Is Spinal Cord Repair a Reality?
Schwann Cell Transplantation for Subacute Spinal Cord Injury

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Potential mechanisms by which cells can contribute to post SCI-repair

- Neuroprotection
- Myelin repair
- Cellular replacement
- Axonal regeneration
- Trophic support
- Neuronal relays
- Immune modulation
Current and completed experimental trials of cell therapy for SCI

- **Proneuron**- Activated macrophages transplanted at the site of injury with 14d of injury. Phase 2 randomized.
- **Geron**- HESC differentiated to oligodendroglial cells transplanted at the site of injury with 14d. Phase 1 open label.
- **Stem cells Inc.**- NSC transplanted at the site of injury in subjects with chronic SCI. Phase 1 open label.
- **Miami Project**- Autologous Schwann cells transplanted in subjects with subacute SCI. Phase 1 open label.
- **Neuralstem (proposed)**- NSC in chronic SCI. Phase 1. Completed Phase 1 in ALS.
Phase 1 open label dose escalation safety and tolerability study of autologous Schwann cell transplantation in subacute Spinal cord injury

clinicaltrials.gov identifier NCT 01739023
Why Schwann Cells?

- Their essential role in peripheral nerve repair
- They can be isolated from sural nerve and purified and expanded in cell culture
- Autologous transplantation
- Substantial preclinical evidence of safety and limited efficacy
- SC enter the injured spinal cord anyway.
SC therapeutic mechanisms

• Growth-promoting extracellular matrix and membrane surfaces
• Production of neurotrophic molecules
• Axonal growth support
• Axonal ensheathment and myelination
Dissection of Fascicles from Nerve

Courtesy of Dr. Pat Wood
Sural nerve biopsy obtained from patient

Manual separation of nerve fascicles

Dissociate nerve fascicles using collagenase and dispase, 16-18 h

Wash out dissociation enzymes

Plate 1 million cells per plate on laminin coated tissue culture dishes in Schwann cell growth medium with antibiotics

Culture for about 1 wk-10 days. Resuspend and replate at 500,000 cells per dish on laminin coated tissue culture dishes in Schwann cell growth medium without antibiotics

Cryo-preserve excess cells

Sterility, purity & endotoxin check

Culture for about 1 wk-10 days. Resuspend cells and prepare for transplantation

Inject into the spinal cord injury site of the same patient (autologous transplantation)

 Courtesy of Dr. Pat Wood
S-100 positive Schwann cells at Passage 2.
EGFP-labeled SCs, implanted at 1 week post-injury, form a substantive bridge across the injured cord.

Courtesy of Dr. Damian Pearse
Day 0 Post contusion

Post contusion +
100 ul SC injection 225,000 cells/ul day 24
Biodistribution of GFP +VE autologous porcine SC autografts

Fluorescent Signal Intensity

# of Schwann Cells Represented by Positive Fluorescent Signal

Distance from Injection Epicenter (mm)

Rostral

Left

Right

Caudal

X-Axis

$45 - 150\mu L - 8DS$

$46 - 100 \mu L - 8DS$

$50 - 50\mu L - 8DS$

$51 - 50\mu L - 8DS$

$62 - 50\mu L - 60DS$

$63 - 45\mu L - 90DS$
5. Ensheathment-Myelination

[A] [B] [C] [D] [E] [F] [G] [H]

- GFP
- NF
- Hoechst

- GFP
- P0
- Hoechst

- GFP
- MBP
- Hoechst
NHP 9221 transplanted 2.3 years post-injury
Long term survival of GFP transplanted SC in the brainsteam of NHP 92221: A, B: anti Neurofilament red-594nm, anti GFP green-488nm, nuclei stained blue-Hoechst 33342; C: anti-CaM Kinase IIα nickel enhanced HRP precipitation. Once the animal reached a behavioral plateau, a transplant of aSCs was stereotaxically placed targeting the right pyramid after which primate presented improvement in its gait. Figures A, B show GFP positive signal at the pyramidal decussation 6 months after transplant (boxes). It is conceivable that improvement could be attributed to the activity of the SC including myelination. C: anti-CaM Kinase IIα specificity for the corticospinal tract stained on an adyacent section labeling the non lesioned left pyramid and its decussation confirm the SC localization. Bar C: 1500 µm, C-1:200 µm.
Figure 55. Myelination by transplanted SCs in NHP 92221. Confocal sequence. Micrographs, immunolabeled for green fluorescent protein, myelin protein zero (P0-red), and nuclei (Hoechst stain blue). Several P0 positive rings are present that clearly colocalize with GFP-ve SC and their nuclei in a very characteristic signet ring appearance. The arrow in A pointing to an interrupted green longitudinal signal, aligned with P0 that resembles a node of Ranvier. Arrows in B, C and D, show several examples of signet ring colocalization classical for SC producing myelin. Scale bar 10μm.
Goals of the Clinical trial

