Virtual histological stain transformations through cascaded deep neural networks

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Abstract: We present a deep learning-based framework to virtually transfer brightfield images of H&E-stained tissue slides to other types of stains using cascaded networks, providing high-quality images of special stains from existing H&E stained tissue images.

1. Introduction

Imaging and interpreting thin tissue sections is an essential part of pathological diagnosis, where various types of histochemical stains are applied to bring contrast to various desired cellular/subcellular features [1]. Histological staining requires well-trained histotechnologists to perform and monitor lengthy tissue processing and staining steps. The staining process usually involves destructive and irreversible histochemical procedures, thus making washing and restaining extremely difficult. Hematoxylin and eosin (H&E) tissue staining is the most commonly applied type of stain. However, H&E staining does not always reveal specific features of tissue, desired in some clinical cases, e.g., for examining basement membrane diseases, where additional special stains such as Periodic acid–Schiff (PAS) stain are needed for more accurate diagnosis.

Deep learning-based virtual staining methods [2-3] are developed to transform label-free images (e.g., autofluorescence) into histochemically stained images to reduce costs and standardize the staining process. An alternative approach is to learn the mapping from one stain type to another on digitalized stained tissue images. This approach, termed stain transfer, can benefit from existing histochemically stained slides, i.e., providing pathologists additional information on desired features not revealed by the original stain. However, unlike virtual staining networks, stain transfer methods face additional challenges in data acquisition and training because a tissue slide can only be stained once, which makes obtaining paired images with different stains almost impossible. Instead of utilizing datasets of unpaired images that may introduce hallucinations, de Haan et al. [4] generated virtually stained images perfectly registered across multiple stain types using pre-trained virtual staining networks using label-free autofluorescence images. However, these stain transfer models were only trained with virtually stained images and did not possess direct access to histochemically stained image data, which is a limitation. Here we report a virtual stain transfer framework using cascaded deep neural networks (termed C-DNN) to perform high-quality stain-to-stain transformations from existing histochemically stained tissue images. The superior performance of this C-DNN method was demonstrated using kidney needle-core biopsy tissue samples by virtually transferring the H&E-stained images into PAS stain [5].

2. Results and Discussions

The C-DNN consisted of two cascaded generator networks (Fig. 1a) to first virtually stain autofluorescence images to H&E stain (C-G_1) and then to perform virtual stain transfer, from H&E-stained images to PAS stain (C-G_2). The training was performed sequentially on two groups of data (Fig. 1a), each containing autofluorescence images, H&E-stained images, and PAS-stained images [5]. For group A, H&E images were histochemically stained, and PAS images were virtually stained, i.e., from virtual staining networks ($G_{AF\rightarrow PAS}$) using autofluorescence input images, while for group B, the opposite staining methods were applied: H&E images were virtually stained ($G_{AF\rightarrow H\&E}$) and PAS images were histochemically stained. Such a cascaded structure allowed the network to directly exploit histochemically stained image data to minimize the distance between the output and histochemically stained target images in both the H&E and PAS domains [5]. Upon convergence of the training phase, only the second generator C-G_2 was used for performing stain transfer of the H&E-stained images to virtual PAS stain.

Using kidney needle-core biopsy tissue sections, we blindly tested the C-DNN model and compared our output images with the standard stain transfer network [4] (Fig 1b). The testing is conducted with two test sets (groups A and B). One testing sample for each group is shown in Fig. 1b, where the output images of C-DNN agree well with the target images. Compared to the standard stain transfer network, the output images generated by C-DNN using

histochemically stained input (group A) had higher contrast in the membranes (labeled with the yellow arrows), which is a unique feature of the PAS stain. When using virtually stained images as input (group B), the color of C-DNN output closely matched the histochemically stained target [5]. In contrast, the color of the standard virtual stain transfer network output images deviated from the target, demonstrating the efficacy of the presented C-DNN framework.

