Computer Simulation of Scaffold Degradation

G Erkizia\textsuperscript{1}, A Rainer\textsuperscript{2}, E M de Juan-Pardo\textsuperscript{1} and J Aldazabal\textsuperscript{1}

\textsuperscript{1} CEIT and Tecnun (University of Navarra), Manuel Lardizábal 15, 20018 San Sebastián, Spain

\textsuperscript{2} CIR - Laboratory of Chemistry & Biomaterials, University Campus Bio-Medico of Rome, Via Alvaro del Portillo 21, Rome, Italy

E-mail: gerkizia@ceit.es

Abstract. Scaffolds are porous biocompatible materials with suitable microarchitectures that are designed to allow for cell adhesion, growth and proliferation. They are used in combination with cells in regenerative medicine to promote tissue regeneration by means of a controlled deposition of natural extracellular matrix by the hosted cells therein. This healing process is in many cases accompanied by scaffold degradation up to its total disappearance when the scaffold is made of a biodegradable material.

This work presents a computational model that simulates the degradation of scaffolds. The model works with three-dimensional microstructures, which have been previously discretised into small cubic homogeneous elements, called voxels. The model simulates the evolution of the degradation of the scaffold using a Monte Carlo algorithm, which takes into account the curvature of the surface of the fibres.

The simulation results obtained in this study are in good agreement with empirical degradation measurements performed by mass loss on scaffolds after exposure to an etching alkaline solution.

1. Introduction

In recent years, biomedical research has been developing strategies to regenerate tissues in the search for new treatments for damaged or diseased organs. Some of these strategies, included in the so-called regenerative medicine field, are the replacement, repair and/or regeneration of tissues (e.g. bone, cartilage, heart valves, bladder, etc.). These tissues substitutes need to fulfil certain requirements on the mechanical and structural properties in order to work properly.

Within the field of regenerative medicine, Tissue Engineering applies engineering principles to produce biological substitutes that maintain, improve or restore the function of organs or tissues of the human body [1]. New biomaterials are often used to replace the damaged part of an organ that has ceased in its functions, or in other cases, as vehicles to transport cells and molecules to the target tissue or organ. These new tissular substitutes act as “scaffolds” that allow for cell growth and formation of natural extracellular matrix.

For a correct operation, the artificially designed scaffolds must be biocompatible, should not present any adverse immunological reaction and in most cases need to be gradually degraded as new biological tissue is formed, thus transferring the withstand of the mechanical loads progressively to the newly formed autologous tissue. The main objective of this work is to model the degradation of scaffolds by hydrolysis. Such degradation will take place in the gradual replacement of a scaffold during bone regeneration.
The time required from the design of a biomaterial scaffold to its clinical application is very long, since it requires several validation steps, thus increasing the overall costs of the final product. It has been estimated that the average development period for a tissue engineering product can range up to 10 years. Therefore, substantial investments are required, in terms of laboratory equipment, men power and validation tests, until the commercialization of the product is reached.

In this framework, it is clear how *in silico* experiments, based on accurate computer models, could help reduce the overall costs of final tissue engineering products in a drastic way.

2. Model

The model proposed in this work was coded in C and was executed in a Linux workstation (DELL Precision T5400). Following subsections show the ideas and principles behind each part of the model.

2.1. Initial microstructure

The model works on scaffolds consisting of equally spaced parallel layers of fibres. The initial microstructure is defined analytically by several parameters: volume size, number of layers, fibres diameter, spacing between fibres in the same layer and tilt angle of the fibres on each layer. The model assumes that the distance between two consecutive layers is zero, i.e., consecutive layers touch each other physically.

2.2. Volume discretisation

Once the microstructure is defined, it has to be discretised into cubic elements. It is assumed that the total volume is composed of many identical cubes, called voxels. Each of these voxels may contain information about the corresponding phase -matrix or fibre- and, in the latter case, to which fibre it belongs. For each voxel, the concentration of the component fibre can also be determined.

