Generation of terminal airway skeletons using synchrotron based X-ray tomographic microscopy

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Introduction

SUBTLE differences in the branching pattern of the acinar airways are crucial for airflow and particle deposition in the pulmonary gas-exchange area. Until now, the generation of skeletons of the gas-exchanging airways was limited by the resolution of the available three-dimensional imaging methods.

USING wide field synchrotron radiation based X-ray tomographic microscopy (SRXTM, Poster I-7 and [1]), we generated large high resolution three dimensional datasets of heavy metal stained and paraffin embedded rat lung samples [2] at an isometric voxel length of 0.74 µm.

Materials and Methods

AT the beamline TOMCAT [4] at the Swiss Light Source (Paul Scherrer Institut, Switzerland) we obtained tomographic datasets of the distal-medial edge of the right lower lung lobe obtained at post-natal days 4–60.

INDEPENDENT acini have been extracted from the lung samples using a region growing algorithm. The acinar skeleton was extracted using a successive erosion technique based on the distance transformation [3] of the extracted segments (see figure 1). The three-dimensional topology of the resulting skeleton corresponds to the extracted acinus (see figure 2). All calculations and visualizations have been made with MeVisLab (Version 1.6.1, MeVis Research GmbH, Bremen, Germany).

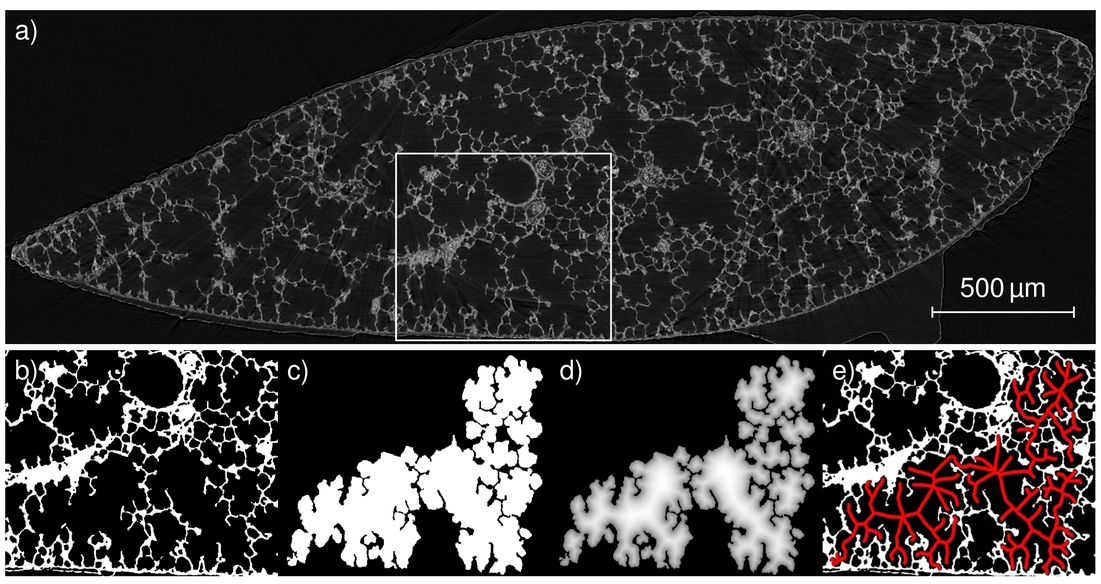

Figure 1: *Two-dimensional skeletonization process: a): Wide field scanned tomographic slice from a rat lung sample obtained at postnatal day 21. The inset corresponds to the region of the images shown in the bottom row. b): Binarized region of interest (ROI). c): Filled structure inside the ROI, representing one connected airspace in two dimensions. d): Distance transformation. e): The local maxima of the distance transformation form the skeleton (overlay on binarized lung structure).*

Results

TOMOGRAPHIC datasets covering a cylindrical field of view with a height of 1.4 mm and a diameter of approximately 4 mm have been obtained. We extracted three independent central terminal airway segments from all samples and the skeletons of these segments have been calculated and visualized as shown in figures 2 and 3.