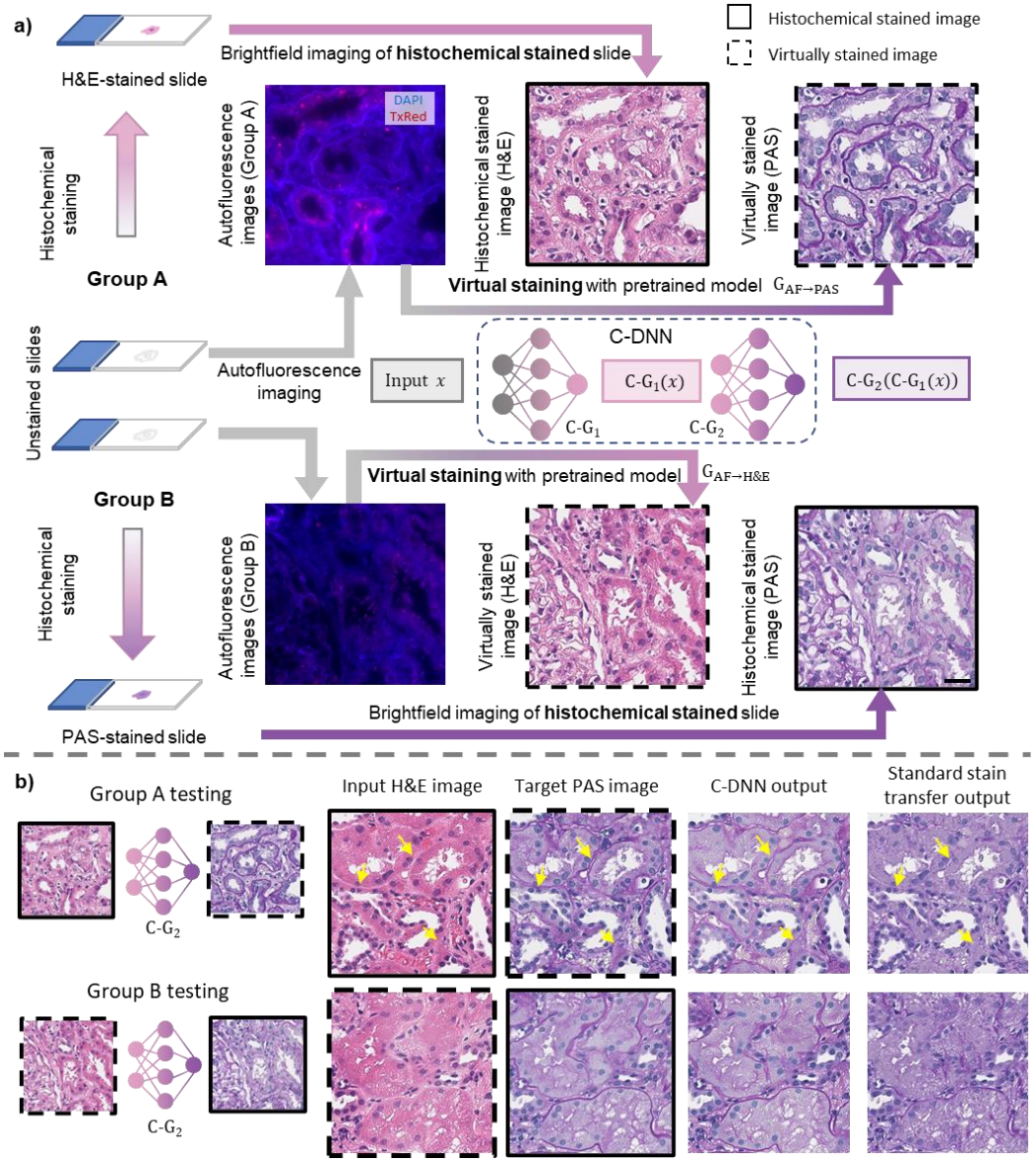

Fig. 1. C-DNN for virtual stain transfer. (a) Data preparation and training scheme of C-DNN [5]. (b) Testing results of C-DNN and comparison with a standard stain transfer network. Scale bar: 30 μm.

In summary, we developed a virtual stain transformation framework using cascaded deep neural networks [5]. This method utilizes existing images of histochemically stained tissue sections, providing pathologists with high-quality virtual images of other stains that are needed for diagnosis.

3. References

[1] H. A. Alturkistani, F. M. Tashkandi, and Z. M. Mohammedsaleh, Glob. J. Health Sci. **8**, 72 (2016).
[2] Y. Rivenson, H. Wang, Z. Wei, K. de Haan, Y. Zhang, Y. Wu, H. Günaydın, J. E. Zuckerman, T. Chong, A. E. Sisk, L. M. Westbrook, W. D. Wallace, and A. Ozcan, Nat. Biomed. Eng. **3**, 466 (2019).
[3] Y. Rivenson, K. de Haan, W.D. Wallace, and A. Ozcan, BME Frontiers (2020).
[4] K. de Haan, Y. Zhang, J. E. Zuckerman, T. Liu, A. E. Sisk, M. F. P. Diaz, K.-Y. Jen, A. Nobori, S. Liou, S. Zhang, R. Riahi, Y. Rivenson, W. D. Wallace, and A. Ozcan, Nat. Commun. **12**, 4884 (2021).
[5] X. Yang, B. Bai, Y. Zhang, Y. Li, K. de Haan, T. Liu, and A. Ozcan. arXiv preprint arXiv:2207.06578 (2022).