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2 | New-generation diagnostics in inflammatory skin diseases: Immunofluorescence and histopathological assessment using ex vivo confocal laser scanning microscopy in cutaneous lupus erythematosus

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ABSTRACT

Ex vivo confocal laser scanning microscopy (CLSM) offers real-time examination of excised tissue in reflectance, fluorescence and digital haematoxylin-eosin (H&E)-like staining modes enabling application of fluorescent-labelled antibodies. We aimed to assess the diagnostic performance of ex vivo CLSM in identifying histopathological features and lupus band test in cutaneous lupus erythematosus (CLE) with comparison to conventional histopathology and direct immunofluorescence (DIF). A total of 72 sections of 18 CLE patients were stained with acridine orange (AO), anti-IgG, anti-IgM and anti-IgA; 21 control samples were stained with AO. Subsequently, ex vivo CLSM examination of all samples was performed in reflectance, fluorescence and digital H&E-like staining modes. Superficial and deep perivascular inflammatory infiltration (94.4%), interface dermatitis (88.9%), spongiosis (83.3%) and vacuolar degeneration (77.7%) were the most common features detected with ex vivo CLSM. Kappa test revealed a level of agreement ranging within "perfect" to "good" between ex vivo CLSM and conventional histopathology. ROC analysis showed that the combination of perivascular infiltration, interface dermatitis and spongiosis detected by ex vivo CLSM has the potential to distinguish between CLE and controls. Basement membrane immunoreactivity with IgG, IgM and IgA was identified in 88.8% (n = 15), 55.5% (n = 10) and 55.5% (n = 10) of the CLE samples using ex vivo CLSM, respectively, whereas DIF showed IgG, IgM and IgA positivity in 94.4% (n = 17), 100% (n = 18) and 88.9% (n = 16) of patients, respectively. In conclusion, ex vivo CLSM enables simultaneous histopathological and immunofluorescence examination in CLE showing a high agreement with conventional histopathology, albeit with a lower performance than conventional DIF. Keywords: diagnostics; direct immunofluorescence microscopy; ex vivo confocal laser scanning microscopy; fluorescence; lupus erythematosus.
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