to an STL file and a three-dimensional printed prototype is shown in Figure 8c.

4.2 VASCULARIZED MODULAR 3D SCAFFOLD

Vascularity is a major issue in tissue scaffold design, so a scaffold with a fully interconnected "vascular" channel system was created using CAD. In the following hypothetical case, two modular scaffolds designs were made that incorporated vessels of different sizes that followed a geometrically scaling pattern. The first scaffold (Figure 9a) had channels with the following dimensions of length and width

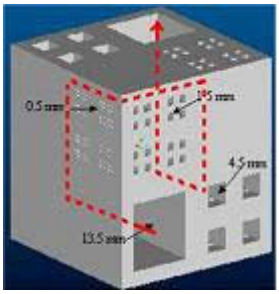


(a)

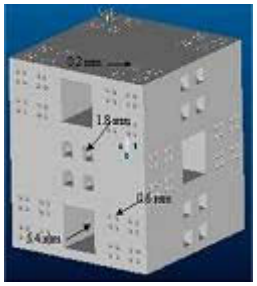


(b) (c)

Figure 8 (a) creation of internal structure using Boolean intersection, (b) completed model with controlled internal and external architecture, (c) rapid prototyped implant for skull defect repair



(a)



(b)

Figure 9 Modular scaffold with fully interconnected channel system, first design showing flow path, second design

going from smallest to largest: 0.5 mm (smallest vessel), 1.5 mm (small vessel), 4.5 mm (medium vessel), and 13.5 mm (large vessel). The dimensions of a module were 80 mm x 80 mm x 80 mm with an overall porosity of 40%. Porosity could be easily changed by adjusting the number and size of the channels. The second design (Figure 9b) had 0.2 mm, 0.6 mm, 1.8 mm, and 5.4 mm channels. In addition, the geometry of the intersecting channels were designed to ensure that the vessels only intersected with the adjacent size channels (Figure 9a), thus fluid would only flow from the large channels into the medium channels into the small channels into the smallest channels, and then vice versa. This would be roughly analogous to the pattern of artery – arteriole – capillary – venule – vein.

The modular design of the scaffold allows it to be interconnected like building blocks as shown in Figure 10. This feature can be used to fill up irregular shapes in a quick manner. This quickly creates a uniform channel system running throughout the entire scaffold.

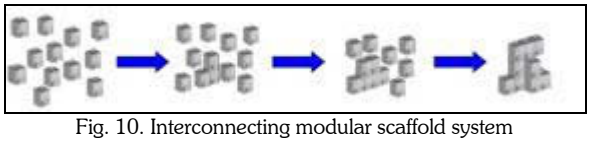


Fig. 10. Interconnecting modular scaffold system

The modular structure also ensures that small channels are located near larger channels immediately within the vicinity. This avoids the problem of long narrow channels within the completed structure that can get clogged. The structure also contains anastamoses, so that even if some of the channels get blocked, there are alternate routes to nourish the cell.

After designing the modular scaffold system, we exported the second modular scaffold design (Figure 9b) into STL format. We then rapid prototyped the scaffold on a 3DP rapid prototyping machine and the prototyped modular unit cell is shown in Figure 11.

Fig. 11 Prototyped modular unit cell (by Z402 3DP rapid prototyping system)

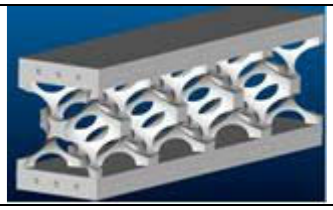


Fig. 12. Strategy III scaffold with compact and porous regions

4.3 DESIGN OF "STRATEGY III" SCAFFOLDS

In general, tissue scaffolds follow two strategies. In Strategy I, the tissue scaffold provides the major support for the implant in vivo until the growing tissue is strong enough to support itself. For this strategy, the biomechanical properties of the scaffold should match

the natural tissue as much as possible. In Strategy II, the tissue scaffold is grown in vitro until it develops enough strength for use in vivo; however, the drawback is that this takes more time to grow the scaffold [19, 20].

With computer-aided tissue engineering, however, we may be able to pursue a third strategy for tissue scaffold design. In the following hypothetical case, a "Strategy III" scaffold (Figure 12) was created in a CAD model that combined Strategies I and II. This scaffold has dense regions similar to compact bone, and porous regions similar to spongy bone where cells can be grown in vitro to increase mechanical strength. The biomimetic "sandwich" resembles flat bone. This scaffold has the advantage that it can be made of a single material yet exhibit different mechanical properties in different regions.

