INTRODUCTION

Peripheral Nerve Regeneration with Nerve Guidance Conduit (NGC)

Scientific Premise: Current man-made NGCs have not succeeded in matching the regenerative capability of autograft nerves, frequently displaying very poor and incomplete functional peripheral nerve repair. It is thus in urgent need to develop NGCs having superior peripheral nerve regeneration capability that can be matched with that of autograft nerves.

Innovation: Exploiting potential intrinsic capacity of graphene to trigger/support neural regeneration.

OBJECTIVE

Graphene Stimulation of Schwann Cell Adhesion, Alignment, and Functional Activity

Rationale: Schwann cell ingrowth and alignment within NGC is a crucial guidance step for later axonal growth and myelination for PNS regeneration.

Specific Aim: Examine the effects of graphene substrate culture on the growth, alignment, and differentiation of Schwann cells.

Hypothesis: Schwann cell functional activity may be enhanced on graphene via FAK and/or p38 MAPK, each constituting key focal adhesion and environmental stress signaling.

METHODS

Schwann Cell Model: Rat S16 (ATCC CRL-2941).

Graphene Monolayer Film: Chemical vapor deposition (CVD) followed by wet transfer on basal glass substrate.

Cell Morphology: Cell spreading area, aspect ratio, and orientation angle. Fluorescent images are analyzed by ImageJ. A total of n = 74 and n = 72 cells analyzed for glass and graphene, respectively.

Cell Proliferation: Alamar blue proliferation assay.

Schwann Cell Phenotype: Fluorescent imaging of Schwann cell marker (myelin basic protein, MBP) and others (GFAP, GAP43, Dhh, etc.).

RESULTS

Graphene Substrate Fabrication and Characterization

Graphene Culture Stimulation of Schwann Cell Alignment

Optical microscopy

Fluorescence microscopy: (Red) Actin (Blue) DAPI

RESULTS (CONT.)

CONCLUSIONS/FUTURE DIRECTION

Conclusions: Schwann cells on graphene had significant alignment behavior (as shown by aspect ratio and orientation angle) but decreased cell area and proliferation relative to those on glass. This suggests the potential benefit of graphene to induce required Schwann cell ingrowth and alignment within NGC.

Future Direction: (1) Mechanism study on graphene control of Schwann cells; (2) Schwann cell-axonal co-culture model to test axonal guidance; (3) Graphene NGCs for testing peripheral nerve regeneration with rat sciatic nerve injury model.

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