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3 | **Confocal Fluorescence Microscopy Platform Suitable for Rapid Evaluation of Small Fragments of Tissue in Surgical Pathology Practice.**

ABSTRACT

CONTEXT.?:Rapid advances in the fields of biophotonics, computer science, and instrumentation have allowed for high-resolution imaging of biologic tissues. OBJECTIVE.?:To evaluate the quality of images from an optimized confocal fluorescence microscopy (CFM) platform for rapid evaluation of small fragments of tissue, compared with hematoxylin-eosin staining. DESIGN.?:Tissue fragments (up to 1.0×0.3 cm) were stained with 0.6 mM acridine orange for 60 seconds and imaged using a CFM platform at 488-nm and 785-nm wavelength. The imaged tissues were then fixed in formalin and processed to generate hematoxylin-eosin-stained tissue sections. The quality of CFM images was scored on a scale of 0 to 3 on the basis of the percentage of the CFM images with recognizable tissue architecture (0, 0%; 1, <20%; 2, 20%-50%; 3, >50%). The diagnoses made using CFM images were compared with those made using histopathologic analysis of the hematoxylin-eosin-stained tissue sections. RESULTS.?:We imaged 118 tissue fragments obtained from 40 breast, 23 lung, 39 kidney, and 16 liver surgical excision specimens. We acquired CFM images in 2 to 3 minutes; 95.8% (113 of 118) of images showed a quality score of 3, and 4.2% (5 of 118) had a score of 2. We achieved a sensitivity of 95.5%, specificity of 97.3%, positive predictive value of 95.5%, and negative predictive value of 97.3%. CONCLUSIONS.?:Our results demonstrate the suitability of the CFM platform for rapid and accurate evaluation of small tissue fragments in surgical pathology practice.