• Explore feasibility of aSC transplantation in SCI.
• Determine safety of aSC implanted at 30-42d post-SCI.
• The study will recruit 8 subjects with acute thoracic spinal cord injury who will provide informed consent and then undergo removal of a portion of one sural nerve from which the Schwann cell transplant will be manufactured in an experienced cGMP facility.

• Dose escalation, 5, 10, 15 million SC.
Aspects of Feasibility

• Subjects that meet study criteria
• Adequate MRI visualization of the injury site
• Nerve harvest
• Nerve quality and initial cell yield
• Culture growth kinetics and purity adequate to meet release criteria.
• Subject consent
• Successful cell delivery
Safety

• **Procedural.** Implantation of aSC does not worsen the neurological deficit

• **Biological and Neurological.** Implanted aSC do not cause serious AE such as a cellular mass, damaging inflammation, increased cavitation, loss of neurological function, worsening of pain, spasticity or other sequelae.
Inclusion Criteria:
1) Persons with traumatic SCI that occurred within the previous 5 (7) days.
2) Between the ages of 18 and 50 (60) at last birthday.
3) SCI at a thoracic level between T3-T11 as defined by MRI and the most caudal level of intact motor and sensory function on the International Standards for Neurological Classification of Spinal Cord Injury (ISNCSCI).
4) Acute SCI with ISNCSCI grade A impairment at time of enrollment.
Exclusion Criteria:
1) Persons with penetrating injury of the spinal cord or complete transection of the cord, including bone fragment lacerations, as identified by magnetic resonance imaging (MRI).
2) Persons with a lesion in the conus medullaris, cauda equina, or lower extremity peripheral nerve.
3) Persons unable to safely undergo an MRI.
4) Persons in whom adequate MRI imaging cannot be obtained.
5) Other traumatic injuries (e.g., CHI, another level of SCI) affecting the ability to provide informed consent and participate fully in rehabilitation.
7) Persons with self-reported persistent severe neuropathic pain, inadequately controlled by non-narcotic medication.
8) Presence of systemic disease that might interfere with subject safety, compliance, or evaluation of the condition under study.
9) Presence of any unstable medical or psychiatric condition that could reasonably be expected to subject the participant to unwarranted risk from participation in the study or result in a significant deterioration of his/her clinical course.
10) Body Mass Index (BMI) > 35.
11) History of active substance abuse.
Current status

• 2 subjects transplanted with 5 million SC.
• Subject 1 is 8 months post-transplant.
• Subject 2 is 5 months post-transplant.
• No loss of neurological function, no unexpected changes on MRI.
• No serious procedural AE
Neuroprotection SCI Clinical trials at the University of Miami

- North American Clinical Trials Network. Completed Phase 1 Riluzole study
- NACTN “RISICS” Phase 2. Cervical A-C
- Hypothermia Observational Study. Cervical ASIA A
- Acorda Therapeutics. AC105-Phase 1/2 . A-C
The Second Step

Testing Schwann Cell Transplantation in Humans

The first step, generating all of the pre-clinical safety and efficacy data to justify the testing of Schwann cell transplantation in humans, has been completed. The Miami Project to Cure Paralysis has submitted its Investigational New Drug (IND) application to the Food and Drug Administration (FDA) requesting permission to begin a Phase I clinical trial to evaluate the safety of autologous human Schwann cell transplantation a few weeks after a spinal cord injury has occurred. The second step, now, is for the FDA to approve the application so we can start the trial.