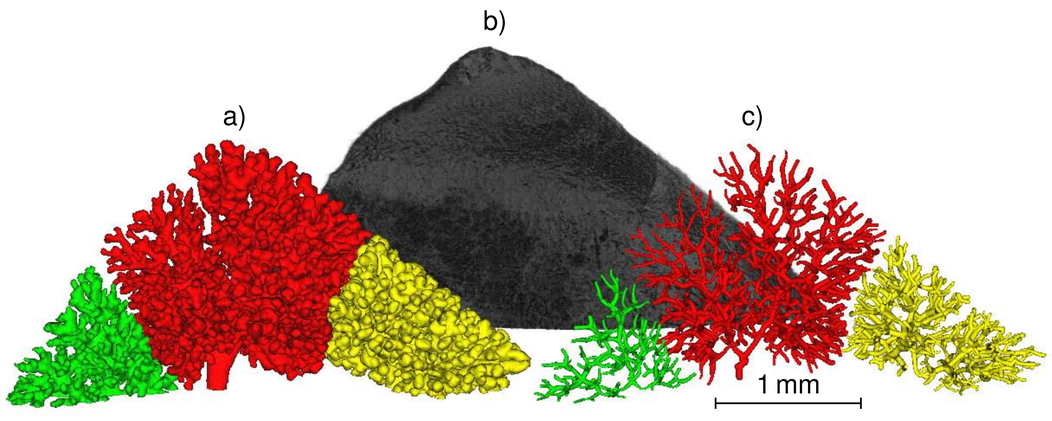

Figure 2: *Overview of a three-dimensional dataset of a Sprague Dawley rat lung sample obtained at postnatal day 4. a) Three independent airway segments have been extracted. The green segment contains one partially cut acinus, the red segment contains two entire acini and the yellow segment contains one complete and one partially cut acinus. b) Three-dimensional view of the full sample. c) Skeletons of segmented acini.*

THE full description of the skeleton has been obtained as an XML-File, which permits us to analyze the amount and positions of the nodal points of the branches of the extracted skeleton. The complexity of the skeletons can thus be analyzed, e.g. by calculating the nodal point or segment density for each sample.

Results (cont.)

WE have successfully extracted airway segments corresponding to acini from multiple datasets as shown in figure 3.

