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A method to design biomimetic scaffolds for bone tissue engineering based on Voronoi lattices

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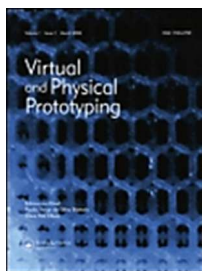
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**A method to design biomimetic scaffolds for Bone Tissue Engineering based on Voronoi lattices**

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## A method to design biomimetic scaffolds for Bone Tissue Engineering based on Voronoi lattices

KEYWORDS: 3d Scaffolds, Voronoi Diagram, Generative Design

### Abstract

In regenerative medicine, 3D scaffolds are used to sustain the regeneration of tissues in removed or damaged parts of the human body. As such practices are being widely experimented in clinical applications, the design, the materials and the manufacturing process to obtain efficient 3D biocompatible lattices are being significantly investigated. Nevertheless, most of the proposed designs are based on regular 3D shapes obtained from the repetition of unit cells disposed in a three-dimensional array. This approach does not exploit the whole potential of Computer Aided Design tools coupled with manufacturing capabilities for freeform shapes. In this paper, we propose a method to model biomimetic lattices controlling the porosity and the pores size of scaffolds to be integrated with the anatomical shape of the defect. The method has been implemented in bone tissue case study and implements a Generative Design approach based on Voronoi diagrams.

### 1. Introduction

Designing synthetic scaffolds for guided tissue regeneration is a very challenging issue. The primary function of a scaffold is to provide a structural template that serves to bear loads of the surrounding tissues, while the cells progressively regenerate in the scaffold volume (Chevallay and Herbage 2000; Yang et al. 2001; Hollister 2005; Saxena 2005). The success of the scaffold depends on several factors which are related to the form of the lattice and to the material used to manufacture it (Ramakrishna et al. 2001; Dee, Puleo and Bizios 2002; Molly and Stevens 2008; Shrivats, McDermott and Hollinger 2014).

On the form side, the requirements that need to be met can be classified into two main levels: i) the external boundary surface of the scaffold must be coherent with the anatomy of the patient to be replaced; ii) the internal porous and interconnected

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microstructure of the scaffold must favor the cells proliferation in the entire volume to be regenerated. Therefore, the scaffold must reproduce the shape of the tissue graft and the ingrowth of cells needs a trabecular architecture, in which the percentage porosity and the pores size must be controllable (Hollister et al. 2000; Wettergreen et al. 2005; Van Cleynenbreugel et al. 2006). These features of the scaffold, together with the degree of pores interconnection (or interconnectivity), have impact on the time and quality of the regeneration (Williams et al. 2006).

Generally, for bone scaffolds, the pores size should be maintained in a given range [150  $\mu\text{m}$  ÷ 600  $\mu\text{m}$ ] (Cornell 1999) and the porosity of the bone can differ according to the specific bone site considered. In literature, it is reported that the bone trabecular morphology is a porous environment variable in a range between {50% ÷ and 90%} whereas the compact bone tissue has a lower porosity [ $<10\%$ ] (Hollister and Kikuchi 1994; Salgado, Coutinho and Reis 2004) due to the *Haversian channels*.

On the material side, the ideal material should be biocompatible and biodegradable, in order to allow the total replacement of biological tissue once the regeneration of tissues is completed (Hollinger and Wong 1996).

Customized porous scaffolds can be obtained by Subtractive Manufacturing (Ciocca et al. 2013a; Ciocca et al. 2013b), starting from blocks of porous material manufactured by different technologies through conventional fabrication methods, including techniques such as solvent casting and particulate leaching, gas foaming, fiber meshes and fiber bonding, phase separation, melt molding, emulsion freeze drying, solution casting and freeze drying (Sachlos and Czernuszka 2003). Also, customized porous scaffolds can be directly manufactured by means of advanced fabrication methods, such as Solid Freeform Fabrication (SFF) or Additive Manufacturing (AM) (Naing et al. 2005; Quadrani et al. 2008; Abdelaal and Darwish 2011) of biocompatible and biodegradable materials.

Several limitations are related to the conventional fabrication methods. These are mainly concerned with the precise control of the pores size, followed by pores geometry, pores interconnection, spatial distribution of pores and construction of internal channels within the scaffold (Peltola et al. 2008). Through Additive Manufacturing, on the other hand, it is virtually possible to control the internal morphology of the scaffold. Limitations concerned with the 3D printed scaffolds are due to material inhomogeneity, limited choice of materials, minimum resolution and

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3 accuracy of machines and, in some cases, difficulty in removing the support material  
4 in very deep and small pores (Hollister, Maddox and Taboas 2002).

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6 For what concerns the biomedical field, Additive Manufacturing has been widely  
7 used for the production of customized surgical guides, bone plates and anatomical  
8 prosthesis (Ciocca et al. 2011; Dobbe et al. 2011; Ciocca et al. 2012; Mazzoli 2013).  
9 Recently, advances in Additive Manufacturing are also opening the way to a better  
10 employment of this technology for the production of customized scaffolds and for the  
11 CAD modeling of anatomic and porous structures to be printed in 3D.  
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14 A modern approach to scaffold design is based on the use of hierarchical structures  
15 created by the repetition of a unit cell of known geometry and known properties and  
16 on the possibility to predict the properties of the scaffolds since the properties of the  
17 unit cell are known. A variety of geometries and geometry libraries have been  
18 proposed and their geometrical and mechanical properties have been compared and  
19 reported (Hoffmann and Arinyo 2002; Chua et al. 2003; Sun et al 2005; Bucklen et al  
20 2008; Gabbrielli, Turner and Bowen 2008). However, the unit cell approach produces  
21 trabecular architectures that do not really mimic the morphology of porous skeletal  
22 structures. These geometries are mainly based on a Constructive Solid Geometry  
23 (CSG) approach and reflect, once combined with the 3D model of the bone graft  
24 substitute, a scarcely natural appearance.  
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27 To overcome this limitation, we propose a method to create customized biomimetic  
28 internal scaffolds with a porous and interconnected microstructure, as it appears in  
29 natural bone, based on a Generative Design approach. Since Generative Design is not  
30 about designing a shape, but it is about designing the process that builds a shape, this  
31 approach allows the user to obtain complex shapes by an automatic modeling process,  
32 and to customize the results by interactively modifying certain parameters.  
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35 The proposed method starts with the generation of Voronoi diagrams. Starting from a  
36 discrete number of points called seeds, a Voronoi diagram is the result of a specific  
37 space partition in which every cell is composed of the points that are closer to the  
38 generating seed. Voronoi diagrams received and receive so much attention because of  
39 their presence in nature and they are being exploited for the generation of shapes in  
40 different fields, such as architecture, jewelry and industrial design in general.  
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43 The paper is organized as follows. The next section gives information on the Voronoi  
44 diagrams and on the way they work. Afterward, the workflow for the generation of a  
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Voronoi-based lattice is described. Results and the way for controlling the features parameters are discussed in the last section before the conclusions.

## 2. Methods: Generation of biomimetic scaffolds based on Voronoi lattice

### 2.1. Voronoi Diagram

Since its introduction and practical application in the early 20<sup>th</sup> century, the Voronoi diagram, initially described by Dirichlet (1850) and Voronoi (1908), has been a popular topic not only in computational geometry ~~and~~ but also in a variety of disciplines. Many natural processes can be used to define particular classes of Voronoi diagrams. Since they are related to several well-known geometrical structures, various authors believe that the Voronoi diagram is one of the most fundamental constructs defined by a discrete set of points. Therefore, such diagrams have increasingly attracted the attention of researchers and computer scientists in the last few years (Aurenhammer 1991).

The ordinary Voronoi diagram, for a point set and its construction, has been studied extensively in both two and higher dimensions (Okabea, Boots and Sugihara 1994). In a 2D vector space, where the Voronoi diagram is modeled, points, lines and polygons are primitives. The primitives, in higher dimensions, are built upon the lower dimensionality primitives. In our case, for the third dimension, we consider the polyhedrons. The Voronoi theory can be thought of as below (Van der Putte and Using 2009).

Let the finite Euclidean space contain a certain number ( $n$ ) of points (with  $n > 1$ ), called seeds. Around each seed a circle is drawn, with a starting radius of 0, expanding at the same rate until two circles touch so that a line or boundary is formed. The circles keep expanding until they cannot expand any further (Fig. 1). The resulting graph is the Voronoi diagram for that set of points. The diagram is built corresponding to the areas in which each point location is closest to the seed corresponding to that area.

Fig. 1 Graphical representation about the formation of a 2D Voronoi diagram

The mathematical equation of such a polygon  $V(p_i)$  can be expressed as follows (Okabe, Boots and Sugihara 1992):

$$V(p_i) = \{ p/d(p, p_j) \leq d(p, p_j), j \neq i, j = 1, \dots, n \}$$

Where:

- $p_1, \dots, p_n$  is a set of distinct seeds located in  $R^d$
- $d(p, p_i)$  represents the Euclidean distance between location  $p$  and seed  $p_i$
- $V(p_i)$  represents the ordinary Voronoi polygon associated with seed  $p_i$

In 2D, Voronoi cells are separated from each other by boundaries that consist of line segments. The Voronoi cell consists of a convex polygon without holes. In 3D, these boundaries consist of planes, and the cells consist of convex polyhedrons without holes. In Fig. 2, an example of the 3D Voronoi diagram is shown.

Fig.2 a) Reference cube, b) 3D Voronoi polyhedron, c) Partition in 3D Voronoi polyhedrons

## 2.2. Voronoi-based lattice generation

In this section, a Generative Design workflow implementing the 3D Voronoi diagram for modeling bone tissue engineering scaffolds is proposed (Fig. 3). The aim is to automatically build a trabecular structure, starting from a set of points randomly distributed in the bounding box volume of the bone defective part to be replaced, hereafter called patient bone geometry. This process allows to obtain a customized scaffold controlling the percentage porosity and pores size in order to reach the desired values.

