

Optogenetically-Controlled Restoration of Muscle Function in ALS

Acronym : OptiMus

Principal Investigator:

Dr Barney Bryson

Grant : 160 000€

Project duration: two years

Project team :

From left to right :

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Machado, Linda Greensmith, Ivo

Lieberam



Summary of the research project

This avant-garde project is managed through a collaborative work between University College London and Kings College London. It started financed by us, in 2010, first results being recently published in Science. Second phase of the project is starting now through a co financing with Motor Neuron Association. T

This project involves a novel approach to restore muscle function following motor neuron loss, using a combination of stem cell based replacement of motor neurons and the revolutionary technique of optogenetics.

Briefly, we have shown that embryonic stem cell-derived motor neurons (ESC-MNs) can be transplanted into injured peripheral nerves that have lost their endogenous motor neuron axons and that these ESC-MNs can then reinnervate the target muscles (See Bryson et al, 2012, Science). Moreover, we genetically manipulated these ESC-MNs to express the light-sensitive ion channel, channelrhodopsin-2 (ChR2), so that when we optically stimulate these ESC-MNs with blue light, they can induce finely-controlled contraction of reinnervated muscles

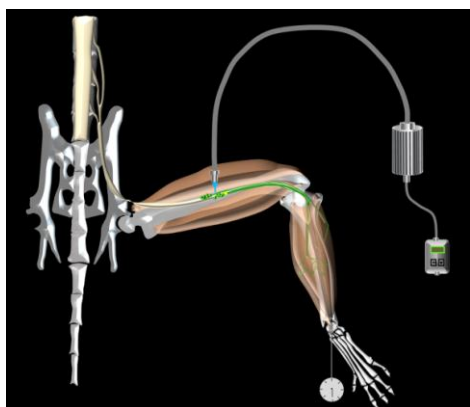


Figure 1:

The schematic illustrates the transplantation of embryonic stem cell-derived motor neurons into specific branches of an injured sciatic nerve. After 35 days, the transplanted motor neuron axons grow towards the muscles in the hind-limb to reinnervate them. At this time point, the nerve in the mid-thigh region was exposed and a blue light-source was used to 'optogenetically' activate the transplanted motor neurons, which resulted in finely controlled muscle contractions.

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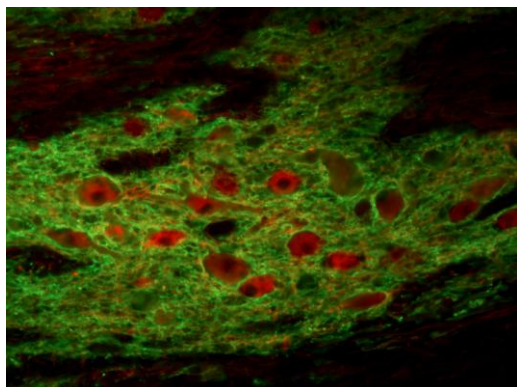


Figure 2:

Image of transplanted embryonic stem cell-derived motor neurons within the sciatic nerve; the transplanted neurons have been labelled with a functional marker of mature motor neurons (choline acetyltransferase; red) and the channelrhodopsin-2 protein (green), which enables the neurons to be controlled optogenetically ie by blue light.

This approach has the potential to overcome the normally permanent atrophy and paralysis of skeletal muscles that can occur as a result of diseases such as ALS as well as by traumatic neurological injury (eg. spinal cord injury). Our long-term aim is to use this approach to maintain function of the diaphragm muscle in models of ALS, since loss of function of this respiratory muscle is the main cause of death in ALS patients. To accomplish this, we are currently investigating whether it is possible to restore muscle function in the long term in mouse models of ALS, using an implantable optical stimulator device.

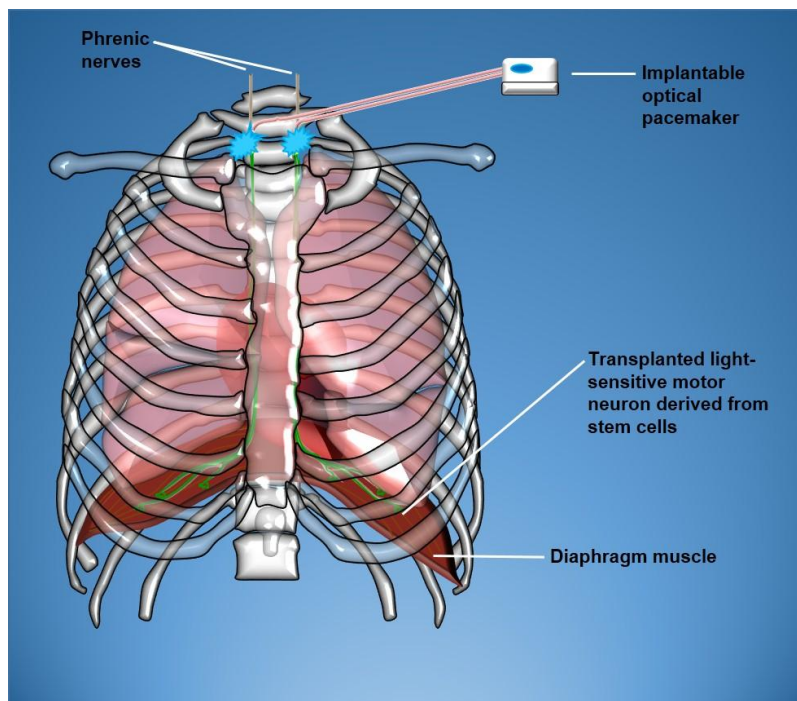
In order to fully evaluate the translational potential of this strategy for ALS patients, and to model the potentially toxic environment that may exist within the neuromuscular system of ALS patients, we now plan to develop this technique using the SOD1G93A mouse model of ALS. We have preliminary evidence that that these ESC-MNs can survive and innervate muscles in SOD1G93A mice *in vivo*, up until late-stage disease. In addition to establishing whether ESC-MNs also survive and functionally innervate target muscles in SOD1G93A mice, we also aim to develop a technique to optically stimulate muscles chronically *in vivo*, which will require the use of an implantable optical stimulator.

