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Development of stabilised aligned CNS co-culture technology to model the behaviour of a range of neural cell types

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The overall aim is to develop robust culture models that recreate the 3D environment of the CNS, thereby allowing neurons and glial cells *in vitro* to behave similarly to their counterparts *in vivo*. A simple, consistent and physiologically relevant model system, which uses a multiwell plate format and can potentially be used at a scale suitable for commercial R&D, has been developed. The model uses an engineered neural tissue which is prepared by a process of initial glial cell self-alignment within a tethered 3D collagen hydrogel and subsequent stabilisation of the gel. Stabilisation is achieved using RAFT technology (www.raft3dcellculture.com), which entails partial removal of interstitial fluid thereby increasing matrix and cell density. A CNS co-culture system suitable for widespread adoption will require various combinations of cells to suit specific neuroscience research requirements. Both primary neuronal and glial cell types and relevant cell lines were used to establish engineered neural tissues which were then assessed using a range of measures including neural cell survival, morphology, proliferation, differentiation and sensitivity to stabilisation. In a bid to determine the complexity of the CNS model, neuron-glial interactions, markers for myelination and reactive gliosis were also investigated. The highly organised nature of the cells and extracellular matrix in this anisotropic system facilitates quantitative analysis of cellular features including neurite length and myelination. Initial studies reveal the new model system can be assembled quickly and reliably using various primary neural cells or cell lines, the approach can be scaled down to facilitate increased throughput, and the co-cultures exhibit characteristic behaviours that mimic *in vivo* scenarios.