Fig. 3 Workflow for the 3D Voronoi scaffold generation

The generative workflow allows to design a porous and interconnected lattice ready to be 3D printed, starting from a data set of four inputs: the patient bone geometry acquired from CT scan and surgery planning, the number of seeds ( $n$ ) and two scale factors ( $S_f$  and  $S_v$ ). The output is generated through a series of sub-routines

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3 implemented in sub-circuits designed in Grasshopper, the graphical algorithm editor  
4 integrated into Rhinoceros 5. The first sub-circuit works on the bounding box of the  
5 patient bone geometry, creating a set of points randomly distributed in the bounding  
6 box volume (*Populate 3D*). Such points will be the seeds to generate the 3D Voronoi  
7 structure that divides the bounding box volume into a set of polyhedral cells (Fig. 4).  
8 Each cell, being a closed polysurface, encloses a volume and each face of the cell  
9 coincides with the boundary face of the adjacent cell. With the aim to gather a porous  
10 and interconnected lattice structure, we need to generate a single closed polysurface,  
11 namely a solid, with the seeds at the center of the pores and void channels to  
12 interconnect these pores.  
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22 **Fig. 4 Partition in Voronoi polyhedrons of the bounding box volume generated from the patient anatomy**

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27 The method here described is based on the idea of building the lattice as a network of  
28 solid beams along the edges of the polyhedral cells generated above. Starting from  
29 these polyhedrons, a deconstruction operation (*DeBrep*) is applied in order to obtain  
30 the boundary representation of each polyhedral cell, in terms of a list of faces, edges  
31 and vertices (F, E, V). Thus, the *DeBrep* component allows the separation of all the  
32 faces from the 3D Voronoi diagram. Two parallel computations are then performed.  
33 The first one starts from the faces of the polyhedral cells and scales each face  
34 assuming its centroid as reference point with a scale factor  $S_f$ . In parallel, each  
35 starting polyhedral cell is scaled assuming its centroid as reference point with a scale  
36 factor  $S_v$ . (Fig. 5).  
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46 **Fig. 5 a) Each face is scaled respect the face's centroid, b) each polyhedron is scaled respect the**  
47 **polyhedron's centroid**

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51 Next, the faces, edges and vertices of such smaller polyhedral cells are obtained again  
52 through the *DeBrep* component. Hence, we have the edges of the scaled faces of the  
53 starting cells and the edges of the faces of the scaled cells. These edges are the  
54 skeleton of the trabecular structure. Then, in order to obtain the beams of the  
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3 trabeculae we need to build a solid on these edges. This is done by generating a series  
4 of quad meshes on the edges vertex, which can be extracted in the proper list (Fig. 6).  
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9 **Fig. 6 a) Generation and deconstruction of faces and polyhedrons, b) particular of scaled faces and**  
10 **polyhedrons, c) quad meshes on the edges to construct the lattice**  
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13 Before the final intersection for producing the customized scaffold, there is another  
14 component that makes an essential task, based on the *Catmull-Clark* algorithm. This  
15 is a technique used in computer graphics to create smooth surfaces by subdivision  
16 surface modeling. Edwin Catmull and Jim Clark devised it in 1978 as a generalization  
17 of bi-cubic uniform B-spline surfaces to arbitrary topology. By this component, which  
18 is contained in the Rhinos plug-in Wavebird for shape reconstructing, a smoothed  
19 lattice mesh can be obtained (Fig. 7).  
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27 **Fig. 7 a) Trabecular mesh, b) Smoothed lattice mesh after *Catmull-Clark* algorithm**  
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30 Finally, a mesh Boolean intersection is performed between the patient bone geometry  
31 mesh and the Voronoi lattice mesh in order to obtain the customized scaffold mesh of  
32 the bone graft (Fig. 8).  
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37 **Fig. 8 Boolean intersection of the patient bone geometry and the lattices to obtain the porous scaffold**  
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41 It is clear that varying the number of seeds ( $n$ ) and the scale factor for the faces ( $S_f$ )  
42 and for the volume ( $S_v$ ) of the polyhedral cells into a range from  $[0\div 1]$ , a different  
43 lattice is obtained in terms of percentage porosity and pores size. In order to find the  
44 correlation between these input parameters (number of seeds and scale factors) and  
45 the target parameters (percentage porosity and pores size), in the next section, an  
46 analysis procedure is proposed. It is based on comparing the key parameters for the  
47 scaffold evaluation such as percentage porosity, pores size, pores interconnection and  
48 trabecular thickness.  
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### 56 **3. Results**

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In order to control the final trabecular morphology of the porous and interconnected Voronoi scaffold, we need to correlate the input parameters with the target ones. The input parameters are:

- the number of seeds ( $n$ ) to be included in the bounding box of the scaffold;
- the scale factors  $S_f$  and  $S_v$ .

The target parameters that we consider in this paper are:

- the percentage porosity  $P\%$ ;
- the pores size.

The percentage porosity  $P\%$  can be easily computed as follows:

$$P\% = \frac{V_{\text{bounding box}} - V_{\text{scaffold}}}{V_{\text{bounding box}}} \times 100$$

Since:

$$V_{\text{void}} = V_{\text{bounding box}} - V_{\text{scaffold}}$$

Actually, we can show how the number of seeds and the scale factors  $S_f$  and  $S_v$  influence the percentage porosity, where:

- $S_f$  is the polyhedrons faces scale factor;
- $S_v$  is the polyhedrons volume scale factor.

First of all, by varying each scale factor in a range of  $[0 \div 1]$  and maintaining the same number of seeds, we derived a sample of scaffold and measured the percentage porosity ( $P\%$ ), which is referred to each pair of values of the two scale factors. In Fig. 9 three snapshots of a cubic scaffold ( $10 \times 10 \times 10$  mm), with the same number of seeds ( $n = 500$ ), generated by setting  $S_f = S_v = 0.25$  for the first one,  $S_f = S_v = 0.50$  for the second one and  $S_f = S_v = 0.75$  for the third one, are depicted.

**Fig. 9 Sample of scaffold ( $10 \times 10 \times 10$  mm) with 500 seeds and setting: a)  $S_f = S_v = 0.25$  ( $P\% = 17.4$ ), b)  $S_f = S_v = 0.50$  ( $P\% = 47.8$ ), c)  $S_f = S_v = 0.75$  ( $P\% = 79.6$ )**

Afterward, we plotted the percentage porosity obtained by varying the  $S_v$  scale factor for different values of the  $S_f$  scale factor. Results are shown in Fig. 10.

**Fig. 10 Effect of the scale factors  $S_f$  and  $S_v$  on the percentage porosity of the lattice (with 500 seeds)**

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3 Then, for each pair of values of the two scale factors ( $S_f$  and  $S_v$ ), we generated a  
4 sample of scaffold by varying the number of seeds in a range of [100÷1000], with  
5 incremental steps of 50, and computed the percentage porosity ( $P\%$ ). Results are  
6 shown in Fig. 11 for the sample of scaffold generated by setting  $S_f = S_v = 0.75$ .  
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11 **Fig. 11 Effect of the number of seeds on the percentage porosity of the scaffold (with  $S_f = S_v = 0.75$ )**  
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14 The first observation on these results concerns the impact of the number of seeds onto  
15 the percentage porosity. By varying the number of seeds, we can observe that the  
16 variation in the percentage porosity of the scaffolds in a sample, characterized by  
17 constant values of  $S_f$  and  $S_v$ , is very low. This can be explained considering that the  
18 number of seeds determines both the number of pores and the number of trabeculae at  
19 the same time. Thus, a larger number of seeds means a larger number of pores  
20 (voids), but also a larger number of trabeculae (solids), whose presence balances the  
21 total amount of the void volume. Therefore, it can be assumed that the scale factors  $S_f$   
22 and  $S_v$  are related to the expected percentage porosity ( $P\%$ ) of the final scaffold with a  
23 very low influence of the number of seeds ( $n$ ): the greater are the scale factors the  
24 higher is the total porosity. To obtain, for example, an expected percentage porosity  
25  $P\% \approx 80\%$ , both scale factors ( $S_f$  and  $S_v$ ) have to be set to 0.75. Of course, the  
26 geometries of the scaffolds will differ for different combinations of  $S_f$  and  $S_v$  that  
27 allow obtaining the same value of percentage porosity.  
28

29 Since for a given combination of  $S_f$  and  $S_v$  we can control the porosity with a good  
30 accuracy, we can now observe the effect of a different number of seeds ( $n$ ) on the  
31 morphology of the scaffold.  
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33 The number of seeds (or the seeds density as the number of seed points per unit  
34 volume) can be given in order to gather a target pores size of the scaffold. Hence, this  
35 is a significant parameter that has to be identified and quantified being also an  
36 important index for cells habitability. The ideal populating input number of seeds can  
37 be estimated in relation to the target percentage porosity and the target pores size, by  
38 setting the relative desired values as input.  
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40 To this aim, starting from the bounding box volume of the scaffold, the total void  
41 volume can be easily estimated as a function of the target percentage porosity ( $P\%$ ).  
42 Assuming that all the pores have a spherical shape with the same diameter ( $D_p$ ), the  
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total number of pores is computed as the target total void volume subdivided by the volume of a single sphere of the given diameter.

By the proposed workflow for the 3D Voronoi scaffold generation, the pores (voids) can be represented by the polyhedral cells and, therefore, the number of pores corresponds to the number of seeds.

In this way, the ideal populating input number of seeds ( $n$ ) is estimated as the ratio of the target total void volume and the volume of the sphere with the diameter ( $D_p$ ) representing the target pores size.

$$n = \frac{V_{\text{bounding box}} \times P\%}{\frac{4}{3}\pi \left(\frac{D_p}{2}\right)^3}$$

Since:

$$V_{\text{void}} = V_{\text{bounding box}} \times P\%$$

Then, to evaluate the actual size of the randomly created pores, we propose the following method. Since the pores have ~~not a~~ irregular shapes, the idea is based on the identification, for each polyhedron, of the sphere that is centered at the centroid of the polyhedral cell and has the same volume of the polyhedron scaled by  $S_v$ . This will be recalled as the polyhedron-sphere method. In Fig. 12 the approximated spherical pores are depicted for a sample of scaffold with the shape of a cube 10 mm sided ( $P\% = 81.6$  and 100 seeds). In this way, we can evaluate the size of each pore as the diameter of the related polyhedron-sphere, so that a comparison with the literature values, included between [150  $\mu\text{m}$  ÷ 600  $\mu\text{m}$ ] can be provided, starting from the number of seeds set as input in the Voronoi diagram.

**Fig. 12 Polyhedron-sphere method: a) polyhedrons scaled by  $S_v$ , b) polyhedrons scaled by  $S_v$  and corresponding spheres, c) spheres representing the pores**

Therefore, we generated a sample of scaffold (10×10×10 mm), with a bounding box volume = 1000 mm<sup>3</sup>, setting  $S_f = S_v = 0.75$  to obtain an expected percentage porosity  $P\% \approx 80\%$ . The number of Voronoi seeds was estimated in relation to target percentage porosity ( $P\% = 80\%$ ) and the target pores size (varying from 5.00 mm to 0.50 mm), as the ratio of the target total void volume (800 mm<sup>3</sup>) to the volume of the sphere with the diameter representing the target pores size (Fig. 13).

**Fig. 13 Number of Voronoi seeds estimated in relation to the target pores size for a cubic scaffold 10 mm sided with a target percentage porosity  $P\% = 80\%$**

Then, the actual outcoming size of each pore of the scaffold, generated with different target pores size (and, therefore, different number of seeds), was measured by applying the polyhedron-sphere method. The results are reported in Fig. 14.

**Fig. 14 Pores size (measured applying the polyhedron-sphere method) of a cubic scaffold 10 mm sided, scale factors  $S_f = S_v = 0.75$ , with a target percentage porosity  $P\% = 80\%$ , generated with different target pores size (and therefore different number of seeds)**

In Fig. 15 two snapshots of a cubic scaffold 10 mm sided, with the same scale factors ( $S_f = S_v = 0.75$ ), generated with a different number of seeds (200, 800), are depicted.

**Fig. 15 Sample of scaffold (10x10x10 mm) with  $S_f = S_v = 0.75$  and setting: a) number of seeds = 200 (target pores size = 2 mm), b) number of seeds = 800 (target pores size = 1 mm)**

The first observation on these results shows as the number of Voronoi seeds rapidly increases with the decreasing of the target pores size: in the sample of scaffold with the shape of a cube 10 mm sided, a target pores size of 5 mm corresponds to 12 seeds, while a target pores size of 0.5 mm correspond to 12'223 seeds. To obtain, for example, the actual size of each pore included in the range  $[150 \mu\text{m} \div 600 \mu\text{m}]$ , a suitable target pores size can be set to 0.6 or 0.5 mm.