A near-term application of this technique would be to restore diaphragm muscle function in ALS patients suffering from respiratory insufficiency, using an implanted optical pacemaker, thereby enabling them to breathe without a mechanical ventilator.

This would not only greatly improve the quality of life of these patients, but could also prevent diaphragm muscle atrophy and may even reduce the risk of ventilator-associated pneumonia which is a common cause of death in ventilator-dependent patients. Ultimately, the ability of this novel biological interface to control complex motor functions is only limited by the sophistication of the optical control device.

Diagram of the ultimate clinical application of this research

Schematic representation of light-activated motor neuron transplants combined with an implantable optical pacemaker, which will enable ALS patients with compromised respiratory function to breathe without the need for mechanical ventilation.



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Relevant research articles for this project are :

1. Bryson J. B. , Barcellos Machado C, Crossley M, Stevenson D, Bros -Facer V, Burrone J, Greensmith L, Lieberam, I. Optical Control of Muscle Function by Transplantation of Stem Cell–Derived Motor Neurons in Mice, *Science*. 2014 Apr 4;344(6179):94-7. doi: 10.1126/science.1248523
2. Boyden, E. S., Zhang, F., Bamberg, E., Nagel, G. & Deisseroth, K. Millisecond-timescale, genetically targeted optical control of neural activity. *Nat Neurosci* **8**, 1263-1268 (2005).
3. Llewellyn, M. E., Thompson, K. R., Deisseroth, K. & Delp, S. L. Orderly recruitment of motor units under optical control in vivo. *Nat Med* **16**, 1161-1165 (2010).
4. Lin, J. Y., Knutsen, P. M., Muller, A., Kleinfeld, D. & Tsien, R. Y. ReaChR: a red-shifted variant of channelrhodopsin enables deep transcranial optogenetic excitation. *Nat Neurosci* **16**, 1499-1508 (2013).
5. Towne, C., Montgomery, K. L., Iyer, S. M., Deisseroth, K. & Delp, S. L. Optogenetic control of targeted peripheral axons in freely moving animals. *PloS one* **8**, e72691 (2013).

link to the lab website:

<http://www.ucl.ac.uk/ion/departments/sobell/Research/LGreensmith>

Dr Barney Bryson is Postdoctoral Research Associate in the Prof. Greensmith's Laboratory : Sobell Dept of Motor Neuroscience and Movement Disorders, Institute of Neurology, UCL, London.

His 5 most relevant publications are:

- Bryson J. B. , Barcellos Machado C, Crossley M, Stevenson D, Bros -Facer V, Burrone J, Greensmith L, Lieberam, I. Optical Control of Muscle Function by Transplantation of Stem Cell–Derived Motor Neurons in Mice, *Science*. 2014 Apr 4; 344(6179):94-7. doi: 10.1126/science.1248523
- **Bryson JB**, Hobbs C, Parsons MJ, Bosch KD, Pandraud A, Walsh FS, Doherty P, Greensmith L. Amyloid precursor protein (APP) contributes to pathology in the SOD1(G93A) mouse model of amyotrophic lateral sclerosis. *Hum Mol Genet*. 2012 Sep 1;21(17):3871-82.
- Petros TJ[†], **Bryson JB**[†], Mason C. Ephrin-B2 elicits differential growth cone collapse and axon retraction in retinal ganglion cells from distinct retinal regions. *Dev Neurobiol*. 2010 Sep 15;70(11):781-94. [†] Denotes equal first authorship
- Ma TC, Campana A, Lange PS, Lee HH, Banerjee K, **Bryson JB**, Mahishi L, Alam S, Giger RJ, Barnes S, Morris SM Jr, Willis DE, Twiss JL, Filbin MT, Ratan RR. A large-scale chemical screen for regulators of the arginase 1 promoter identifies the soy isoflavone daidzein as a clinically approved small molecule that can promote neuronal protection or regeneration via a cAMP-independent pathway. *J Neurosci*. 2010 Jan 13;30(2):739-48.v
- Deng K, He H, Qiu J, Lorber B, **Bryson JB**, Filbin MT. Increased synthesis of spermidine as a result of upregulation of arginase I promotes axonal regeneration in culture and in vivo. *J Neurosci*. 2009 Jul 29;29(30):9545-52.