The necessity of X-ray computed tomography to reveal the hierarchical structure of materials

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Abstract

Two recent developments of µCT to reveal the hierarchical structure of materials are presented. In the first case study, the determination of the permeability of a non-crimp carbon textile reinforcement using simulations of the fluid dynamics with µCT based voxel models is revealed. The voxel models are constructed from the µCT images of the material samples using a statistical algorithm for image segmentation, based on the Gaussian mixture model. The method is architecture independent and allows studying the variability of the permeability for different unit cells of the textile. In the second case study, the use of contrast agents for the visualization and quantification of soft biological tissues is described. More specifically, contrast-enhanced µCT (CE-µCT) facilitates the distinction between bone from cartilage, bone marrow and blood vessels in implanted biomaterials that were seeded with osteoprogenitor cells, to be screened as potential therapy for the healing of large bone defects.

Keywords: hierarchical structure, materials, X-ray computed tomography

1 Introduction

X-ray computed tomography is a very important tool for doctors, material scientists, geologists, biologists, civil engineers, bioengineers, dentists, etc., all dealing with materials of which the fine internal structure or the changes within the material are of utmost importance to understand the behaviour of the material or to have insights in the processes going on. X-ray CT is now well accepted in those disciplines as well as submicron or nano tomography facilities. In this paper details are given of two recent case studies, obtained at the Department of Materials Engineering, to reveal the hierarchical structure of materials (textiles and biological tissues) for behaviour studies or modelling purpose or materials optimization.

2 Materials and Methods

2.1 Permeability of a non-crimp carbon textile reinforcement

The material used in the study is a non-crimp carbon/epoxy composite from Saertex (540 g/m\(^2\), +45/-45, French stitch). The manufactured test plate had a thickness of 4.0 mm and a resulting fibre volume fraction of 45.5\%. The data on the permeability of the studied NCF reinforcement is presented in [1]: at a fibre volume fraction of 50.8\% the saturated permeability was measured as 0.5×10\(^{-4}\) mm\(^2\) (in the 45\(^\circ\) direction to the production direction). Based on linear fit of the fibre volume fraction – log (permeability) dependencies presented in [2] for non-crimp fabrics, similar to the one studied here, the permeability at fibre volume fraction of 45.5\% can be estimated as (1…2)×10\(^{-4}\) mm\(^2\). Three samples of different size were cut from the plate and scanned with a Nanotom S system (GE Measurement and Control Solutions, Germany). The total number of unit cells in the scanned samples is 27.

In order to construct the voxel models, the µCT images of the material samples were segmented using a method proposed in [3]. The segmentation is based on two feature variables calculated from the image: average grey value and structural anisotropy. The structural anisotropy is calculated based on the structure tensor [4]. Based on the two variables, a statistical Gaussian mixture model is constructed, which consists of a set of two-dimensional Gaussian distributions for each component of the model (fluid/solid). The voxel model is constructed through classifying each voxel of the model to one of the components, based on the maximum probability, where the probabilities are calculated from the statistical model and the values of the feature variables at each voxel. Permeability calculations were performed with FlowTex software [5], developed at KU Leuven in collaboration with the Institute for Numerical Simulation at the University of Bonn.

2.2 Contrast-enhanced µCT (CE-µCT)

Organs and biological tissues have a spatial heterogeneity. Thus, 2D measurements like histomorphometry only partially reveal the degree of change during tissue formation and integration. As a solution, the use of µCT combined with tissue-specific X-ray opaque contrast agents (i.e. contrast-enhanced µCT – CE-µCT) is put forward. This combination allows 3D visualization and quantification of both mineralized and soft tissue formation. To validate the potency of CE-µCT as virtual histology tool for skeletal tissues, this study focuses on bone tissue engineering (TE), a multidisciplinary field of science focusing on healing large

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bone defects. It typically involves a 3D carrier material (scaffold) with cells, soluble signaling molecules or their combination (i.e. TE construct), to support bone forming processes [6]. The combination of a calcium phosphate/collagen composite scaffold, a growth factor (bone morphogenetic proteins - BMP) and human periosteal derived cells (hPDCs) at 2, 4 and 6 weeks post implantation in an ectopic nude mouse model was evaluated. All samples were first imaged without contrast agent (isotropic voxel size 3 µm). Then Hexabrix® 320 (Guerbet Nederland B.V., The Netherlands) was used as contrast agent and the samples were scanned again. Finally, after washing overnight, all samples were immersed in phosphotungstic acid (PTA) and scanned a third time. Avizio® Fire (FEI Visualization Sciences Group), DataViewer (Bruker MicroCT, Belgium) and Mimics (Materialise, Belgium) were applied for 3D image processing and visualization of the different CE-CT datasets. After CE-CT imaging, the samples were fixed, decalcified and embedded in paraffin and 5 µm sections were stained with haematoxylin and eosin (H&E) and Toluidine Blue. The histological sections were matched with the corresponding CE-CT images using DataViewer and an in-house developed MatLab tool.

3 Results

3.1 Permeability of a non-crimp carbon textile reinforcement
The permeability varies quite significantly across the unit cells, in the range of (0.5…3.5)×10⁻⁴ mm², which is however in a good agreement with the experimental data (1.0…2.0×10⁻⁴ mm²). Figure 1 shows average values of predicted permeability over the unit cells in the samples. Analysis of the correlation of permeability with the solid volume fraction in the unit cell models showed a significant negative correlation, i.e. permeability is lower with a higher solid volume fraction.

3.2 Contrast-enhanced µCT (CE-CT)
By comparing to Toluidine Blue staining, we show that Hexabrix®, a negatively charged iodinated contrast agent, allows visualizing glycosaminoglycan-containing tissues such as cartilage. H&E staining confirmed that PTA stains bone marrow, but does not stain fat cells and large blood vessels (Fig. 2). Based on their different size and interconnectivity, the 3D blood vessel network and fat cells in the bone marrow compartment were individually distracted and quantified.

Acknowledgements
The first case study has received funding from the EU Seventh Framework Programme for research, technological development and demonstration under grant agreement no ACP2-GA-2012-314562-QUICOM, µCT measurements of the carbon textile reinforcement were done by Ch. Hahn, E. Winterstein (TU Munich) and B. Plank (University of Applied Sciences Upper Austria). For the second case study, the Research Foundation - Flanders (FWO/12R4315N) is acknowledged. This work is part of Prometheus, the Leuven Research & Development Division of Skeletal Tissue Engineering of the KU Leuven: www.kuleuven.be/prometheus. The CE-CT images have been generated on the X-ray computed tomography facilities of the Department of Materials Engineering of the KU Leuven, financed by the Hercules Foundation (project AKUL 09/001: Micro- and nano-CT for the hierarchical analysis of materials).

References

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