The second observation concerns the size of the actual pores that is almost entirely smaller than the target one. Since on the generated scaffold the pores have different dimensions, their measures can slightly differ from the target value. We observed that in the generated scaffold the maximum pores size approximate with a good accuracy the target pores size, and the variance is relatively low, remaining below 1 and decreasing with the increasing of the number of seeds. This can be explained considering that the total pores void, computed by the polyhedron-sphere method, results, as expected, in  $421.875 \text{ mm}^3$ , since the starting Voronoi polyhedrons (with total volume  $1000 \text{ mm}^3$ ) are scaled by  $S_v = 0.75$  in the 3 dimensions ( $1000 \text{ mm}^3 \times 0.75 \times 0.75 \times 0.75 = 421.875 \text{ mm}^3$ ). The remaining part of void ( $378,125 \text{ mm}^3$ ) to reach the expected 80% porosity ( $800 \text{ mm}^3$ ) is due to connective channels between the pores, essential to assure a correct pores interconnection.

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3 Moreover, the number of seeds can give us information about the number of pores  
4 inside the customized scaffold. Since the input is given as the number of points to be  
5 included in the bounding box of the patient bone geometry, we initially assumed that  
6 the number of seeds in the model of the customized scaffold depends on the ratio  
7 between the volume of this model and the volume of its bounding box. Therefore, a  
8 rough estimation of the number of seeds (and, therefore, pores) in the final scaffold  
9 can be based on this proportion. Actually, depending on the shape of the patient bone  
10 geometry, a different number of expected seeds (and therefore pores) can be included  
11 in the final mesh of the customized scaffold.  
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14 Another key parameter for the scaffold evaluation is the pores interconnection (or  
15 interconnectivity) that is the property, which allows the perfusion of nutrients and the  
16 extrusion of wasting material inside the cancellous bone structure. This is the main  
17 reason for preferring scaffolds with a high permeability structure that also ensures the  
18 availability of higher surface area for enhanced cell adhesion and proliferation.  
19 Therefore, the degree of pores interconnection can be considered as important as the  
20 pores size.  
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23 In the case of the generated Voronoi scaffold, we tried to understand how each pore  
24 communicates with its neighbors, and in particular to find the number and the size of  
25 the connective channels departing from each pore.  
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28 We have already seen that the pores (voids) can be represented by the polyhedral cells  
29 scaled by  $S_v$  and that the actual size of the pores can be evaluated by means of the  
30 polyhedron-sphere method.  
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33 If we look at the void component (pores plus connective channels) of a very simple  
34 cubic Voronoi scaffold (10 mm sided), generated with only 8 seeds corresponding to  
35 the bounding box vertices and minimal scale factors ( $S_f = S_v = 0.25$ ), we can observe  
36 how the 8 pores are mutually interconnected (Fig. 16a).  
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48 **Fig. 16 Void component (pores plus connective channels) for a Voronoi scaffold with minimal scale factors**  
49 **( $S_f = S_v = 0.25$ ) and a) number of seeds = 8, b) number of seeds = 9**  
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51  
52 For each of the 8 polyhedrons (that are cubes in this case), six connective channels  
53 depart, one from each face of the cube, giving the impression that the resulting  
54 structure is six-fold connected. If we add another seed (the centroid of the bounding  
55 box), we can now observe 9 polyhedral cells with different shapes and more faces  
56 with respect to the previous case (Fig. 16b). Since the connective channels are still  
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3 departing from each face of the polyhedrons to run out to the corresponding face of  
4 the neighbor, the level of interconnectivity increases with the number of Voronoi  
5 seeds. Moreover, we can also observe that the size of each connective channel is  
6 represented by the area of the departing face of the polyhedral cell.  
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9 Therefore, we can conclude that the mean number of faces of the polyhedrons could  
10 give the level of interconnectivity of the generated Voronoi scaffold, and the mean  
11 area of the faces of the polyhedrons could give the mean size of the connective  
12 channels. The properties of the connective channels have been computed as the mean  
13 pores interconnection and mean channels size for a Voronoi scaffold with a target  
14 percentage porosity ( $P\% = 80\%$ ) and varying the target pores size (from 5.00 mm to  
15 0.50 mm), as depicted in Fig. 17.  
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23 **Fig. 17 Mean pores interconnection and mean channels size of a cubic scaffold 10 mm sided, scale factors  $S_f$**   
24 **=  $S_v = 0.75$ , with a target percentage porosity  $P\% = 80\%$ , generated with different target pores size (and**  
25 **therefore different number of seeds)**  
26

27 Finally, the trabecular thickness (that represents the solid part of the scaffold opposite  
28 to the total void volume) can be evaluated as the mean value of the distance between  
29 the centroids of the coupled faces of neighboring polyhedral cells scaled by  $S_v$ . We  
30 have seen that Voronoi polyhedrons, scaled by  $S_v$ , can represent the pores (voids) of  
31 the scaffold structure, evaluated by means of the polyhedron-sphere method,  
32 therefore, the trabecular skeleton is built up in the free space between these cells  
33 (Fig. 18).  
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40 **Fig. 18 Trabecular thickness evaluation: a) Voronoi polyhedral cells scaled by  $S_v$  with the distance of the**  
41 **centroids of coupled faces; b) Voronoi polyhedral cells scaled by  $S_v$  with the trabecular mesh; c) the final**  
42 **scaffold ( $S_f = S_v = 0.75$ ; number of seeds  $n = 12$ ; target pores size  $D_p = 5$  mm)**  
43

44 The trabecular thickness has been computed for a Voronoi scaffold with a target  
45 percentage porosity ( $P\% = 80\%$ ) and by varying the target pores size (from 5.00 mm  
46 to 0.50 mm), as depicted in Fig. 19.  
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50 **Fig. 19 Mean pores interconnection and mean channels size of a cubic scaffold 10 mm sided, scale factors  $S_f$**   
51 **=  $S_v = 0.75$ , with a target percentage porosity  $P\% = 80\%$ , generated with different target pores size (and**  
52 **therefore different number of seeds)**  
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#### 55 4. Discussion

56 The main findings of this work concern two aspects, which are the general design  
57 method and the CAD approach followed. The general design method resulted very  
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3 efficient since the generation of biomimetic porous lattices, shaped on the patient  
4 anatomy, is very intuitive and fast. Moreover, great accuracy in the expected  
5 percentage porosity and pores size has been achieved by setting the suitable input  
6 parameters. The CAD approach also resulted positive since a closed and watertight  
7 mesh of the customized porous scaffold, for different bone grafts, have been obtained  
8 in a short time. This allows to directly interfacing the generative modeling module for  
9 the generation of porous interconnected scaffolds with Additive Manufacturing  
10 systems.

11  
12 These findings can be explained by the fact that the interactive Generative Design  
13 approach, which has been developed from Voronoi diagram, is independent by the  
14 final shape of the scaffold and allows to simply modify the input parameters in  
15 suitable ranges for satisfying the surgical requirements in terms of both percentage  
16 porosity and pores size.

17  
18 In a recent review about the design of scaffold for bone tissue engineering,  
19 considering the emergence of Solid Freeform Fabrication (SFF) methods (Velasco,  
20 Narváez-Tovar and Garzón-Alvarado 2015), the main reported design techniques, in  
21 previous relevant works, are based on Constructive Solid Geometry (CSG), Boundary  
22 Representation (B-Rep) and, in the last years, on implicit surfaces like Triply Periodic  
23 Minimal Surfaces (TPMS). Only a case study based on Voronoi diagrams for general  
24 irregular porous structures modeling has been reported (Kou and Tan 2010).  
25 However, this paper does not investigate key parameters for bone replacement  
26 scaffolds generation, such as percentage porosity, pores size, pores interconnection  
27 and trabecular thickness. Moreover, no practical recommendations are provided for  
28 guiding the design of internal trabecular morphology of the bone scaffold.

29  
30 Furthermore, the new and important aspect of this study is not the design of a  
31 specific scaffold, but the development of a general interactive method to design  
32 different customized scaffolds according to the specific requirements provided by the  
33 clinical team. Therefore, the proposed method can be a useful tool for the design of  
34 biomimetic scaffolds for bone regeneration.

35  
36 Nevertheless, together with the design, ~~also~~ materials and manufacturing processes  
37 also play ~~a~~ fundamental roles in obtainingto-obtain efficient 3D biocompatible  
38 scaffolds with this internal trabecular microstructure. For this reason, the capabilities  
39 of Additive Manufacturing, according to the biomaterials used, should allow the  
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3 manufacturability of these scaffolds in terms of percentage porosity, pores size, pores  
4 interconnection and trabecular thickness.

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6 Additive Manufacturing is evolving from rapid prototyping to the end-of-use product  
7 manufacturing and offers numerous benefits for innovative design solutions.  
8  
9 Nowadays, design freedom has been significantly broadened, including shape  
10 complexity, material complexity, hierarchical complexity, and functional complexity  
11 (Gibson, Rosen and Stucker 2010; Yang and Zhao 2015).  
12  
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14 The emerging field of Design for Additive Manufacturing (DFAM) is exploring new  
15 methodological frameworks and design rules, but is also highlighting geometrical  
16 limitations and constraints to be considered (Adam and Zimmer 2015; Kumke,  
17 Watschke and Vietor 2016).  
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20 Therefore, to validate the manufacturability of the proposed biomimetic scaffolds  
21 based on Voronoi diagram, a review of possible methods that can be used in the  
22 actual fabrication, in terms of Additive Manufacturing systems and biomaterials, is  
23 reported. The three main classes of synthetic biomaterials used today are metals,  
24 ceramics and polymers.  
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27 The class of biomaterials with a long history of application in bone implants  
28 comprises of metals and alloys. Among them, the use of stainless steels, cobalt (Co),  
29 titanium (Ti) and relative alloys are well proven due to their good biocompatibility,  
30 satisfactory mechanical strength and superior corrosion resistance. Therefore, Metal-  
31 based Additive Manufacturing (MAM) techniques, such as Selective Laser Melting  
32 (SLM) and Electron Beam Melting (EBM), are increasingly being used for the  
33 fabrication of porous metals for bone scaffolds and orthopaedic implants, with  
34 predefined external shape and internal architecture (Wang et al. 2016).  
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37 For example, Yan et al. (2015) reported that Ti-6Al-4V biomorphic lattices for bone  
38 implants were manufactured by Selective Laser Melting (SLM). The design was  
39 based on Gyroid and Diamond Triply Periodic Minimal Surface (TPMS), having an  
40 interconnected high porosity of 80-95% and pores size in the range of 560-1600  $\mu\text{m}$   
41 and 480-1450  $\mu\text{m}$  respectively. The manufacturability, microstructure and mechanical  
42 properties were evaluated and the comparison between 3D micro-CT reconstructed  
43 models and original CAD models of the Ti-6Al-4V TPMS lattices showed excellent  
44 reproduction of the designs.  
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47 Also, Taniguchi et al. (2016) studied the effect of pores size on bone ingrowth into  
48 porous titanium implants fabricated by Selective Laser Melting (SLM), since this  
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3 Additive Manufacturing technique has the ability to produce metallic scaffolds with  
4 accurately controlled porosity, pores size and interconnectivity for orthopedic  
5 applications. Three porous titanium implants, based on a Diamond lattice basic  
6 structure (with an intended porosity of 65% and pores size of 300, 600, and 900  $\mu\text{m}$ ),  
7 were successfully manufactured by EOSINT-M270 SLM system. The porous  
8 implants were evaluated using Computed Tomography and the actual average pores  
9 size resulted of 309, 632, and 956  $\mu\text{m}$ , respectively.