4.4 DESIGN OF MULTI-LAYERED SCAFFOLD WITH CONTROLLED ARCHITECTURE

The scaffold design shown previously is useful for flat bones, but would be difficult to use in irregularly shaped bones. However, by using CATE, multi-layered scaffolds with compact and porous layers can be created through Boolean and scaling operations. To create the channel structure, we made a negative mold and represented it as a CAD model (Figure 13a), and did a Boolean intersection between it and a scaled-down copy of the implant (Figure 13b) to create a new mold. We then subtracted this new mold pattern from the center of the implant (Figure 13c) to create the final implant scaffold with a compact outer layer and a porous, interconnected center (Figure 13d). Also, since cells tend to migrate and thrive in the outer layer of tissue scaffolds, it would be wasteful to have an outer layer with low porosity. By using these same simple techniques, a triple-layered structure with a porous outer layer, a compact middle layer, and a porous inner layer was created (Figure 14a-d).

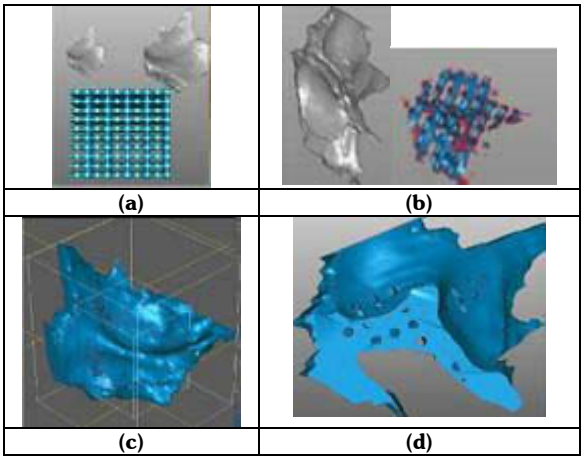


Fig. 13. Creation of a multi-layered scaffold showing (a) mold pattern, implant, and scaled-down implant, (b) Boolean intersection, (c) finished implant, and (d) cutaway view

4.5 DESIGN SCAFFOLD WITH A VASCULAR TREE AND A BUILT-IN DRUG DELIVERY

The power of CATE can be seen in using Boolean operations with pre-designed structures. A vascular tree was created in CAD that followed a basic pathway analogous to artery – arteriole – capillary – venule – vein (Figure 15a). Using scaling and Boolean operations a portion of an implant was quickly "vascularized" (Figure 15b-c).

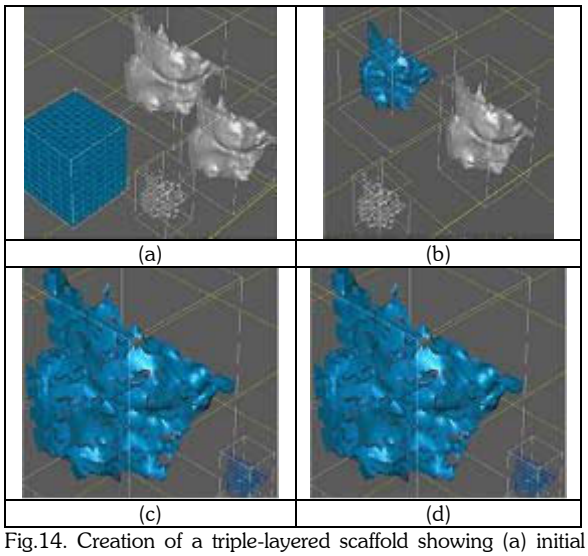


Fig.14. Creation of a triple-layered scaffold showing (a) initial structures, (b) creation of porous outer layer, (c) creation of compact middle layer, (d) cutaway view of finished implant with porous inner layer

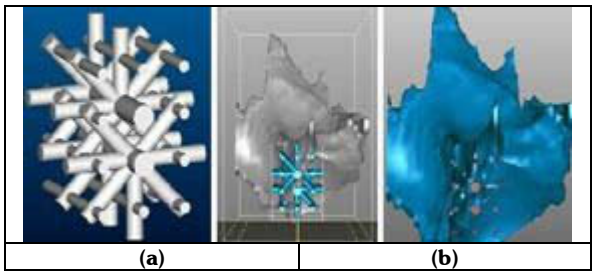


Fig 15. Using CATE to create channels in an implant (a) vascular tree created in CAD, (b) imported and rescaled STL files in Geomagic, and (c) scaffold structure after Boolean operation

Because of the power of CATE, new ideas to improve tissue scaffold design can also be explored. Tissue scaffolds can be designed with built-in functional components that are non-biological in nature. For example, growth factors and drugs play vital roles in

tissue engineering, and so we designed a drug chamber in CAD (Figure 16a) and then added the feature to the scaffold (Figure 16b-d). The existing vascular tree design was then also incorporated into the scaffold (Figure 16d). Other non-biomimetic features such as inlet and outlet ports and attachment interfaces could be added. CATE allows us to quickly assemble sophisticated scaffolds by using pre-designed features.