The accuracy of the discretisation process can be adjusted by controlling the size of voxels. Certainly, the number of voxels used for discretising a given volume strongly affects computational effort and requirements.

Working with cubic elements has the following advantages [2]:

- All voxels are of equal size.
- The number of neighbours for all voxels is constant, as well as the area shared with each surrounding neighbour.
- For a voxel, the position of the vertices can be derived from the coordinates of its centre.
- Coding benefits due to the simple geometry of a voxel.

2.3. Boundary conditions

Periodical boundary conditions cannot be systematically applied as a rule to a microstructure. The geometry of the scaffold will depend on the number of fibres per layer and the tilt angle. Once these parameters are fixed, it is frequent that the final microstructure is not periodic. As a consequence of this lack of periodicity, a special set of boundary conditions must be applied, where the simulated volume should be extended over a voxel on each side, that is, if the domain size along the *i* axis is *l_i* the model should work with a volume with a dimension of *l_i* + 2 voxels. In this way, the so called extended volume is obtained to define the boundary conditions of the simulation. The extended faces parallel to fibre layers are considered in the model as liquid phase, i.e., there are no fibres, while on the other four sides the extended volume is completely filled with “fibres voxels”.
Figure 1 represents the geometric relationship between the simulated volume and the extended volume. The simulated volume is constituted by different fibre layers, as shown in the first image (a). Parallel to these layers, the extended volume is filled by the liquid phase (b) and perpendicular to the layers (c) the extended volume is filled with a solid phase, i.e. with fibre voxels.

![Figure 1](image.png)

**Figure 1.** Construction of the extended volume around the simulated volume. Different boundary conditions for the extended volume are reported (b,c).

2.4. Monte Carlo Method

In this work we have used the Monte Carlo Q-Potts algorithm to simulate the degradation of scaffolds [3].

Once the initial three-dimensional microstructure has been discretised, the Monte Carlo method randomly selects a voxel within the simulated volume (not within the extended volume). The dissolution probability of a voxel of the scaffold is related with the status (solid or liquid) of its surrounding neighbours. The algorithm works by counting how many neighbours of each phase are present. For example, if the chosen voxel is solid, i.e., belongs to a fibre, and has many liquid neighbours, its dissolution probability is high. If the voxel belonging to the solid phase is inside the fibre, i.e. all its neighbours are solid, it will never dissolve. This process is repeated for all voxels in the simulated volume.

In this kind of models it is very common to measure the magnitude of the simulation time using as a time unit Monte Carlo Steps (MCS). A MCS is reached when all voxels of the domain have been chosen randomly once, i.e. the whole volume was sampled. One MCS is equivalent to a number of trials equal to the total number of voxels.

2.5. Assigning weights to neighbours

There are, in total, three types of neighbours: face, edge or vertex neighbours. Since the influence of each type of neighbour is different, different weights should be assigned to each one. The proposed model considers only two types of neighbours: face and edge neighbours. The weight assigned to each type of neighbour is inversely proportional to the distance between the voxel and the neighbour. These weights are shown in Table 1 [4].

2.6. Assigned probabilities

Before defining the probability distribution function (PDF), some considerations are needed.

When a voxel is completely surrounded by solid neighbours, i.e. there is no surrounding liquid, it cannot be dissolved, so its dissolution probability is 0.
Table 1. Normalised neighbour weights

| Neighbour type | Number of neighbours | Weight   | Normalised weight |
|----------------|----------------------|----------|-------------------|
| Face           | 6                    | 1        | 0.069             |
| Edge           | 12                   | $1/\sqrt{2}$ | 0.049             |
| Maximum weight | 6$(1 + \sqrt{2})$   | 1.000    |                   |

\[ P_d(0) = 0 \]  
\[ P_d(1) = 1 \]  

On the other hand, when a solid voxel is in the middle of the liquid, it will be always dissolved.

Summarising, we can state that the probability of dissolution, \( P_d \), depends on the number of liquid neighbours, \( n \). Between the two extreme cases previously described, there are other key configurations to determine the PDF.

