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Plastic compression of collagen: development and assessment of a new biomaterial in nerve repair

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Plastic Compression of Collagen: Development and Assessment of A New Biomaterial in Nerve RepairS.Ashraff¹ R.A.Brown¹ J.B.Phillips²¹*Tissue Repair and Engineering Centre, Institute of Orthopaedics, UCL, London HA7 4LP, U.K.*²*Department of Biological Sciences, The Open University, Milton Keynes, U.K.*

INTRODUCTION: Nerve reconstruction is a surgical challenge. Current neural conduits provide sub-optimal clinical results. We have developed a device from a composite material comprising a fibronectin core and a collagen outer layer, using collagen made by a new technique called plastic compression. This collagen element has the strength to improve the mechanical properties of nerve repair devices which is an important design consideration.¹ Preliminary studies have been performed to assess the suitability of the new collagen material for use in nerve repair conduits.

METHODS: A 6ml gel was made from type I rat tail collagen (First Link), substituted with 10xMEM and 10 % EBSS. This was neutralized with NaOH and left to set for 30 minutes in a rectangular metal mould. The gel was removed from the mould and compressed, leaving a flat sheet of collagen. This material can be rolled to create an implantable conduit. In order to investigate the ability of the compressed collagen to support neuronal growth, flat sheets were used in this experiment.

To assess stability, a 6ml flat sheet was divided and incubated at 4°C and 37°C in EBSS for 2 weeks. At the end of the incubation period, the material was degraded with collagenase to quantify the protein remaining in the material. The rate of protein release from the material into the solution was assessed using a BCA assay.

Dissociated dorsal root ganglia (DRG) from 250g Sprague Dawley rats were seeded upon the surface of the collagen material and maintained in culture for 4 days at 37°C, 5% CO₂. Controls were equivalent cultures grown on shear aggregated fibronectin mats, an established substrate for neuronal growth.²

The constructs were fixed in 4% paraformaldehyde, then mouse anti-β₃ tubulin, (1:300 for 1h, Sigma) was used to detect neurones. The secondary antibody was TRITC

conjugated anti-mouse IgG, (1:100, 45 min). Labelled cells growing on the materials were visualized using fluorescent microscopy.

RESULTS:

Graph to show stability of compressed collagen

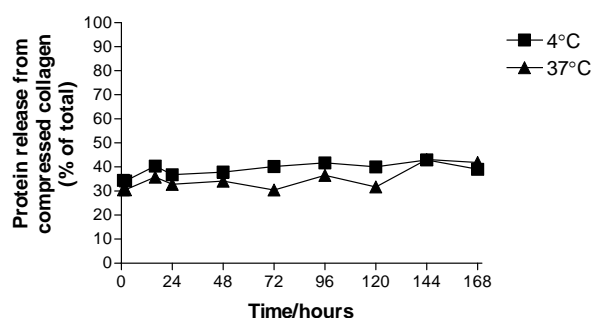


Fig 1: Protein loss from compressed collagen sheets incubated for 7 days in EBSS.

After incubation for 1h at 4 and 37°C, protein loss from the material was 34 and 31% respectively. There was minimal further loss in the remaining 7 days.

Culture studies showed that both the compressed collagen and fibronectin control supported neurite outgrowth from the DRGs .

DISCUSSION: This novel biomaterial shows potential for use in a neural conduit. It is sufficiently stable to be incorporated into composite devices and supports neural growth.

REFERENCES: ¹J.B. Phillips, X. Smit, et al.(2004) *J Physiol* **557(3)** 879-887 ²I.H. Whitworth, R.A. Brown, et al.(1995) *J Hand Surg* **21B** 514-522.

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