10  
11 Finally, Sing et al. (2015) investigated the effect of design and process parameters on  
12 the dimensional accuracy of titanium lattice structures fabricated by using Selective  
13 Laser Melting (SLM). Geometrical design parameters, such as unit cell type and strut  
14 diameter, had not significant effects on the dimensional accuracy of the lattice  
15 structures. On the other hand, the processing parameters, such as laser power and  
16 laser scan speed, had a significant effect on the powder adhesion on the struts and  
17 consequentially affected the dimensional accuracy.

18  
19 In the field of ceramic biomaterials, Sabree et al. (2015) reported on how highly  
20 porous ceramic scaffolds have been fabricated by using stereolithography and the  
21 lost-mould process combined with gel-casting. All scaffolds had a simple cubic strut  
22 structure with an internal porosity of approximately 42% and internal pore dimensions  
23 in the range of 300-600  $\mu\text{m}$ .

24  
25 For what concerns the class of polymers as biomaterials, another method for the  
26 actual fabrication of Voronoi-based lattices could be the manufacturing as  
27 Poly(propylene fumarate) (PPF) scaffolds by using an EnvisionTEC Perfactory 3D  
28 Printer. Poly(propylene fumarate) is an important biodegradable and crosslinkable  
29 polymer designed for bone tissue-engineering applications. EnvisionTEC Perfactory  
30 3D Printer is a rapid prototyping manufacturing system, based on Digital Light  
31 Processing (DLP) technology, to create 3D models that range from the conceptual to  
32 the fully functional. Some examples of fabrication of porous PPF scaffolds, as a huge  
33 opportunity for the regenerative medicine, have been proposed.

34  
35 Wang et al. (2015) reported the manufacturing of porous, cylindrical scaffolds,  
36 consisting of modular ring-shaped bases, with 500  $\mu\text{m}$  wall thickness, two pore sizes  
37 (400  $\mu\text{m}$  and 800  $\mu\text{m}$ ) and two different porosities (25% and 50%). More recently,  
38 Luo et al. (2016) have described the fabrication of a porous, cylindrical scaffold, by  
39 using the Schoen Gyroid Triply Periodic Minimal Surface (TPMS) geometry, with  
40 400  $\mu\text{m}$  strut thickness, pore diameter of 1400  $\mu\text{m}$ , and porosity of 88.2%.

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3 Actually, the next step of this work will be the concrete fabrication of the proposed  
4 Voronoi-based lattices to directly assess the manufacturability. Moreover, as a  
5 natural extension of this work could aim to find a proper seeds distribution to allow  
6 the generation of scaffolds with non-homogenous porosity, for example, with higher  
7 density for the compact external part (cortical or compact bone tissue) and lower  
8 density for the trabecular internal one (cancellous or spongy bone tissue). Since the  
9 initial distribution of Voronoi seeds influences the final scaffold trabecular  
10 morphology, non-homogenous scaffolds could be obtained with a higher  
11 concentration of seeds distribution in the external part and lower in the internal one.  
12 This method can be also refined and/or expanded on other requirements, such as the  
13 mechanical properties, once the density of seeds has been related to such properties.  
14 However, some limitations of this study should be also considered, since a drawback  
15 of this approach is related to the generation of very large scaffolds due to the  
16 computational resources required.  
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## 28 **5. Conclusions**

29 In this paper, we proposed a method to design interconnected porous lattices that  
30 mimic specific tissue characteristics for the realization of bone regenerative scaffolds.  
31 The shape of reference is a healthy spongy bone, which presents a heterogeneous  
32 structure to enhance the cells proliferation and the tissue regeneration. The greater  
33 advantage of this method, compared to other approaches, is to provide geometrical  
34 heterogeneity thus resulting in a really biomimetic shape (Fig. 20).  
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39 The method is applied to provide an intuitive and fast tool to create biomimetic  
40 scaffolds and is expandable to other tissue engineering requirements or to other  
41 domains in which the design porous interconnected structures is needed.  
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46 **Fig. 20 a) 3D generated Voronoi Scaffold, b) Healthy spongy bone (Marinozzi et al. 2012)**  
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## References

Abdelaal OA, Darwish SM. Fabrication of Tissue Engineering Scaffolds Using Rapid Prototyping Techniques. World Academy of Science, Engineering and Technology. 2011;59:11-27.

Adam GAO, Zimmer D. On design for additive manufacturing: evaluating geometrical limitations Rapid Prototyping J. 2015;21(6):662-670.

Aurenhammer F. Voronoi diagrams – A survey of a Fundamental Geometric Data Structure. AMC Computing Surveys. 1991;23(3):345-405.

Bucklen BS, Wettergreen MA, Yuksel E, Liebschner MAK. Bone-derived CAD Library for Assembly of Scaffolds in Computer-Aided Tissue Engineering. Virtual Phys Prototyp. 2008;3(1):13-23.

Chevallay B, Herbage D. Collagen-based biomaterials as 3D scaffold for cell cultures: applications for tissue engineering and gene therapy. Med Biol Eng Comput. 2000;38(2):211-218.

Chua CK, Leong KF, Cheah CM, Chua SW. Development of a Tissue Engineering Scaffold Structure Library for Rapid Prototyping. Part 1: Investigation and Classification. Int J Adv Manuf Technol. 2003;21:291-301.

Ciocca L, Fantini M, De Crescenzo F, Corinaldesi G, Scotti R. Direct metal laser sintering (DMLS) of a customized titanium mesh for prosthetically guided bone regeneration of atrophic maxillary arches. Med Biol Eng Comput. 2011;49(11):1347-1352.

Ciocca L, Mazzoni S, Fantini M, Persiani F, Baldissara P, Marchetti C, Scotti R. A CAD/CAM-prototyped anatomical condylar prosthesis connected to a custom-made bone plate to support a fibula free flap. Med Biol Eng Comput. 2012;50(7):743-749.

1  
2  
3 Ciocca L, Donati D, Ragazzini S, Dozza B, Rossi F, Fantini M, Spadari A, Romagnoli  
4 N, Landi E, Tampieri A, Piattelli A, Iezzi G, Scotti R. Mesenchymal Stem Cells and  
5 Platelet Gel Improve Bone Deposition within CAD-CAM Custom-Made Ceramic HA  
6 Scaffolds for Condyle Substitution. *Biomed Res Int.* 2013; Article ID 549762, 10  
7 pages.  
8  
9

10  
11  
12  
13 Ciocca L, Donati D, Fantini M, Landi E, Piattelli A, Iezzi G, Tampieri A, Spadari A,  
14 Romagnoli N, Scotti R. CAD-CAM generated hydroxyapatite scaffold to replace the  
15 mandibular condyle in sheep: preliminary results. *J Biomater Appl.* 2013;28(2):207-  
16 218.  
17  
18

19  
20  
21 Cornell CN. Osteoconductive materials and their role as substitutes for autogenous  
22 bone grafts. *Orthopedic Clinics of North America.* 1999;30(4):591-598.  
23  
24

25  
26 Dee KC, Puleo DA, Bizios R. *An Introduction to Tissue-Biomaterial Interaction.* John  
27 Wiley & Sons. 2002;3:37-52.  
28  
29

30  
31 Dobbe JDD, Vroemen JC, Strackee SD, Streekstra GJ. Patient-tailored plate for bone  
32 fixation and accurate 3D positioning in corrective osteotomy. *Med Biol Eng Comput.*  
33 2011;51(1):19-27.  
34  
35

36  
37  
38 Gabbrielli R, Turner IG, Bowen CR, Development of Modelling Methods for  
39 Materials to be Used as Bone Substitutes. *Key Engineering Materials.* 2008;361-  
40 363:903-906.  
41  
42

43  
44  
45 Gibson I, Rosen DW, Stucker B. *Additive Manufacturing technologies: rapid*  
46 *prototyping to direct digital manufacturing.* 2010, Springer, US.  
47  
48

49  
50 Hoffmann C, Arinyo RJ. Parametric Modelling. *Handbook of CAGD,* Farin G,  
51 Hoschek J, Kim MS, eds. Elsevier. 2002;519-541.  
52  
53

54  
55 Hollinger J, Wong ME. The integrated processes of hard tissue regeneration with  
56 special emphasis on fracture healing. *Oral Surg Oral Med Oral Pathol Oral Radiol*  
57 *Endod.* 1996;82(6):595-606.  
58  
59  
60

1  
2  
3  
4  
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52  
53  
54  
55  
56  
57  
58  
59  
60

Hollister SJ. Porous scaffold design for tissue engineering. *Nat Mater*. 2005;4(7):518-524.

Hollister SJ, Kikuchi N. Homogenization Theory and Digital Imaging: A Basis for Studying the Mechanics and Design Principles of Bone Tissue. *Biotechnology and Bioengineering*. 1994;43(7):586-596.

Hollister SJ, Maddox RD, Taboas JM. Optimal design and fabrication of scaffolds to mimic tissue properties and satisfy biological constraints. *Biomaterials*. 2002;23:4095-4103.

Hollister SJ, Levy RA, Chu TM, Halloran JW, Feinberg SE. An image based approach for designing and manufacturing craniofacial scaffolds. *Int J Oral Maxillofac Surg*. 2000;29(1):67-71.

Kou XY, Tan ST. A simple and effective geometric representation for irregular porous structure modelling. *Comput Aided Des*. 2010;42(10): 930-941.

Kumke M, Watschke H, Vietor T. A new methodological framework for design for additive manufacturing. *Virtual Phys Prototyp*. 2016;11(1):3-19.

Luo Y, Dolder CK, Walker JM, Mishra R, Dean D, Becker ML. Synthesis and Biological Evaluation of Well-Defined Poly(propylene fumarate) Oligomers and Their Use in 3D Printed Scaffolds. *Biomacromolecules*. 2016;17(2):690-697.

Marinozzi F, Marinozzi A, Bini F, Zuppante F, Pecci R, Bedini R. Variability of morphometric parameters of human trabecular tissue from coxo-arthritis and osteoporotic samples. *Ann Ist Super Sanita*. 2012;48(1):19-25.

Mazzoli A. Selective laser sintering in biomedical engineering. *Med Biol Eng Comput*. 2013;51(3):245-256.