In Figure 2a, the model tries to dissolve the voxel at the surface. This configuration has a normalised number of liquid neighbours \( n \) equal to 0.26, therefore it will sometimes be allowed to initialize the degradation of the flat surfaces.

Figure 2b shows another key case. This configuration corresponds to a thin edge with 0.60 normalized neighbours. The probability of dissolution assigned to this case is equal to one: that is, if a voxel is chosen randomly within an edge like this will always dissolve.

\( P_d(0.26) = 0.1 \)  

Several tests were performed to ensure an isotropic behaviour. This behaviour was obtained assigning a probability of 0.1 to the configuration shown in Figure 2a [5].

\[ P_d(0.26) = 0.1 \]  

Once these key cases were studied a linear probability distribution between these two cases was assigned, as shown in Figure 3.
2.7. Degradation

The degradation simulated in this model is hydrolysis, which is related to the contact surface between fibres and liquid.

As mentioned above, the method used to degrade the fibres in the microstructure is based on the Monte Carlo algorithm.

In this method, the standard number of liquid neighbours is measured. As shown in Figure 3, it is possible to know the probability of degradation of a selected voxel. The number of liquid neighbours for a specific voxel will determine its probability of degradation, using a randomly generated number between 0 and 1 as a comparator. If the probability to dissolve is greater than the comparator, the voxel will be degraded, i.e. converted into liquid; and if the probability is lower, the voxel will remain in its previous configuration, i.e. as a solid fibre.

3. Results

To validate the model, results of the simulations were compared with empirical degradation experiments of a scaffold made of poly-ε-caprolactone/β-tricalcium phosphate (PCL/TCP). PCL is a semi-crystalline and biodegradable polymer belonging to the family of aliphatic polyesters, while TCP is a biocompatible and bioactive ceramic material with a stoichiometry similar to the mineral phase of bone. It is widely used as a scaffold in bone tissue engineering due to its osteoconductivity and its ability to bond directly to bone. The empirical data were obtained from the literature, relative to a scaffold degradation in a diluted NaOH solution, a process commonly used to accelerate the scaffold degradation by hydrolysis in in-vitro experiments [6].

The simulated geometry of the scaffold was generated modelling the real microstructure of the scaffold used in the empirical tests performed in [6], as shown in Figure 4.

The model was adjusted to generate a scaffold of a fibre diameter of 266 μm and a scale of 20 μm/voxel, while the cube side was adjusted to a simulation volume 4000 μm. 15 multifibre-layers were arranged with a tilt angle of 90°, 30° and -30°, and distance between fibres belonging to the same layer of 866 μm.

Once the microstructure was generated, it was introduced into the model to simulate its degradation. Figures 5 and 7 show the evolution of the geometry obtained by the model during the degradation. From these results, it is possible to confirm that the degradation process obtained with the model is homogeneous.

In particular, Figure 5 shows a comparison between the empirical data obtained on real
scaffolds (SEM micrographs) and the results of the simulation process, in the 0-48 hours time range. Results confirm that the simulated degradation matches very well the empirical degradation data.

Given the correctness of the qualitative behaviour of the model, we tried to quantify the degradation process by estimating the evolution of the volume fraction of the solid phase. In order to easily compare computational results with empirical data, a conversion rate from simulated to real-time was estimated: 1 MCS ≈ 4.16 hours.

Figure 6 shows the empirically measured values for volume fraction versus the results of the computational model.

From Figure 6, it is possible to conclude that the model adjusts very well to empirical data.
Figure 6. Evolution of volume fraction of fibres: comparison of the results obtained by the computational model (dividing the number of voxels belonging to fibres by the total number of voxels) versus the empirical mass loss measurements obtained by Lam et al in [6].

Figure 7. Initial and final steps of the 3D simulation.

4. Conclusions
The proposed model is capable of generating solid geometries which resemble real scaffold microstructures.

The proposed degradation algorithm is isotropic and reasonably matches the empirical results for the degradation of scaffolds under accelerated hydrolysis tests. Yet, validation of the model against other real geometries needs to be performed.

References
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