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Confocal mosaicing microscopy in Mohs skin excisions: feasibility of rapid surgical pathology

Gareau D, Li Y, Huang B, Eastman Z, Nehal K, Rajadhyaksha M.; *Journal of Biomedical Optics* 13 5, 054001 September/October 2008

ABSTRACT

Mosaicing of confocal images enables observation of nuclear morphology in large areas of tissue. An application of interest is rapid detection of basal cell carcinomas (BCCs) in skin excisions during Mohs surgery. A mosaic is currently created in less than 9 min, whereas preparing frozen histology requires 20 to 45 min for an excision. In reflectance mosaics, using acetic acid as a contrast agent to brighten nuclei, large and densely nucleated BCC tumors were detectable in fields of view of 12 x 12 mm (which is equivalent to a 2x-magnified view as required by Mohs surgeons). However, small and sparsely nucleated tumors remained undetectable. Their diminutive size within the large field of view resulted in weak light backscatter and contrast relative to the bright surrounding normal dermis. In fluorescence, a nuclear-specific contrast agent may be used and light emission collected specifically from nuclei but almost none from the dermis. Acridine orange of concentration 1 mM stains nuclei in 20 s with high specificity and strongly enhances nuclear-to-dermis contrast of BCCs. Comparison of fluorescence mosaics to histology shows that both large and small tumors are detectable. The results demonstrate the feasibility of confocal mosaicing microscopy toward rapid surgical pathology to potentially expedite and guide surgery.