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## **Determination of meningioma brain tissue grades using Raman hyperspectral imaging**

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Raman spectroscopy is a powerful tool used to analyse biological materials, where spectral biomarkers such as proteins (1500-1700  $\text{cm}^{-1}$ ), carbohydrates (470-1200  $\text{cm}^{-1}$ ) and phosphate groups of DNA (980, 1080-1240  $\text{cm}^{-1}$ ) can be detected. A major advantage is that it is reagent-free and unaffected by water interference, which is ideal for biological applications. Raman hyperspectral imaging combines the chemical sensitivity of this spectrochemical technique with spatially distributed information. Herein, Raman microspectroscopy imaging ( $50 \times 50 \mu\text{m}$  tissue area,  $50\times$  magnification, 50% laser power, 0.1 ms exposure time, 780-1858  $\text{cm}^{-1}$  spectral range) was used to investigate 79 brain tissue samples (sourced from the Brain Tumour North West) in order to differentiate meningioma Grade I ( $n=55$ ) *versus* Grade II ( $n=24$ ). Meningioma is the commonest type of brain tumour with the majority of them being benign tumours (Grade I) whilst a few are aggressive or malignant (Grade II). Grade II tumours have a poor prognosis by their nature, hence new sensitive diagnostic tools are essential. Using partial least squares discriminant analysis (PLS-DA) with 20 LVs (99% explained variance), we were able to differentiate Grade I *versus* Grade II meningioma with a classification accuracy of 99% in an internal validation set of 23 samples (16 Grade I, seven Grade II); this was with an accuracy of 85% for an external test set of similar size. These findings highlight the potential of Raman hyperspectral imaging for differentiation of meningioma tumours.