Full Title: Spatial transcriptomics of compartmentalised inflammation in Multiple Sclerosis

Short title: Spatial transcriptomics of MS inflammation

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Abstract:
Multiple Sclerosis (MS) is a chronic inflammatory demyelinating disease of the central nervous system (CNS). In the later and progressive stages, inflammation can become compartmentalised in the connective tissue spaces of the vasculature and leptomeninges, occurring behind an intact blood-brain-barrier. The relative grade of meningeal inflammation and the presence of tertiary-lymphoid structures (TLS) is associated with a more severe disease course and patients entering the progressive phase sooner. The immune cell composition of TLS in MS is unclear and it is not known whether the cellular profiles of TLS are distinct in different CNS compartments that harbour inflammatory cells. This study aims to characterise the molecular profiles of TLS and other CNS inflammatory areas.

Using formalin-fixed paraffin embedded (FFPE) post-mortem tissue from the Dame Ingrid Allen Tissue Collection, Queen's University Belfast (QUB) in-depth molecular profiling of TLS and other CNS inflammatory compartments was undertaken. Nanostring digital spatial profiling whole transcriptome analysis (WTA) was performed on TLS, meningeal and perivascular regions of 2 MS cases (Case D28 and Case D192). Using the Nanostring dataset, we identified the highest and most differentially expressed genes in TLS and other CNS inflammatory compartments. A subset of these RNA targets, were selected for validation in a single TLS+ MS case with the RNAscope HiPlex assay. This assay allows detection of up to 12 targets simultaneously at the cellular level. Preliminary findings confirm detection of immune-related genes associated with the TLS transcriptomic profile. Further, it indicates a possible association between cellular phenotypes occurring in TLS and in GM perivasculature adjacent, but not distal, to the TLS. These findings demonstrate that long-archived human FFPE CNS tissue can be used for spatial transcriptomic profiling and this unbiased approach can identify novel molecular signatures in MS brain tissue.

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Disclosure of potential conflicts of interest:

There are no potential conflicts of interest to declare on behalf of the authors.