Automatic Repair of Acquisition Defects in Reconstruction of Histology Sections of a Human Brain

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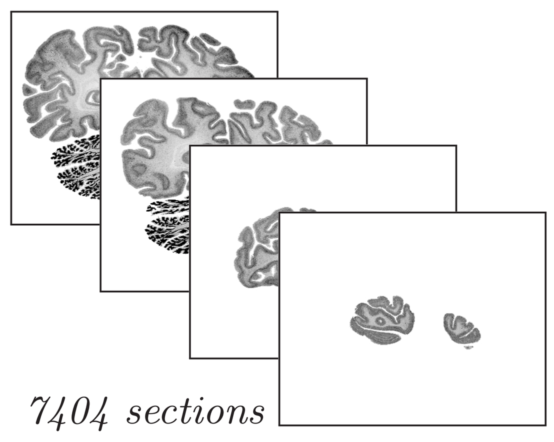
Introduction

Histological sections are a prerequisite for the analysis of the cerebral cortex of the human brain with its cytoarchitectonic areas, cortical layers, and cell columns represented at a microscopical resolution. A 3-D reconstruction of the histological dataset is, however, necessary to understand the spatial organization of these structures. The size and the complex folding pattern of the human brain introduce several challenges for its high resolution reconstruction: the size of the data (1TB), the intensity imbalance within and across sections, and the various defects introduced during histological processing, e.g., rips, tears, missing and displaced pieces, distortion (shear), stain crystallization. An algorithm is presented for the automatic repair of non-displaced artefacts such as rips, tears, and missing pieces.

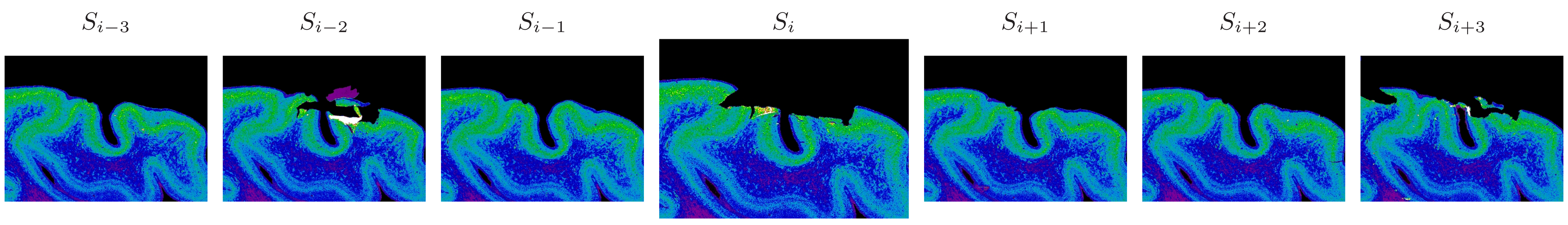
Methods

A MRI scan at $0.4 \times 0.4 \times 0.8$mm resolution of a human post-mortem brain was first obtained using a MP-RAGE sequence. The brain was then fixed in 4% formalin for several months, embedded in paraffin, and sectioned at 20μm in the coronal plane. The 7404 histological sections were mounted and stained for cell bodies. The sections were digitized using a flat bed scanner with a resolution of 2400 dpi (max $13,000 \times 11,000$ pixels, $10 \times 10\mu\text{m}^2$).



Repair Algorithm

Damaged and incomplete sections need to be repaired prior to their 3-D reconstruction at high-resolution, with the MRI serving as an undistorted spatial reference. A section S_i containing artefacts is repaired by extracting information from its immediate 3 previous and next section neighbours $S_{i\pm1,2,3}$.



The neighbouring sections S_j are first non-linearly warped to the reference section S_i; their intensities are normalized to that of S_i, after masking. A block-averaging technique is used to produce an average $\bar{S}_i$, without artefacts by removing intensity outliers, from which S_i is repaired. The process is repeated twice, using the repaired sections in the first iteration to improve the non-linear alignment and intensity normalization for the second iteration. The iterative nature of the procedure makes it self-correcting: as sections are repaired, masking and registration errors are corrected too.

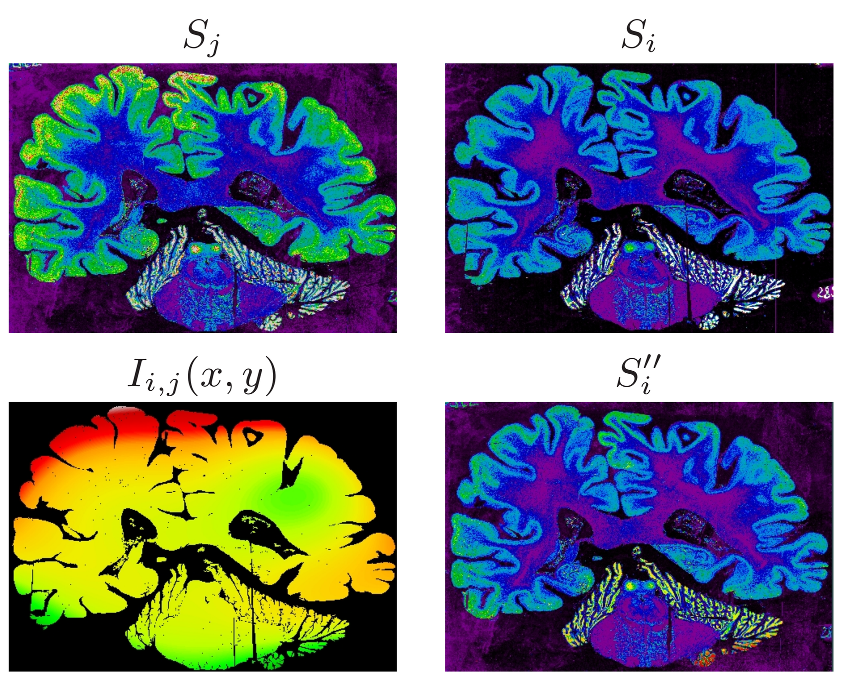
Summary:

1. *Non-linear alignment of $S_{i\pm1,2,3}$ to S_i*
2. *Intensity normalization of $S_{i\pm1,2,3}$ to S_i*
3. *Block averaging of $\bar{S}_i$*
4. *Correction of artefacts of S_i by $\bar{S}_i$*
5. *Repeat 1–4 using $\bar{S}_i$ as registration target*

Intensity Normalization

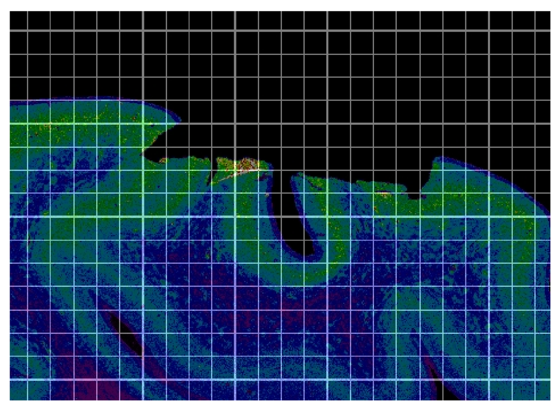
Section-to-section intensity normalization is performed relative to the reference section S_i. The normalization field $I_{i,j}(x,y)$ is obtained by matching fitted cubic b-splines between pairs of non-linearly aligned masked sections S_j to S_i:

1. *Scale and shift:* $S'_j = \alpha S_j + \beta, \ \alpha, \beta$ *const.*
2. *Field varying:* $S''_j = I_{i,j}(x,y) S'_j$



Block Averaging

The aligned normalized sections are first averaged, then intensity "outliers" (defects) are detected by means of a similarity function based on the mean and variance of the tissue intensities of the neighbouring aligned sections. The key idea in the detection of the outliers is not to look at the similarity on a pixel by pixel basis, which is too sensitive to biological variations of the tissues, but to consider the total variance in a small neighbourhood in $m \times m$ blocks. For each $m \times m$ block, compute:



$$F_s = \sum_{j=1}^{m}\sum_{k=1}^{m} \operatorname{erfc}\left(\frac{|I^s_{jk} - \mu_{jk}|}{\sqrt{2}\sigma_{jk}}\right), \quad s = i-3, ..., i+3$$

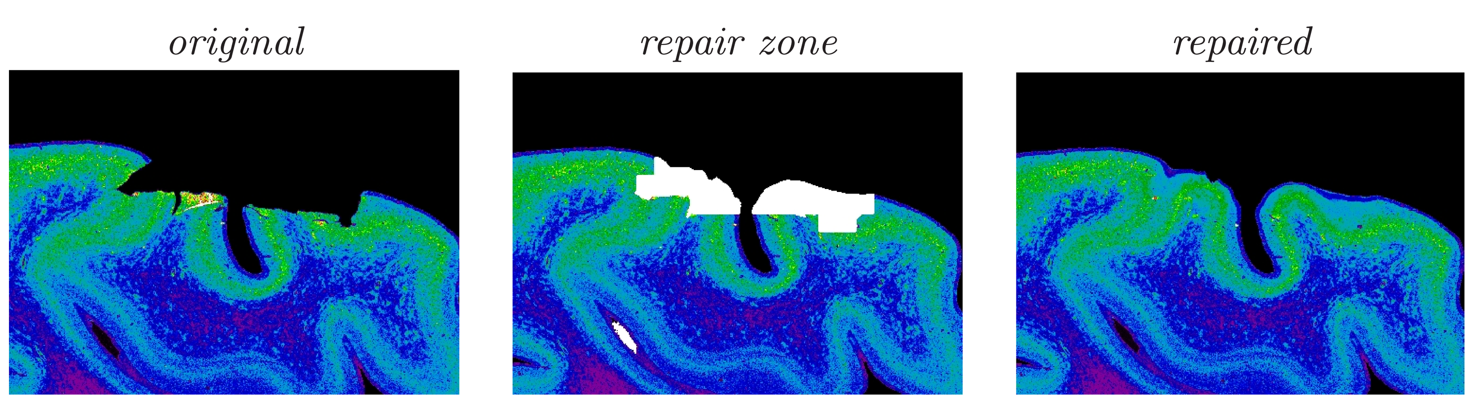
where

erfc	=	complementary error function
I^s_{jk}	=	intensity of section s at voxel j, k
μ_{jk}	=	mean of sections $i-3$,...,$i+3$ at voxel j, k
σ_{jk}	=	standard deviation of sections $i-3$,...,$i+3$ at voxel j, k

Outliers are removed on a block-by-block basis from the section average $\bar{S}_i$. The block size is $m = 25$ for the first iteration, for the gross damaged features, and $m = 10$ for the second iteration, for the finer tears.

Repair of Artefacts

The section is repaired by replacing the outlying blocks by the block-averaged section $\bar{S}_i$. Only the outlying intensities in S_i are replaced by $\bar{S}_i$.



Discussion

The automatic repair of histological artefacts is a first step towards global intensity balancing and 3-D reconstruction of large-scale datasets of human brain sections with hitherto unprecedented precision. High accuracy of the section-to-section alignment is a crucial step towards 3-D architectonic studies of the cerebral cortex: it enables the analysis in planes orthogonal to the cortical surface without any bias caused by tangential sectioning. This will open new perspectives for investigating the architecture of the cerebral cortex.

Acknowledgments

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