



P2.233: Autofluorescence- Based Virtual H&E Image Generation to Assist Intraoperative Diagnosis of Lung Adenocarcinoma: An Exploratory Study

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INTRODUCTION

Intraoperative H&E staining is time-consuming, labor-intensive, and prone to inter-institutional variability that limits both intraoperative efficiency and AI model generalizability. Autofluorescence (AF) imaging offers a rapid, label-free alternative. We developed a diffusion-based framework to translate AF images into high-fidelity virtual H&E images, aiming to provide a standardized and efficient alternative to conventional histochemical staining.

METHODS

Design:

This prospective two-center study included 215 paired intraoperative AF and H&E whole-slide images (Guangdong Provincial People's Hospital n=165 for training and internal testing; Jiangxi Cancer Hospital n=50 for external validation). AF images were acquired under the DAPI channel at 20× magnification, followed by H&E staining and re-scanning of the same sections. After strict image registration using affine transformation followed by SSIM-guided fine alignment, a conditional diffusion model was trained on 256×256 pixel patches and compared against a CycleGAN baseline. Progressive distillation compressed inference from 1000 to 8 sampling steps for rapid intraoperative use. Three independent board-certified pathologists, blinded to image origin, evaluated staining quality (nuclear, cytoplasmic, and extracellular details on a 1–4 scale), performed a visual discrimination test, and assessed five-class frozen pathology diagnostic concordance (Benign/AAH/AIS/MIA/IAC) against final paraffin diagnosis as the gold standard. SSIM and PSNR served as the primary objective image quality metrics.

DISCUSSION AND CONCLUSION

Actionable Take Away Message

- AF-based virtual H&E matches diagnostic accuracy of standard H&E while eliminating staining variability and reducing workflow complexity. Ready for integration into intraoperative pathology suites.

Future Direction for Research

- We plan to extend this framework to virtual multiplex immunofluorescence and immunohistochemistry (PD-L1, Ki-67) from a single AF scan, explore 3D virtual histology, and validate in larger cohorts to support broad clinical adoption and regulatory clearance.

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RESULTS

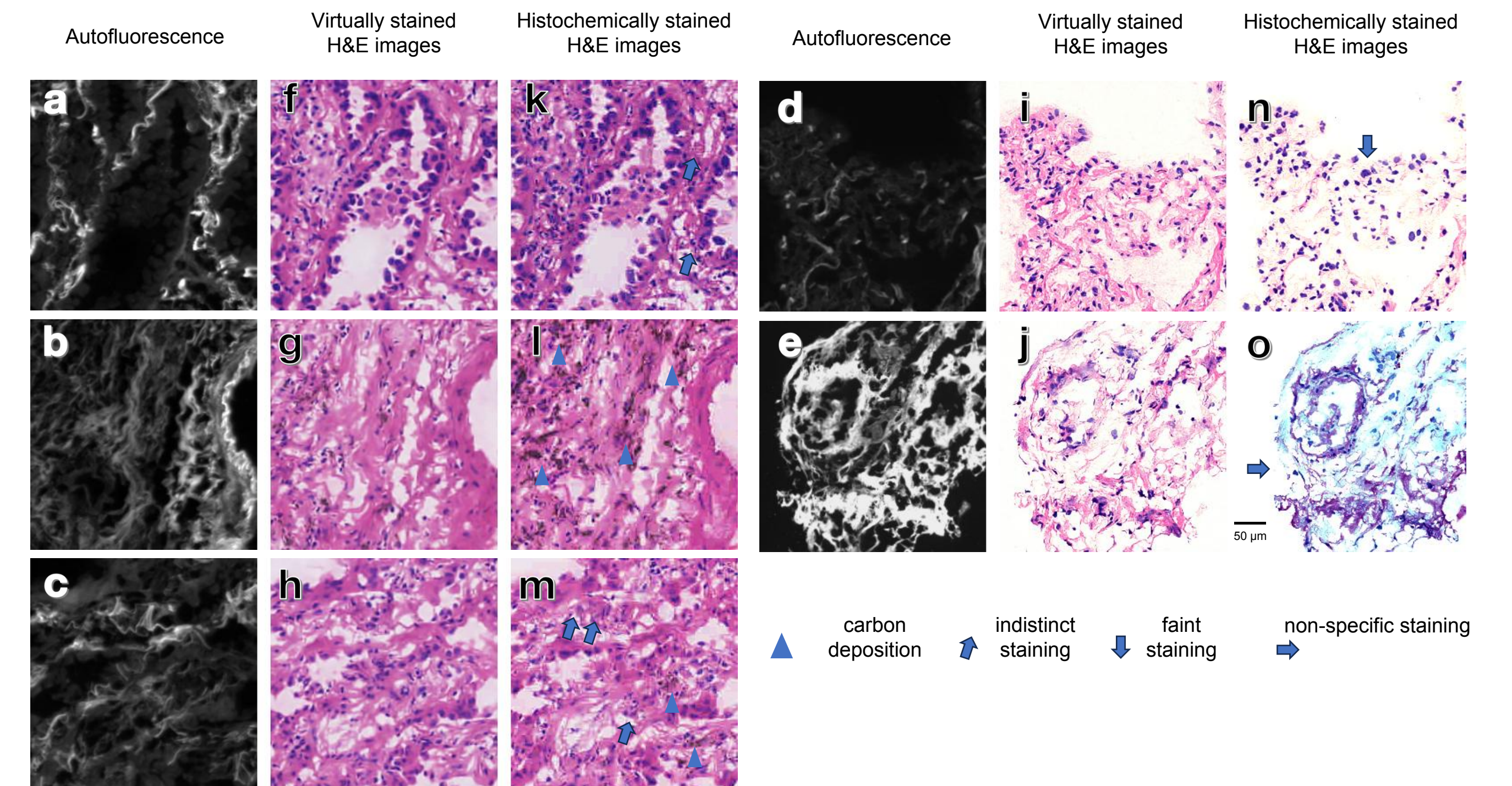


Figure1. Visual comparison between virtual staining and ground truth H&E staining. a- e: AF images, f- j: Corresponding virtually stained H&E images generated from AF images; k- o: Corresponding ground-truth H&E images.