1  
2  
3 Molly M, Stevens MM. Biomaterials for bone tissue engineering. *Materials Today*.  
4 2008;11(5):18-25.  
5

6  
7  
8 Naing MW, Chua CK, Leong KF, Wang Y. Fabrication of customised scaffolds using  
9 computer-aided design and rapid prototyping techniques. *Rapid Prototyping J*.  
10 2005;11(4):249-259.  
11

12  
13  
14 Okabe A, Boots B, Sugihara K. *Spatial Tessellations - Concepts and Applications of*  
15 *Voronoi Diagrams*. Wiley, 1992.  
16

17  
18  
19 Okabea A, Bootsb B, Sugiharac K. Nearest neighbourhood operations with  
20 generalized Voronoi diagrams: a review. *Int J Geogr Inf Sci*. 1994;8(1):43-71.  
21

22  
23  
24 Peltola SM, Melchels FP, Grijpma DW, Kellomäki M. A review of rapid prototyping  
25 techniques for tissue engineering purposes. *Ann Med*. 2008;40(4):268-280.  
26

27  
28  
29 Quadrani P, Pasini A, X Mattioli-Belmonte M, Zannoni C, Tampieri A, Landi E,  
30 Giantomassi F, Natali D, Casali F, Biagini G, Tomei-Minardi A. High-resolution 3D  
31 scaffold model for engineered tissue fabrication using a rapid prototyping technique.  
32 *Med Biol Eng Comput* 2005;43(2):196-199.  
33

34  
35  
36  
37 Ramakrishna S, Mayer J, Wintermantel E, Leong KW. Biomedical applications of  
38 polymer-composite materials: a review. *Compos Sci Technol*. 2001;61:1189-1224.  
39

40  
41  
42 Sabree I, Gough JE, Derby B. Mechanical properties of porous ceramic scaffolds:  
43 Influence of internal dimensions. *Ceramics International*. 2015;(41):8425-8432.  
44

45  
46  
47 Sachlos E, Czernuszka JT. Making tissue engineering scaffolds work. Review on the  
48 application of solid freeform fabrication technology to the production of tissue  
49 engineering scaffolds. *Eur Cell Mater*. 2003;5:29-40.  
50

51  
52  
53  
54 Salgado AJ, Coutinho OP, Reis RL. Bone Tissue Engineering: State of the Art and  
55 Future Trends. *Macromol Biosci*. 2004;4(8):743-765.  
56

1  
2  
3 Saxena AK. Tissue engineering: Present concepts and strategies. *J Indian Assoc*  
4 *Pediatr Surg.* 2005;10:14-19.  
5  
6

7  
8 Shrivats AR, McDermott MC, Hollinger JO. Bone tissue engineering: state of the  
9 union. *Drug Discov Today.* 2014;19(6):781-786.  
10

11  
12 Sing SL, Yeong WY, Wiria FE, Tay BY. (2015) Characterization of Titanium Lattice  
13 Structures Fabricated by Selective Laser Melting Using an Adapted Compressive Test  
14 Method. *Experimental Mechanics.* Advance online publication. DOI 10.1007/s11340-  
15 015-0117-y.  
16  
17  
18

19  
20 Sun W, Starly B, Nam J, Darling A. Bio-CAD modelling and its applications in  
21 computer-aided tissue engineering. *Comput Aided Des.* 2005;37:1097-1114.  
22  
23

24  
25 Taniguchi N, Fujibayashi S, Takemoto M, Sasaki K, Otsuki B, Nakamura T,  
26 Matsushita T, Kokubo T, Matsuda S. Effect of pore size on bone ingrowth into porous  
27 titanium implants fabricated by additive manufacturing: An in vivo experiment. *Mater*  
28 *Sci Eng C Mater Biol Appl.* 2016;59:690-701.  
29  
30  
31

32  
33 Van Cleynenbreugel T, Schrooten J, Van Oosterwyck H, Vander Sloten J. Micro-CT-  
34 based screening of biomechanical and structural properties of bone tissue engineering  
35 scaffolds. *Med Biol Eng Comput.* 2006;44(7):517-525.  
36  
37  
38

39  
40 Van der Putte, T. Using the discrete 3D Voronoi diagram for the modelling of 3D  
41 continuous information in geosciences. Master Thesis, Geographical Information  
42 Management and Applications. Utrecht University, Netherlands, 2009.  
43  
44

45  
46 Velasco MA, Narváez-Tovar CA, Garzón-Alvarado DA. Design, Materials, and  
47 Mechanobiology of Biodegradable Scaffolds for Bone Tissue Engineering. *Biomed*  
48 *Res Int.* 2015, Article ID 729076, 21 pages.  
49  
50

51  
52 Wang MO, Vorwald CE, Dreher ML, Mott EJ, Cheng MH, Cinar A, Mehdizadeh H,  
53 Somo S, Dean D, Brey EM, Fisher JP. Evaluating 3D-printed biomaterials as  
54 scaffolds for vascularized bone tissue engineering. *Adv Mater.* 2015;27(1):138-144.  
55  
56  
57  
58  
59  
60



1  
2  
3  
4 Wang X, Xu S, Zhou S, Xu W, Leary M, Choong P, Qian M, Brandt M, Xie YM.  
5 Topological design and additive manufacturing of porous metals for bone scaffolds  
6 and orthopaedic implants: A review. *Biomaterials*. 2016;83:127-141.  
7  
8  
9

10  
11 Wettergreen MA, Bucklen BS, Sun W, Liebschner MAK. Computer-Aided Tissue  
12 Engineering of a Human Vertebral Body. *Ann Biomed Eng*. 2005;33(10):1333-1343.  
13  
14

15  
16 Williams JM, Adewunmi A, Schek RM, Flanagan CL, Krebsbach PH, Feinberg SE,  
17 Hollister SJ, Das S. Bone tissue engineering using polycaprolactone scaffolds  
18 fabricated via selective laser sintering. *Biomaterials*. 2005;26(23):4817-4827.  
19  
20  
21

22  
23 Yan C, Hao L, Hussein A, Young P. Ti-6Al-4V triply periodic minimal surface  
24 structures for bone implants fabricated via selective laser melting. *J Mech Behav*  
25 *Biomed Mater*. 2015;51:61-73.  
26  
27  
28

29  
30 Yang S, Leong KF, Du Z, Chua CK. The Design of Scaffolds for Use in Tissue  
31 Engineering. Part I Traditional Factors. *Tissue Eng*. 2001;7(6):679-689.  
32  
33

34  
35 Yang S, Zhao YF. Additive manufacturing-enabled design theory and methodology: a  
36 critical review. *Int J Adv Manuf Technol*. 2015;80:327-342.  
37  
38  
39  
40  
41  
42  
43  
44  
45  
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## List of figures

Fig. 1 Graphical representation about the formation of a 2D Voronoi diagram

Fig.2 a) Reference cube, b) 3D Voronoi polyhedron, c) Partition in 3D Voronoi polyhedrons

Fig. 3 Workflow for the 3D Voronoi scaffold generation

Fig. 4 Partition in Voronoi polyhedrons of the bounding box volume generated from the patient anatomy

Fig. 5 a) Each face is scaled respect the face's centroid, b) each polyhedron is scaled respect the polyhedron's centroid

Fig. 6 a) Generation and deconstruction of faces and polyhedrons, b) particular of scaled faces and polyhedrons, c) quad meshes on the edges to construct the lattice

Fig. 7 a) Trabecular mesh, b) Smoothed lattice mesh after Catmull-Clark algorithm

Fig. 8 Boolean intersection of the patient bone geometry and the lattices to obtain the porous scaffold

Fig. 9 Sample of scaffold (10×10×10 mm) with 500 seeds and setting: a)  $S_f = S_v = 0.25$  (P% = 17.4), b)  $S_f = S_v = 0.50$  (P% = 47.8), c)  $S_f = S_v = 0.75$  (P% = 79.6)

Fig. 10 Effect of the scale factors  $S_f$  and  $S_v$  on the percentage porosity of the lattice (with 500 seeds)

Fig. 11 Effect of the number of seeds on the percentage porosity of the scaffold (with  $S_f = S_v = 0.75$ )

Fig. 12 Polyhedron-sphere method: a) polyhedrons scaled by  $S_v$ , b) polyhedrons scaled by  $S_v$  and corresponding spheres, c) spheres representing the pores

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3 Fig. 13 Number of Voronoi seeds estimated in relation to the target pores size for a  
4 cubic scaffold 10 mm sided with a target percentage porosity  $P\% = 80\%$   
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8 Fig. 14 Pores size (measured applying the polyhedron-sphere method) of a cubic  
9 scaffold 10 mm sided, scale factors  $S_f = S_v = 0.75$ , with a target percentage porosity  
10  $P\% = 80\%$ , generated with different target pores size (and therefore different number  
11 of seeds)  
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15 Fig. 15 Sample of scaffold (10x10x10 mm) with  $S_f = S_v = 0.75$  and setting: a) number  
16 of seeds = 200 (target pores size = 2 mm), b) number of seeds = 800 (target pores size  
17 = 1 mm)  
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22 Fig. 16 Void component (pores plus connective channels) for a Voronoi scaffold with  
23 minimal scale factors ( $S_f = S_v = 0.25$ ) and a) number of seeds = 8, b) number of seeds  
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29 Fig. 17 Mean pores interconnection and mean channels size of a cubic scaffold 10  
30 mm sided, scale factors  $S_f = S_v = 0.75$ , with a target percentage porosity  $P\% = 80\%$ ,  
31 generated with different target pores size (and therefore different number of seeds)  
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36 Fig. 18 Trabecular thickness evaluation: a) Voronoi polyhedral cells scaled by  $S_v$   
37 with the distance of the centroids of coupled faces; b) Voronoi polyhedral cells scaled  
38 by  $S_v$  with the trabecular mesh; c) the final scaffold ( $S_f = S_v = 0.75$ ; number of seeds  
39  $n = 12$ ; target pores size  $D_p = 5$  mm)  
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44 Fig. 19 Mean pores interconnection and mean channels size of a cubic scaffold 10  
45 mm sided, scale factors  $S_f = S_v = 0.75$ , with a target percentage porosity  $P\% = 80\%$ ,  
46 generated with different target pores size (and therefore different number of seeds)  
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51 Fig. 20 a) 3D generated Voronoi Scaffold, b) Healthy spongy bone (Marinozzi et al.  
52 2012)  
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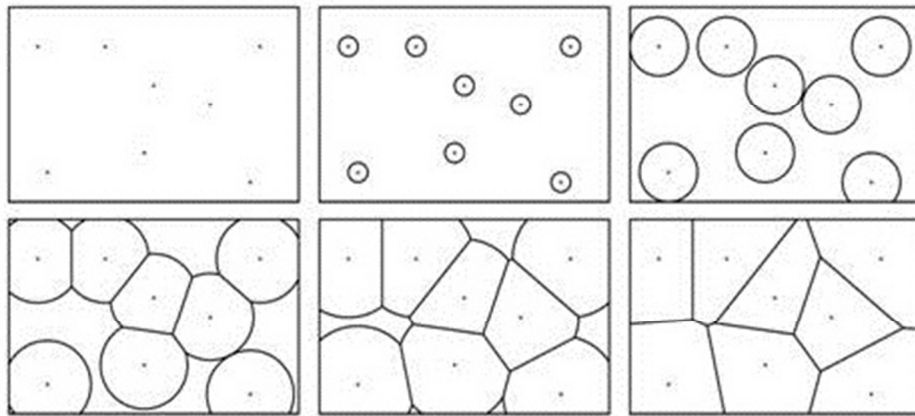


Fig. 1 Graphical representation about the formation of a 2D Voronoi diagram  
123x56mm (96 x 96 DPI)

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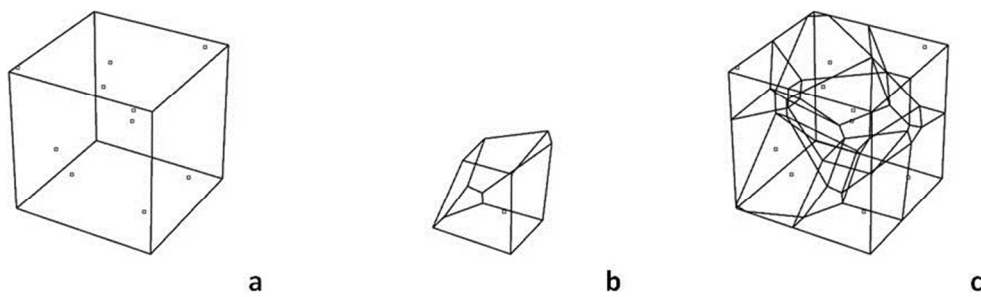


Fig.2 a) Reference cube, b) 3D Voronoi polihedron, c) Partition in 3D Voronoi polihedrons  
233x72mm (96 x 96 DPI)

