Controlled Brain Derived Neurotrophic Factor Release from Layer-by-Layer Coated Hydrogel Scaffolds for Nerve Repair

Daniel A Lynam,1 Dena Shahriari,1 Kayla Felger,1 Kobi Koffler,2 Mark H Tuszynski,2 Jeffrey S Sakamoto,1 1Michigan State University, East Lansing, MI, USA and 2University of California-San Diego, La Jolla, CA, USA

Introduction

Paralysis, or the functional loss of voluntary movement resulting from damage to nerve or muscle tissue, is a devastating condition with no treatment available. Recently, agarose hydrogel nerve guidance scaffolds (NGS) have been fabricated [1] and functionalized with brain derived neurotrophic factor (BDNF), demonstrating substantial axon regeneration in vivo [2]. However, these reports employed a cellular BDNF delivery strategy to promote regeneration. The following work focuses on functionalizing agarose hydrogel NGS to release BDNF by utilizing an acellular mechanism, hydrogen-bonded layer-by-layer (LBL). To this end, we report efforts to tailor the LBL process for optimized release of BDNF from templated hydrogel scaffolds. Replacing hydrochloric with acetic acid for LBL assembly will improve BDNF stability, but also affect cumulative dosages and release rates. From this work, incorporation and controlled release of nerve growth factors from LBL-coated NGSs will promote and sustain axon growth by an acellular method.

Materials and Methods

Nerve growth proteins can be incorporated into the NGS via a hydrogen-bonded, dual polymer LBL system under acidic conditions [3]. By depositing LBL on the highly porous hydrogel, the gradual release of BDNF or lysozyme, a suitable analog for BDNF, is achieved by release of polymer layers under physiological pH. This system consists of depositing alternating layers of acetic acid-adjusted polyacrylic acid, polyethylene glycol, and BDNF within the NGS hydrogel framework and measuring protein release every 24-hours.

Results

Release of lysozyme, a BDNF analog, from LBL-coated agarose hydrogels has been shown to reach milligram/milliliter concentrations over the course of one month [3]. Likewise, we have shown that augmenting the mesopore volume within the hydrogel through the addition of sucrose during gelation results in a substantial increase in release dose of lysozyme, surpassing 2500µg/mL cumulative release in some samples [4]. However, this work utilized hydrochloric acid to adjust LBL assembly conditions. Modifications to the LBL assembly are needed to provide BDNF stability. Incorporating acetic acid into the LBL process has shown controlled release of lysozyme in clinically relevant dosages on the week timescale. Acetic acid not only affects LBL kinetics, but also stabilizes proteins; an important aspect for maintaining bioactivity. This work has thus allowed for release of BDNF from LBL-coated agarose NGS (Fig. 1).

Discussion and Conclusions

This work demonstrates the controlled release of BDNF from LBL-coated agarose NGS over a 12-day period. Although release concentrations are below the clinical relevance of 20ng/mL per day, localized concentrations within the agarose NGS may be higher. Continued work will explore ways of increasing BDNF release concentrations. This work shows that the LBL method is a viable technique for prolonged, controlled delivery of growth proteins for use in spinal cord repair therapies.

References List


Acknowledgments

This work acknowledges funding from NIH 1R01EB014986-01.

Disclosures

The authors have nothing to disclose.