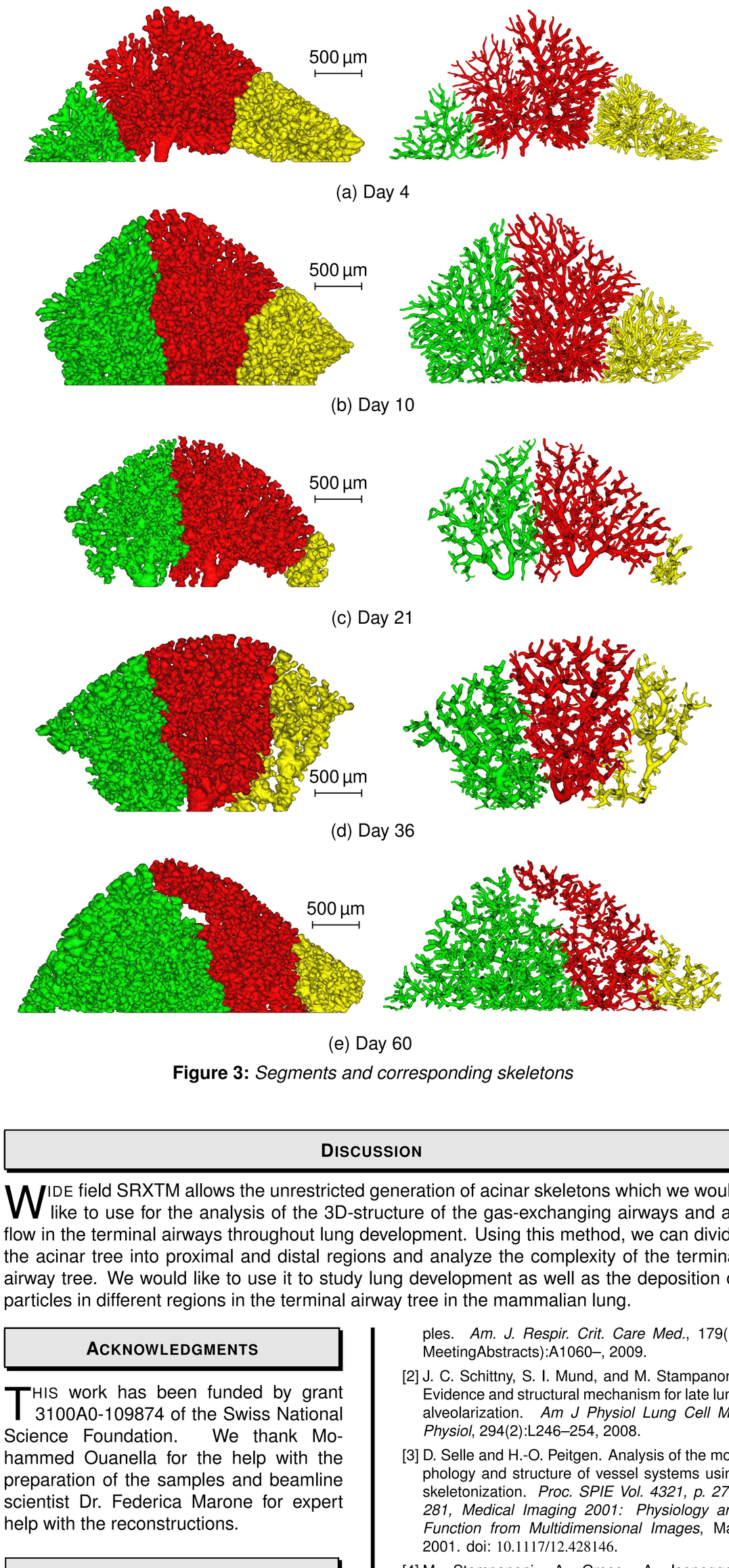

Figure 3: *Segments and corresponding skeletons*

Discussion

WIDE field SRXTM allows the unrestricted generation of acinar skeletons which we would like to use for the analysis of the 3D-structure of the gas-exchanging airways and air flow in the terminal airways throughout lung development. Using this method, we can divide the acinar tree into proximal and distal regions and analyze the complexity of the terminal airway tree. We would like to use it to study lung development as well as the deposition of particles in different regions in the terminal airway tree in the mammalian lung.

Acknowledgments

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References

[1] D. Haberthür, C. Hintermüller, J. C. Schittny, and M. Stampanoni. Quality Guided Synchrotron Radiation Based X-Ray Tomographic Microscopy of Large Lung Samples. *Am. J. Respir. Crit. Care Med.*, 179(1-MeetingAbstracts):A1060–, 2009.

[2] J. C. Schittny, S. I. Mund, and M. Stampanoni. Evidence and structural mechanism for late lung alveolarization. *Am J Physiol Lung Cell Mol Physiol*, 294(2):L246–254, 2008.

[3] D. Selle and H.-O. Peitgen. Analysis of the morphology and structure of vessel systems using skeletonization. *Proc. SPIE Vol. 4321, p. 271-281, Medical Imaging 2001: Physiology and Function from Multidimensional Images*, May 2001. doi: 10.1117/12.428146.

[4] M. Stampanoni, A. Groso, A. Isenegger, G. Mikuljan, Q. Chen, D. Meister, M. Lange, R. Betemps, S. Henein, and R. Abela. TOMCAT: A beamline for TOmographic Microscopy and Coherent rAdiology experimenTs. *AIP Conference Proceedings*, 879(1):848–851, 2007. doi: 10.1063/1.2436193.