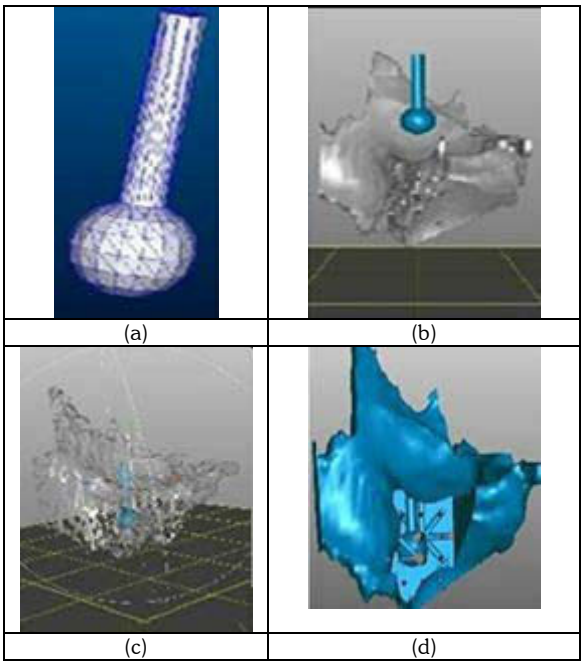


Fig 16. Scaffold design with an integrated delivery system (a) chamber created in CAD, (b) parts before subtraction, (c) chamber insertion, and (d) cutaway showing chamber and channels

5. CONCLUSIONS

This paper was intended to demonstrate the power of applying CATE, particularly CAD in modeling and design of novel tissue scaffolds. The case studies presented in Section 4 show that the enabling CAD technology can help us to: 1) design a scaffold to match a patient's anatomy by using CT or MRI data, 2) control the scaffold's internal architecture (pore size, porosity, interconnectivity), 3) incorporate branching channels into the tissue scaffold, 4) create multi-layered, dense and porous regions with different mechanical properties, 5) integrate artificial, non-biomimetic features into tissue scaffolds, and 6) rapidly assemble sophisticated scaffolds by using pre-designed features.

In general, there is a tradeoff between good mechanical properties and good biological properties. Materials with good mechanical strength tend to be less porous and thus more restrictive for cell growth, while highly porous structures have weaker strength. In addition, because of the sensitivity of cells to local geometries at the microscopic level (such as porosity and pore size), precise control of internal architecture is necessary for the most effective design of tissue scaffolds. Scaffolds are also limited by vascularity, and getting viable cells deep within the interior of scaffolds is a problem. Poor blood supply results in cell death and can affect cell morphogenesis. Experiments have been done in creating vascularized scaffolds by using a hybrid technique of 3D printing and salt leaching [21], etching microvasculature patterns as small as 10 μm using MEMS [22], and creating geometrically scaled, branching channels [23]. CATE proves to be an effective tool in address above biophysical and biological issues.

CAD and freeform fabrication are the two most important components in CATE. In conjunction CAD with solid freeform fabrication make it possible to design and manufacture very complex tissue scaffolds with functional components that are difficult, if not impossible, to create with conventional techniques. Although CATE is still in its early development stage, it eventually will play a significant role in tissue engineering, especially for tissue scaffold design by providing precise and controlled architecture and multi-material printing for different types of biological factors, cells, and scaffold materials. In a near future, CATE can help to design scaffolds that are built to work as mini-bioreactors, for example, designed with a perfusion system that can be loaded to create mechanical stimuli. CATE can also help to design engineering tissue structures at different hierarchical levels: from microscopic to macroscopic, and ultimately, complex scaffolds could be designed to incorporate bioactive materials to interact with the cell and even have external ports or interfaces to give a physician access to the scaffold for drug administration or monitoring.

6. ACKNOWLEDGEMENTS

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