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Accuracy Of Whole Slide Imaging Stack Alignment In Consecutive Sections Of The Carotid Artery

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Introduction/ Background

Atherosclerosis is a chronic inflammatory disease of middle-sized and large arteries, characterized by the accumulation of inflammatory cells, especially macrophages [1] . A detailed visualization of the presence and distribution of macrophages in the atherosclerotic plaque contributes to a better understanding of the pathogenesis of atherosclerosis and the onset of acute coronary syndromes after atherosclerotic plaque rupture. Three-dimensional (3D) reconstruction of histology sections has the potential to improve both the detection of lesions as well as understanding in plaque growth and destabilization.

Aims

The objective of this study is to implement a image marker independent 3D histology reconstruction method in order to visualize the arteriosclerotic vessel and evaluate its accuracy.

Methods

A dataset comprising 48 consecutive cross-sections with a slice thickness of 10µm of a formalin-fixed paraffinembedded (FFPE) carotid artery was used. The slideswere double stained with monoclonal antibodies and were scanned with an Olympus dotSlide scanner with a 10x objective leading to 0.65 micron pixel size. In these images, the smooth muscle cells and macrophages were visualized in blue and red, respectively. Rigid, rigid & affine, and rigid & affine & b-spline (non-rigid) automatic stack alignment was performed using elastix, an open-source toolbox for alignment of images [2]. As a consequence of the image deformation in non-rigid approaches, the diagnostic accuracy might be hindered. Therefore a small bending energy, i.e., sum of the spatial second-order derivatives of the transformation, was al- lowed. In order to increase processing speed, the stack alignment was performed on downsampled data. An automatically determined mask of the vessel was used for pair-wise reconstruction of the vessel with respect to the middle slide that was chosen as a reference section. Accuracy was visually assessed using a surface plot of the lumen of the vessel. In addition, the Dice similarity coefficient, which is a measure of spatial image overlap, of consecutive pairs of slides was calculated for the different stack alignment approaches.

Results

Visual assessment of the surface plot of the vessels' lumen after pair-wise stack alignment, showed a relatively smooth surface of the lumen. This was the case for the rigid (i.e. translation and rotation), rigid & affine, and rigid & affine & b-spline approaches. The Dice similarity coefficient of the registered masks increased with each additional alignment step. Slides alignment using rigid, rigid & affine and rigid & affine & b-spline approaches resulted in average Dice similarity coefficient of 0.85, 0.87, and 0.98, respectively. A more accurate result of the alignment comes at the cost of an increase in computation time by roughly a factor of two in each additional alignment step.

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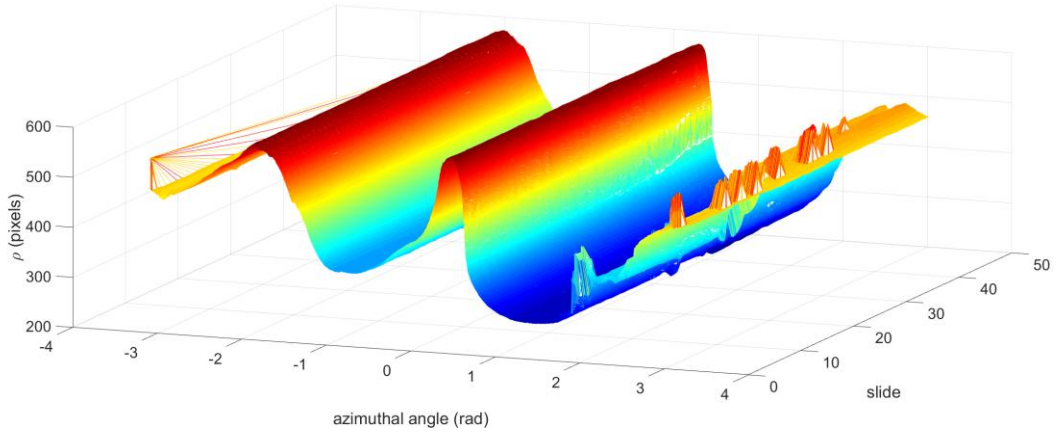

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