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Cross-Registration of Whole Slide Images from Different Modalities in AstroPath

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In this work, we established routines and methods for the registration of digital whole-slide histological imaging specifically focused on the challenges associated with combining multiplexed immunofluorescent imaging with immunohistochemical ones. There exist multiple platforms for whole-slide digital image registration, however, these are primarily limited to brightfield hematoxylin and eosin and brightfield immunohistochemical microscopy, or registering multiplex images in the same marker or narrow filter band. Other studies have demonstrated the ability to use mutual information to register immunofluorescent images with brightfield hematoxylin and eosin, however, this process requires the acquisition of a fluorescent nuclear image for each additional registered image.

We have developed and implemented a fully automated process to perform a hierarchical, parallelizable, and deformable registration of multiplexed digital whole-slide images, that is applicable even between different modalities (e.g immunofluorescent vs H&E). We generalized the calculation of mutual information as a registration criterion to an arbitrary number of dimensions, a task well-suited for multiplexed imaging. We also further leveraged the power/utility of information theory by using self information of a given immunofluorescent channel to determine its suitability for the registration process. This allows for the identification and avoidance of channels containing staining artifacts and the identification and selection of channels containing adequate information for the registration.

Challenges associated with multiplexed image registration include the potential sparseness of channels and independence between channels. Registration algorithms focused on multispectral images should be designed to accommodate these unique challenges. This effort builds on and extends existing whole-slide image registration studies by demonstrating the ability to register whole-slide multispectral immunofluorescent images with whole-slide immunohistochemical brightfield images using the fluorescent channels determined by their self information. We present our first results from successfully registering whole-slide six-channel multiplex immunofluorescent images with whole-slide brightfield immunohistochemical ones. Future work will focus on sequential slide registration and reconstruction.

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