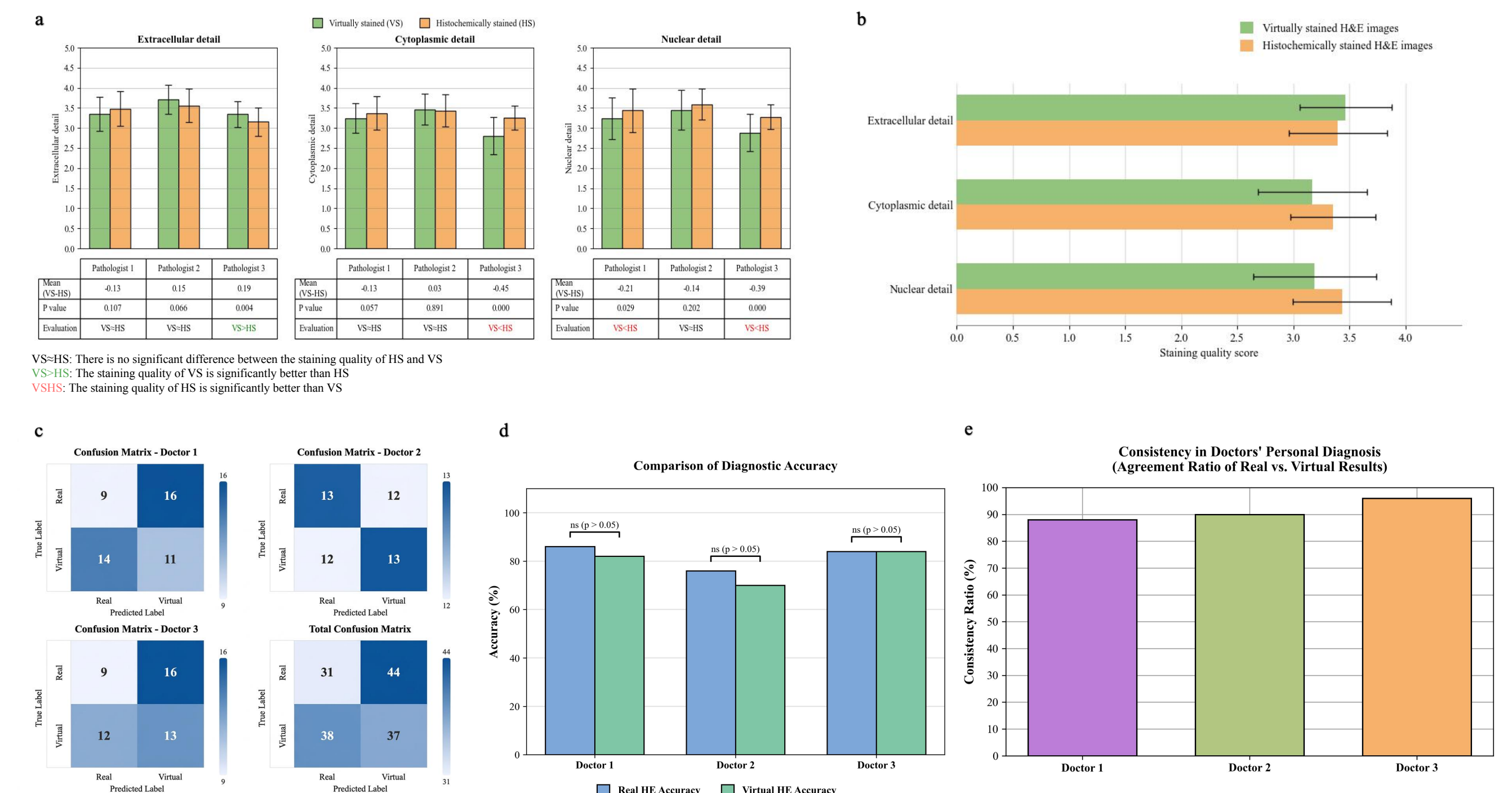


Figure 2. Subjective evaluation experiment for virtual staining. (a) Subgroup staining quality assessment from three independent pathologists, comparing virtually stained (VS) and histochemically stained (HS) H&E images across extracellular, cytoplasmic, and nuclear details, with corresponding P values and evaluation conclusions. (b) Overall mean staining quality scores, demonstrating highly comparable results between VS and HS with overlapping confidence intervals. (c) Confusion matrices of pathological diagnoses for each pathologist and the over all cohort, comparing diagnostic results based on VS versus HS images. (d) Comparison of diagnostic accuracy between VS and HS images, with no significant difference observed across all three evaluators. (e) Intra- observer diagnostic consistency, showing the agreement ratio of each pathologist's diagnose based on VS versus HS images.