

***Diagnostic Segregation of Human Brain Tumours Using Fourier-transform Infrared and/or Raman Spectroscopy Coupled with Discriminant Analysis***

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**Background:** During surgery for removal of a brain tumour, it is very important to remove the diseased tissue in its entirety to prevent recurrence whilst minimizing damage to normal adjacent tissue. Fourier-transform infrared (FTIR) and Raman spectroscopy has shown promise as a non-invasive approach with diagnostic potential in cancer detection.

**Aim:** To determine whether IR and/or Raman spectroscopy combined with multivariate analysis could be applied to discriminate between normal and various grades of brain tumours.

**Methods:** With appropriate ethical approval, 52 paraffin-embedded (FFPE) tissue blocks were obtained. Thin cut sections were mounted on low-E IR reflective slides or on glass slides (H&E-stained tissue) and dewaxed. IR spectra were acquired using a Bruker Tensor 27 FTIR spectrometer or Raman spectra with an InVia Renishaw Raman spectrometer. Towards analyzing the data, principal component analysis (PCA) and/or linear discriminant analysis (LDA) was used. Immunohistochemical analysis was carried out for IDH1 and P53 on glioma samples in order to correlate these conventional markers of disease with novel spectral biomarkers.

**Results:** Clear segregation was observed between normal and various grades of brain tumours. In derived PCA-LDA scores plots, marked within-category variation (*i.e.*, heterogeneity) was noted; even so, excellent discrimination between different grades of glioma was also observed (*i.e.*, towards between-category discriminating biomarkers). Spectral biomarkers appeared to be more robust towards identifying aberrant tissue than immunohistochemical markers. Using linear discriminant classifier, following diagnostic accuracy of 96 % diagnostic for normal brain, 93 % for glioblastoma and 93.3 % for astrocytomas were obtained for IR spectra.

**Conclusions:** This exploratory study indicates that IR and/or Raman spectroscopy coupled with multivariate analysis may provide a novel diagnostic approach especially towards identifying tumour margins has the potential to be used to differentiate brain tumours.