

MODIFIED CHONDROITINASE ABC FOR TREATMENT OF SPINAL CORD INJURY

Dr John Rogers and Dr Liz Muir have modified the bacterial chondroitinase ABC gene so that mammalian cells are able to synthesise and secrete active chondroitinase ABC to degrade CSPGs which inhibit axon regrowth. The optimised gene sequence with preferred mammalian codons gives efficient expression from:

- transfection in COS, Neu7, SCTM41 and primary astrocyte cells
- lentiviral transformation of HEK293T, Neu7, SCTM41, primary astrocyte cells, primary Schwann cells and fibroblasts

In vivo studies using lentiviral vectors in a rat model of wound injury have shown that active enzyme is secreted at the injury site.

Potential Uses

Treatment of spinal cord injury (SCI) and other nervous system damage by:

- Gene Therapy - controlled production of chondroitinase ABC in mammalian neurons and/or glia to deliver enzyme directly to injury site
- Cellular Therapy - generation of Schwann cells that secrete chondroitinase ABC for in-vivo bridge grafts at injury site

For further information please contact:

Dr Emma Barker

✉ emma.barker@enterprise.cam.ac.uk

☎ +44 (0)1223 760339

Cambridge Enterprise Limited, University of Cambridge
10 Trumpington Street, Cambridge CB2 1QA UK
www.enterprise.cam.ac.uk

Case Ref: Rog-1024-03

Background

About 250,000 people in the US live with spinal cord injury and there are ~12,000 new spinal cord injuries every year. These injuries cause a glial scar to form in the spinal cord and the inhibitory molecules present, such as chondroitin sulphate proteoglycans (CSPGs) inhibit axon regrowth.

Chondroitinase ABC is an enzyme made naturally by some bacteria which cleaves the glycosaminoglycan (GAG) chains on CSPGs which are responsible for most of the inhibition of axon regrowth.

This enzyme has shown considerable promise in a rat model of SCI after repeated injections of the enzyme at the site of injury and promotes regeneration of injured axons and functional recovery following spinal cord injury. However, repeated injections increase the risk of further trauma and infection, so development of suitable administration at the site of injury is required.

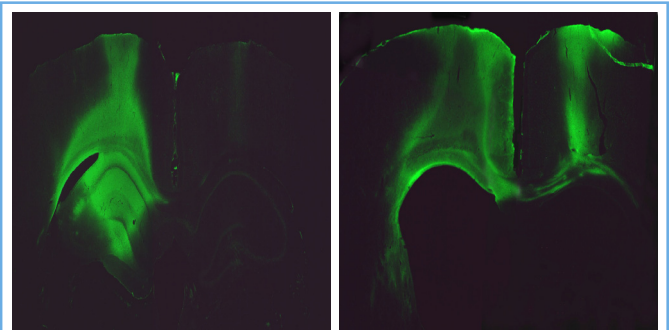
Technology

The bacterial gene encoding chondroitinase ABC cannot be adequately expressed in mammalian cells, because these cells, unlike bacteria, glycosylate the protein. Dr John Rogers and Dr Liz Muir at the University of Cambridge have modified the bacterial chondroitinase ABC gene so that mammalian cells are able to synthesise and secrete active enzyme.

The modified gene has mutations that eliminate key/specific glycosylation sites and a signal sequence derived from another mammalian gene. It has been re-synthesised with preferred mammalian codons and gives efficient expression from:

- transfection in COS, Neu7, SCTM41 and primary astrocyte cells
- lentiviral transformation in HEK293T, Neu7, SCTM41, primary astrocyte cells, primary Schwann cells and fibroblasts

In vivo studies using lentiviral vectors in a rat model of wound injury have shown that active enzyme is secreted at the injury site.



Sections of rat brain at 2 and 4 weeks - the 1B5 antibody recognises an epitope exposed when the GAG chains on CSPGs have been cleaved. The green staining indicates the area where the secreted enzyme is active.

Image courtesy of R-R. Zhao, J. Fawcett, J. Verhaagen & E. Muir.

Future work will entail assessing the efficacy of the construct in the rat model of SCI using the following approaches:

- **Gene therapy** - controlled production of chondroitinase ABC in mammalian neurons and/or glia to deliver the active enzyme directly to the injury site and facilitate repair of neural tissue
- **Cellular Therapy** - use of Schwann cells secreting chondroitinase ABC for in-vivo bridge grafts at injury site to encourage regenerating axons to traverse the lesion

Commercialisation and patent status

We are seeking to establish collaboration and licensing relationships for the commercialisation of this exciting technology.

This technology is protected by the UK patent application number 0712302.9 filed on 22/06/07.