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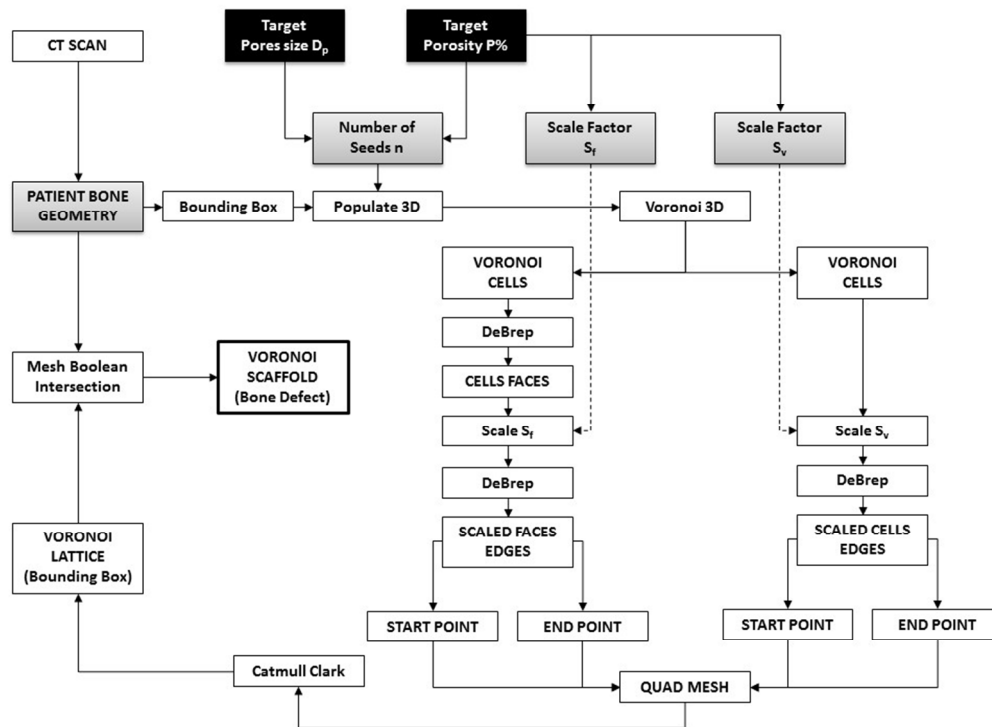


Fig. 3 Workflow for the 3D Voronoi scaffold generation  
254x190mm (96 x 96 DPI)

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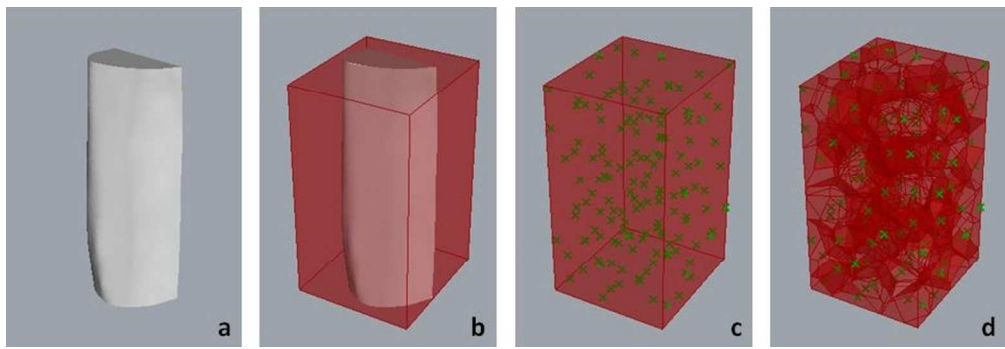


Fig. 4 Partition in Voronoi polyhedrons of the bounding box volume generated from the patient anatomy 253x86mm (96 x 96 DPI)

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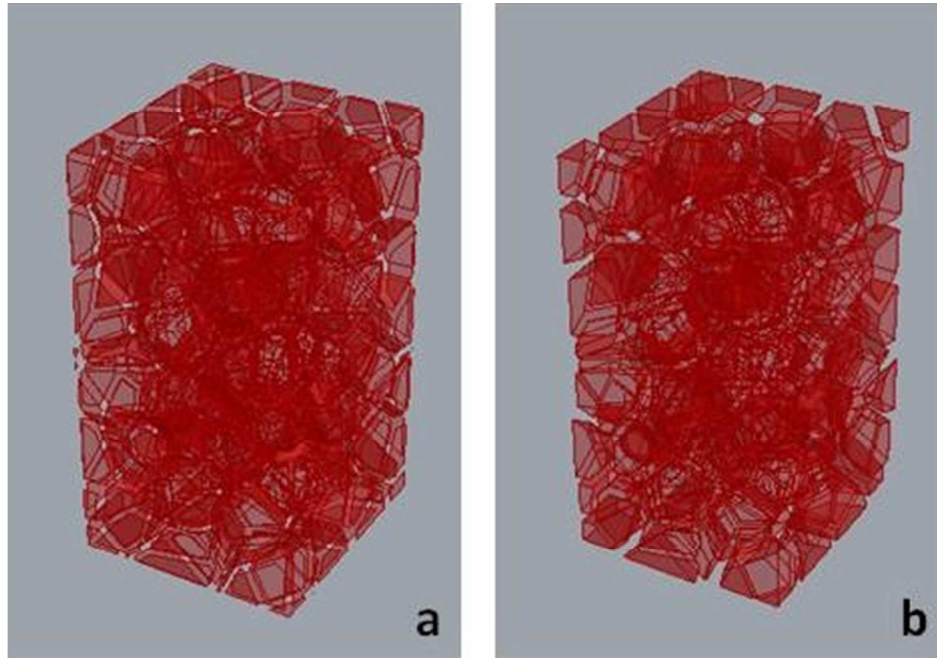


Fig. 5 a) Each face is scaled respect the face's centroid, b) each polyhedron is scaled respect the polyhedron's centroid  
124x86mm (96 x 96 DPI)



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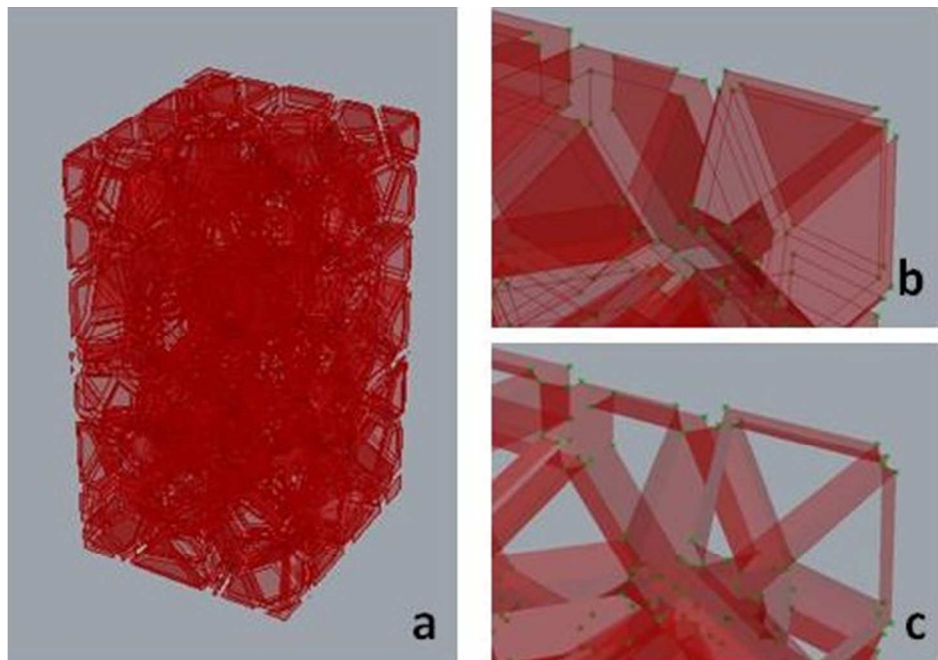


Fig. 6 a) Generation and deconstruction of faces and polyhedrons, b) particular of scaled faces and polyhedrons, c) quad meshes on the edges to construct the lattice  
124x86mm (96 x 96 DPI)

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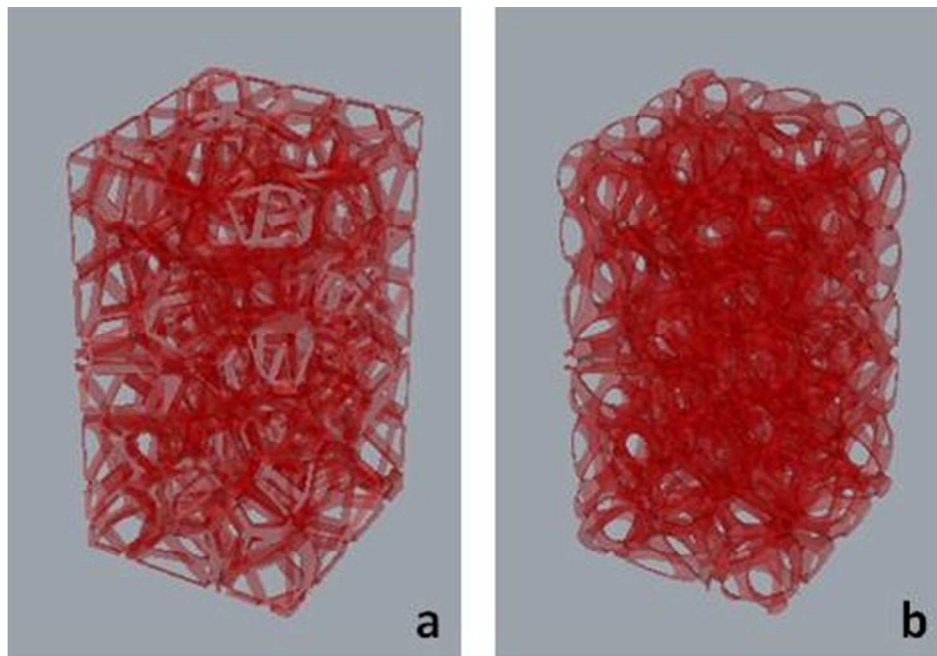


Fig. 7 a) Trabecular mesh, b) Smoothed lattice mesh after Catmull-Clark algorithm  
124x86mm (96 x 96 DPI)

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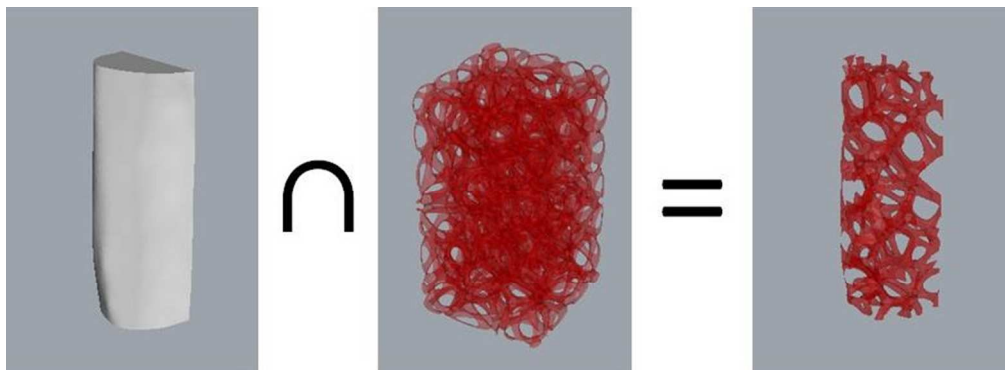


Fig. 8 Boolean intersection of the patient bone geometry and the lattices to obtain the porous scaffold  
236x86mm (96 x 96 DPI)

