

## Investigation of Spider Egg Case Silks for Cartilage Tissue Engineering

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**INTRODUCTION:** Spider silks are some of the toughest natural materials, and they have shown potential for use as scaffolds for cartilage tissue engineering. Our group previously reported the use of silk harvested from *Nephila edulis* egg cases, and processing methods to improve their efficacy as tissue engineering scaffolds. Here, the distinct regions of the egg case are investigated.

**METHODS:** The morphological features of the silk fibres taken from the inner and outer portions of egg cases were observed by scanning electron microscopy (SEM). Scaffolds for cartilage tissue engineering were formed from these silk samples and sterilized by autoclaving. Freshly harvested mature chondrocytes obtained from bovine metacarpophalangeal joints were seeded onto the scaffolds using published methods<sup>1</sup>. After 7 days, samples were observed by SEM. Further cell-scaffold constructs were cultured for 40 days, to engineer hyaline-like cartilage. After 40 days, constructs were analysed biochemically<sup>2</sup> and histologically to determine glycosaminoglycan (GAG) content and distribution. Collagen type I and type II were detected immunohistochemically. Poly(glycolic acid) scaffolds were used as comparative materials.

**RESULTS:** SEM showed scaffolds had similar fibre morphology. Cells had attached to all scaffolds and formed confluent sheets after 7 days. After 40 days in culture, extracellular matrix proteins were detected in all constructs.

*Fig 1 Morphology of tissue engineering scaffolds. A: Nephila edulis outer egg case silk, B: N. edulis inner egg case silk, C: Poly(glycolic acid) non-woven felt*

**DISCUSSION & CONCLUSIONS:** We have demonstrated that different regions of *Nephila edulis* egg case silks support the attachment, proliferation and re-differentiation of bovine articular chondrocytes *in vitro*. We found that all scaffolds supported the formation of a hyaline-like cartilage, and therefore conclude that spider silks have potential as scaffolds for cartilage tissue engineering.

**REFERENCES:** <sup>1</sup> in *Biopolymer Methods in Tissue Engineering*, AP Hollander & PV Hatton (Eds), Humana Press (2004). <sup>2</sup> RW Farndale, DJ Buttle, AJ Barrett, *Improved quantitation of glycosaminoglycans by use of dimethylmethylene blue*. *Biochemica et Biophysica Acta*, **883** (1986): 173-177.

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