Evaluation of digital staining for ex vivo confocal laser scanning microscopy.


ABSTRACT
BACKGROUND: Ex vivo confocal laser scanning microscopy (CLSM) is a novel diagnostic tool for the fast examination of native tissue. However, CLSM produces black/white/green images, depending on the refraction indices of the tissue structures, complemented by nuclear fluorescence staining, which the vast majority of Mohs surgeons and dermatopathologists are not trained to interpret. Digital staining is applicable to ex vivo CLSM investigations to simulate the images of conventional slides stained with haematoxylin and eosin (H&E). OBJECTIVES: The aim of our study was to evaluate in detail the appearance of human skin structures using digitally stained ex vivo CLSM images and compare the results to that of conventional H&E slides of the same specimen.

METHODS: After providing informed consent, 26 patients donated their Burow's triangles (healthy skin) that resulted from plastic reconstruction after the R0 excision of skin tumours. After being investigated by ex vivo CLSM, including automated digital staining (VivaScope 2500M-4G, MAVIG GmbH), the specimens were fixed in formalin, embedded in paraffin and stained with H&E.

RESULTS: Almost all skin structures in the digitally stained ex vivo CLSM images morphologically resembled the structures in the histopathological images acquired from H&E slides. Due to the high refraction index of melanin, the hair shafts appeared bright pink, and the melanocytes and melanophages were poorly imaged, resulting in a strong pink appearance that vastly differed from the appearance of conventional H&E-stained histopathology.

CONCLUSIONS: Digital staining of ex vivo CLSM images is an easy and highly useful tool to facilitate the interpretation of black-field images generated by confocal laser scanning microscopy for dermatopathologists and Mohs surgeons who are familiar with H&E staining. Unlike the pigmented structures, the cutaneous and subcutaneous structures had excellent visualization with only minimal differences from their appearance on H&E slides. © 2019 The Authors. Journal of the European Academy of Dermatology and Venereology published by John Wiley & Sons Ltd on behalf of European Academy of Dermatology and Venereology. PMID: 31732988 DOI: 10.1111/jdv.16085