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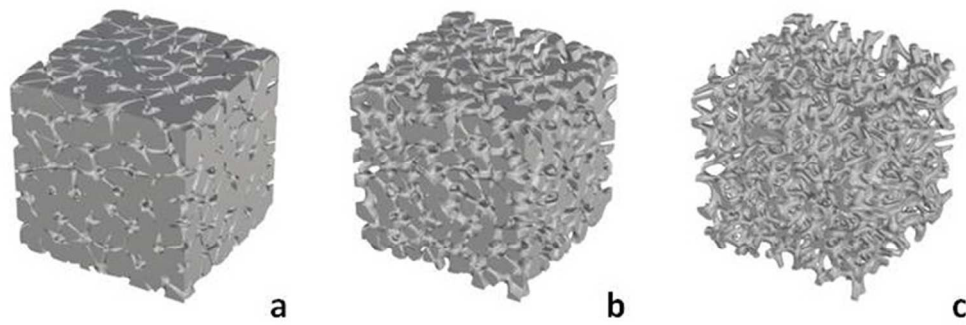


Fig. 9 Sample of scaffold (10×10×10 mm) with 500 seeds and setting: a)  $S_f = S_v = 0.25$  ( $P\% = 17.4$ ), b)  $S_f = S_v = 0.50$  ( $P\% = 47.8$ ), c)  $S_f = S_v = 0.75$  ( $P\% = 79.6$ )  
190×65mm (96 x 96 DPI)

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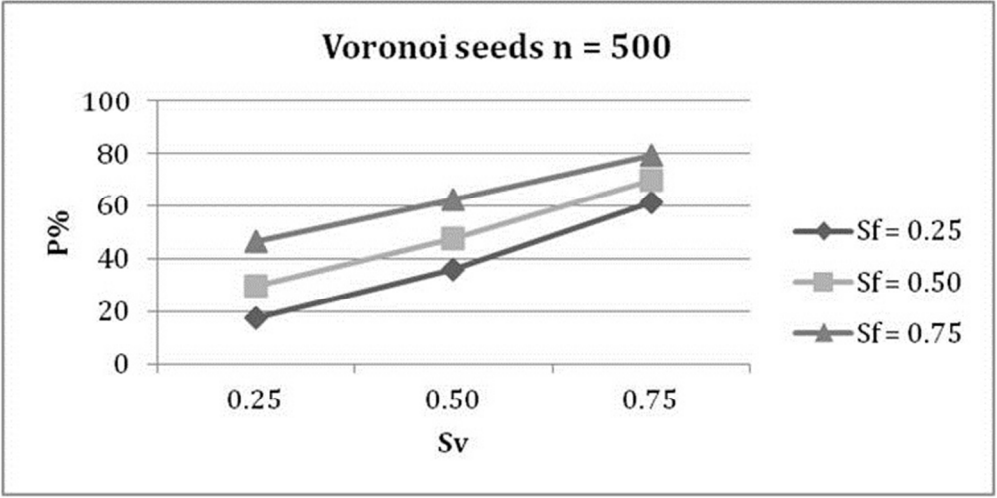
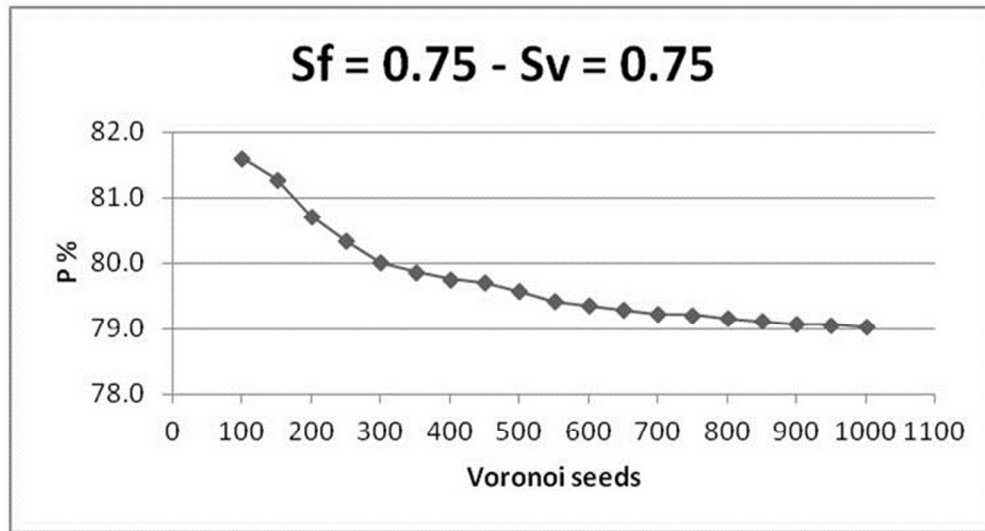


Fig. 10 Effect of the scale factors Sf and Sv on the percentage porosity of the lattice (with 500 seeds) 200x101mm (96 x 96 DPI)

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Fig. 11 Effect of the number of seeds on the percentage porosity of the scaffold (with  $S_f = S_v = 0.75$ )  
201x108mm (96 x 96 DPI)

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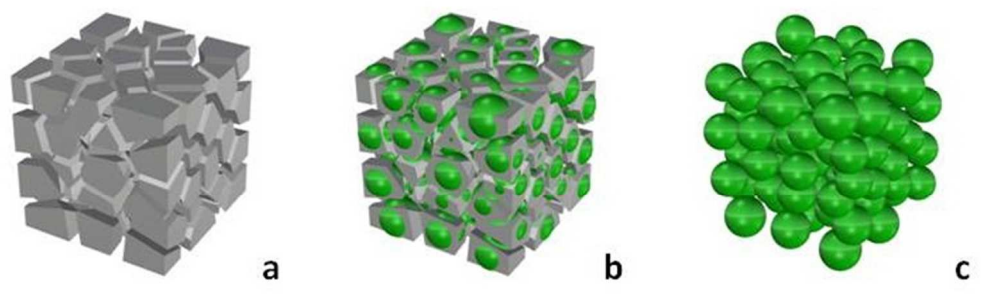


Fig. 12 Polyhedron-sphere method: a) polyhedrons scaled by  $S_v$ , b) polyhedrons scaled by  $S_v$  and corresponding spheres, c) spheres representing the pores  
187x58mm (96 x 96 DPI)

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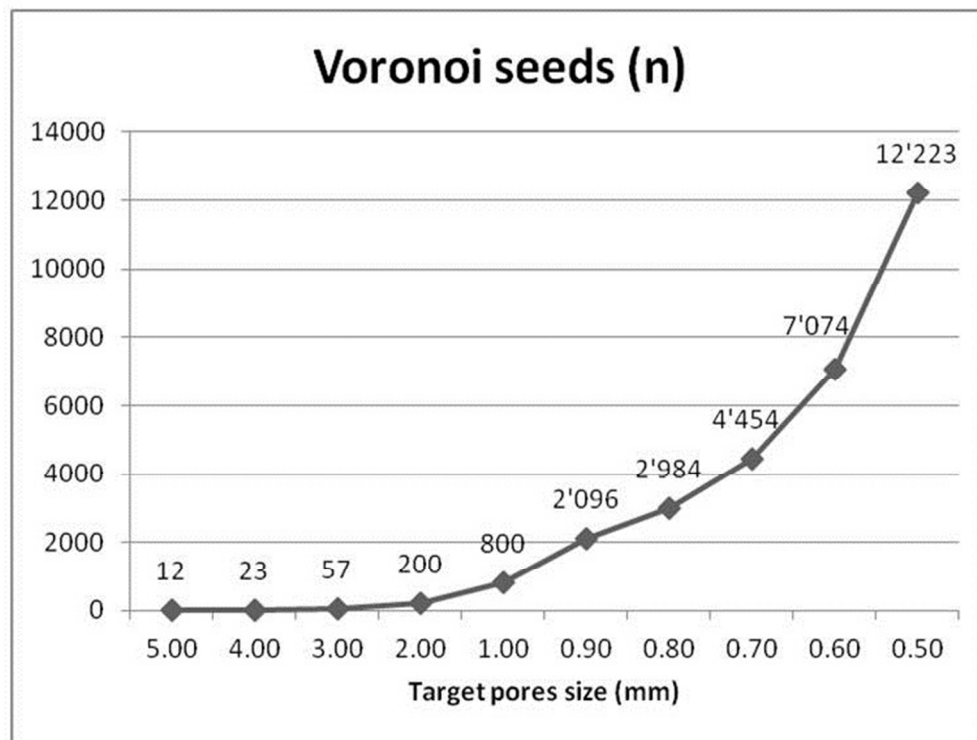


Fig. 13 Number of Voronoi seeds estimated in relation to the target pores size for a cubic scaffold 10 mm sided with a target percentage porosity  $P\% = 80\%$   
203x154mm (96 x 96 DPI)



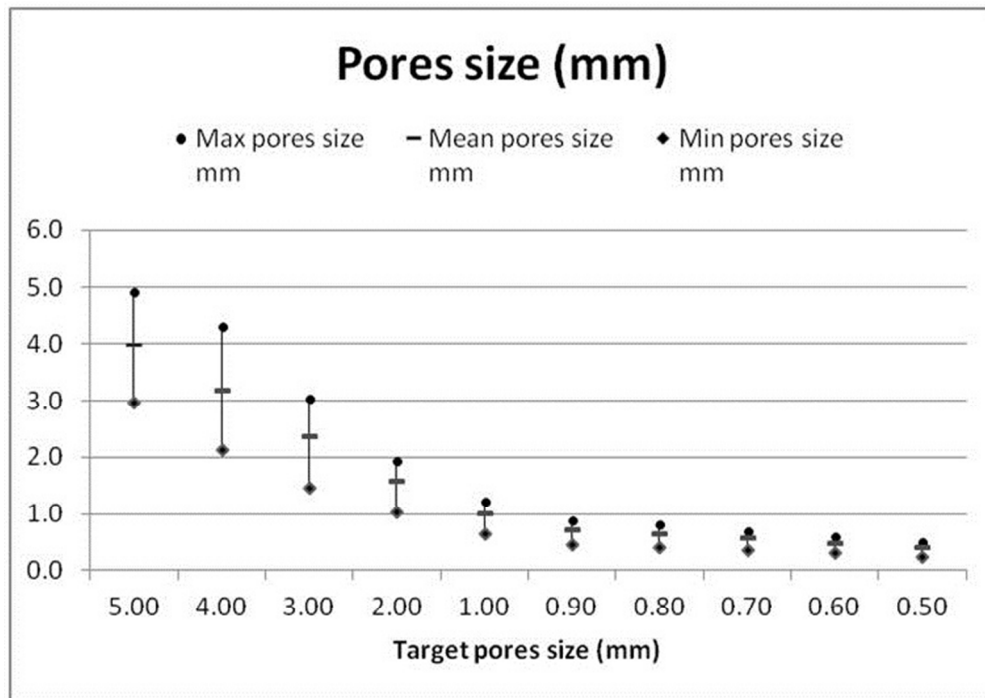


Fig. 14 Pores size (measured applying the polyhedron-sphere method) of a cubic scaffold 10 mm sided, scale factors  $S_f = S_v = 0.75$ , with a target percentage porosity  $P\% = 80\%$ , generated with different target pores size (and therefore different number of seeds)  
201x142mm (96 x 96 DPI)

