



Title	Multi-channel Collagen Gel (MCCG) as a Biomaterial Scaffold for Tissue Engineering [an abstract of dissertation and a summary of dissertation review]
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Abstract of Doctoral Dissertation

Degree requested Doctor of Life Science

Applicant's name KOH Isabel Siew Yin

Title of Doctoral Dissertation

Multi-channel Collagen Gel (MCCG) as a Biomaterial Scaffold for Tissue Engineering
(組織工学のための生体材料足場としてのマルチチャンネルコラーゲンゲル (MCCG))

Multi-channel collagen gel (MCCG) is prepared simply by dialyzing a phosphate buffer solution (gelation PBS) against collagen solution, and the phase separation of collagen solution yields a porous anisotropic hydrogel. The anisotropic property of the MCCG is supplied by the arrangement of collagen fibres parallel to the circumference of the channel lumen, and perpendicular to the axis of the channel. In this thesis, the aim is to explore the potential of the MCCG as a biomaterial for neural tissue engineering, and as a model for studying phase separation processes in biology.

The MCCG presents a porous hydrogel scaffold onto which cells may be seeded, or encapsulated within, to achieve a 3D culture system. In chapter 3, these seeding methods were tested using single cells and cell spheroids, and it was observed that kidney epithelial cells adhered to the surface of the channels when seeded but formed cysts when encapsulated within the collagen matrix. Furthermore, PC12 cells encapsulated in normal collagen gel (COL) extended more neurites and grew in larger aggregates compared to those in MCCG. These findings suggest that even though various methods have been proposed for 3D culture systems, they may still not be comparable.

The alignment of collagen fibres and the restriction of available collagen-rich regions by the presence of the channels in MCCG make it a prospect for neural tissue engineering, given that the guidance of neurite growth is an important criteria for neural guidance conduits. In chapter 4, the alignment of collagen fibres in MCCG was investigated, and it was shown that the extension of PC12 cell neurites were significantly guided in MCCG compared to COL. It was also suggested that the MCCGs prepared with different ionic concentrations in this study were all within the threshold fibre alignment range required for contact guidance of neurites.

The two phase separation processes of nucleation and growth (NG) and spinodal decomposition (SD) are often investigated as separate studies and not compared to one another. As the two hydrogels used in this study, COL and MCCG, are formed by NG and SD phase separation, respectively, this provides an opportunity to study both processes simultaneously. In Chapter 5, microspheres of 6 μm and 0.5 μm were used as models of cells and organelles, respectively, to investigate the movement of particles during the phase separation process. It was observed that in COL, the movement of the particles were largely downwards, whereas in MCCG, the particles were largely moved upwards and sideways away from the forming channel. The particles of different sizes and density did not appear to have an effect on the rate of gelation. The velocity of 6 μm microspheres were not affected by the density of particles, but with 0.5 μm microspheres, the particle velocity decreased with increasing particle density.

Overall, the simple experiments conducted in this thesis point to the potential applications of MCCG as a scaffold in 3D culture systems and tissue engineering, as well as a tool for studying phase separation processes with relevance to the extracellular matrix (ECM), of which collagen is a major component. Nevertheless, further comprehensive studies are required to put these into full use.