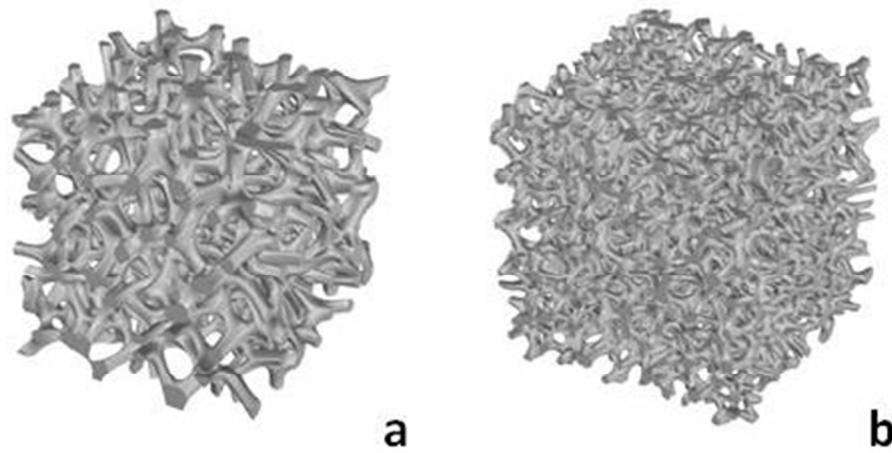


Fig. 15 Sample of scaffold (10x10x10 mm) with  $S_f = S_v = 0.75$  and setting: a) number of seeds = 200 (target pores size = 2 mm), b) number of seeds = 800 (target pores size = 1 mm)  
123x65mm (96 x 96 DPI)

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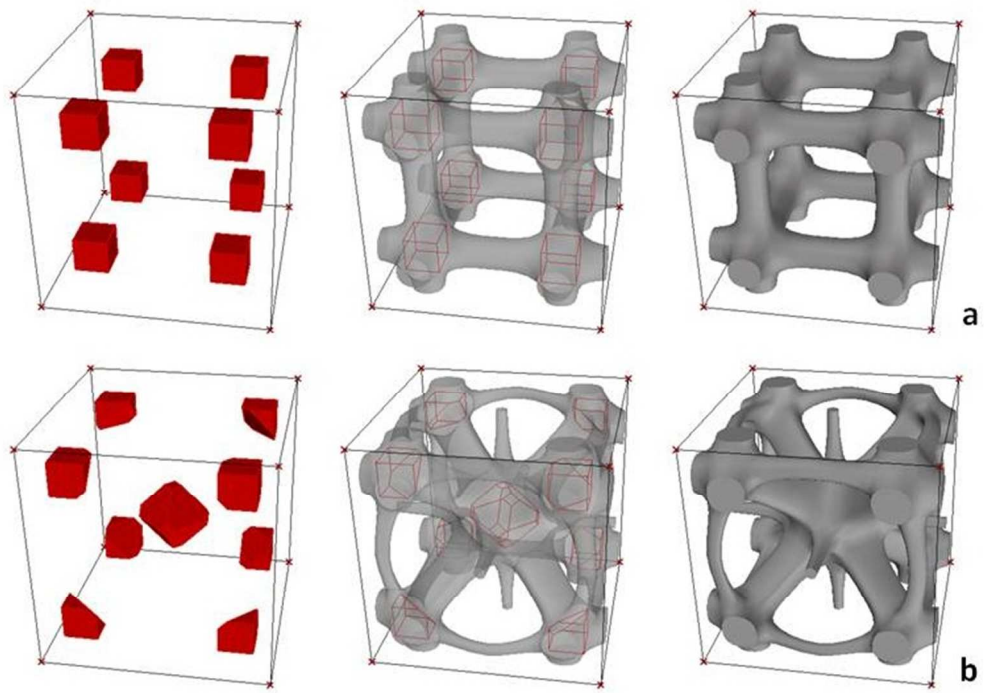


Fig. 16 Void component (pores plus connective channels) for a Voronoi scaffold with minimal scale factors ( $S_f = S_v = 0.25$ ) and a) number of seeds = 8, b) number of seeds = 9  
200x142mm (96 x 96 DPI)

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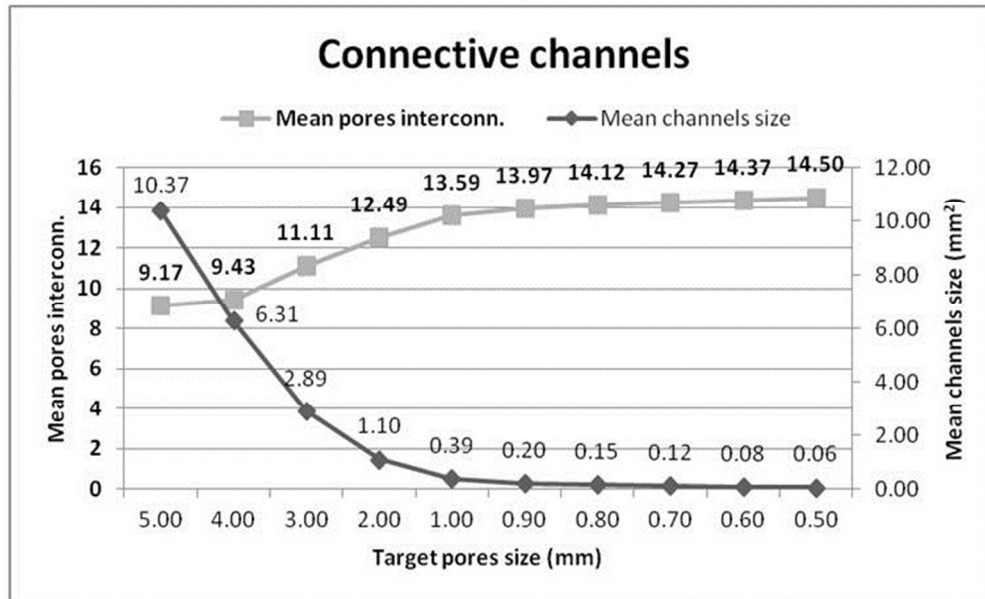


Fig. 17 Mean pores interconnection and mean channels size of a cubic scaffold 10 mm sided, scale factors  $S_f = S_v = 0.75$ , with a target percentage porosity  $P\% = 80\%$ , generated with different target pores size (and therefore different number of seeds)  
201x123mm (96 x 96 DPI)

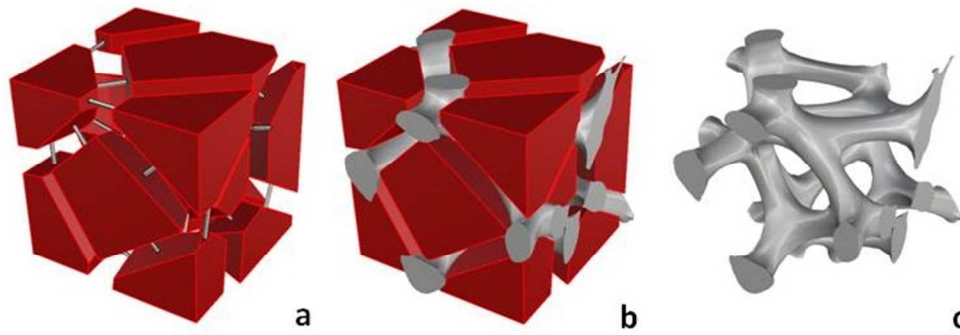


Fig. 18 Trabecular thickness evaluation: a) Voronoi polyhedral cells scaled by  $S_v$  with the distance of the centroids of coupled faces; b) Voronoi polyhedral cells scaled by  $S_v$  with the trabecular mesh; c) the final scaffold ( $S_f = S_v = 0.75$ ; number of seeds  $n = 12$ ; target pores size  $D_p = 5$  mm) 203x73mm (96 x 96 DPI)

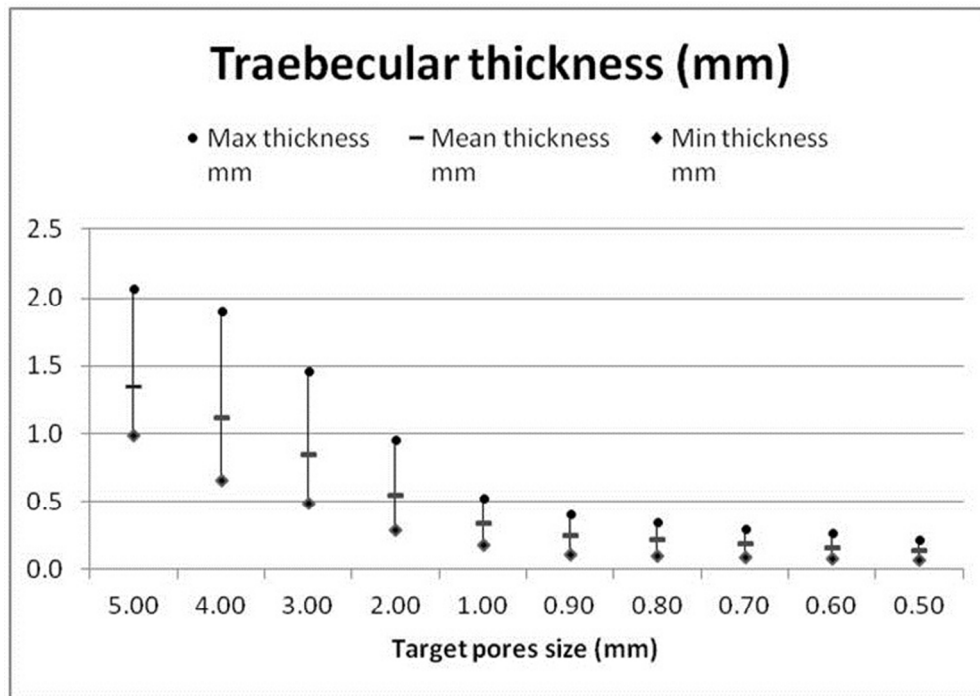


Fig. 19 Mean pores interconnection and mean channels size of a cubic scaffold 10 mm sided, scale factors  $Sf = Sv = 0.75$ , with a target percentage porosity  $P\% = 80\%$ , generated with different target pores size (and therefore different number of seeds)  
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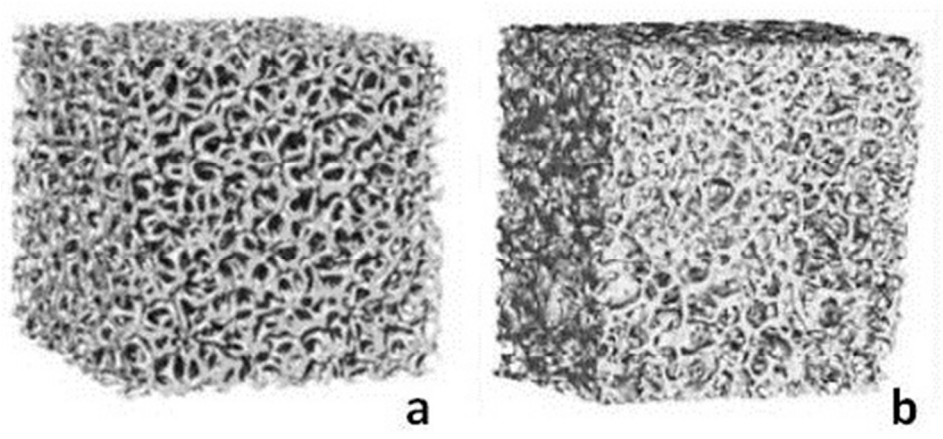


Fig. 20 a) 3D generated Voronoi Scaffold, b) Healthy spongy bone (Marinozzi et al. 2012)  
125x63mm (96 x 96 DPI)

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