Kenny Li--EN.585.685.81--Methods in Neurobiology

Module 11 Case Study

Background

Studies, such as the one done by Al-Majed et al., have proven that “electrical stimulation applied immediately after a surgical repair of a cut femoral rodent’s nerve promoted regeneration of motor axons over a 25 mm distance from the site of injury after 3 weeks” [3]. In another study, electrical stimulation was associated with approximately 30-50% improvement in axon regrowth and regeneration [4]. As a result, we want to investigate if the same technique can be applied to injuries involving neuroma-in-continuity, which is a neuroma that results in the distal part of a nerve to no longer function [5].

Aim

To establish an experiment in order to determine whether or not electrical stimulation will be an effective treatment or method in improving axon regeneration in the peripheral nerves as well as the overall functional recovery in neuroma-in-continuity nerve injury in rat animal models.

Research Plan

- As stated in the aims, we will be using rat animal models, for a couple reasons:
    - Compared to mice, they have larger nerves, making it easier to perform microsurgeries as well as having more standardized and comparable function tests (as we have learned in lecture).
    - Rats are more resilient compared to mice, and are able to maintain longitudinal rate of nerve regeneration at a rate of ~1mm/day [1].
- We will be performing neuroma-in-continuity (NIC) type injuries on the right sciatic nerve of our rat models, and there will be a total 3 groups of rats to be studied in our experiments:
    - The first group will act as our control: we will place 3-4 rats under anesthesia and then perform a cut on the right sciatic nerve. The ends of the nerve will then be capped using silicone caps to ensure that the nerves will not be able to regenerate. No electrical stimulation will be carried out.
    - The second group will consist of 5-6 rats undergoing a similar procedure carried out in our control rats; however, no silicone caps will be placed on the nerves, but rather, the right sciatic nerve will be crushed to simulate NIC injury. This group will not receive electrical stimulation.
    - Our third and final group consisting of 5-6 rats will undergo the same procedure as the second group; however, after crushing the nerve, electrical stimulation will be administered to test if there are any therapeutic results. This will be done by placing electrodes onto the proximal nerve and applying a pulse voltage for approximately an hour.
- In order to test whether or not electrical stimulation can improve nerve regeneration and restore some function, we will have our 3 groups of rats undergo behavioral tests at weeks 4,6,8, and 10. After the physicals, the rats will be sacrificed to analyze for axonal regrowth.
    - The physical/behavioral test will likely be the horizontal ladder rung walking task, which is commonly used to assess the walking skill of the forelimb and hindlimb

of rats. Slips or missteps will be counted, scored, and recorded [2]. We will then compare the results between the 3 groups of rats.

- If our hypothesis is correct, we expect to see the rats with no electrical stimulation perform better on the physical test compared to our control rats, but poorer to the rats that received the electrical stimulation. (Rats in Group 3 recover faster than Group 2 Rats).
    - We should also see more evidence of axonal regrowth in Group 3 rats compared to Group 2 rats.

Application

If we find that applying electrical stimulation immediately after an in-continuity nerve injury can indeed allow for faster axonal recovery and restoration in function, then we can assume after further experimentation, a similar procedure can be applied to humans suffering from in-continuity nerve injuries.

Works Cited

[1] Farah, Zayd, et al. “A Concise Review of Common Animal Models for the Study of Limb Regeneration.” *Organogenesis*, Taylor & Francis, 2 July 2016, https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4993303/.

[2] Metz, Gerlinde A, and Ian Q Whishaw. “The Ladder Rung Walking Task: A Scoring System and Its Practical Application.” *Journal of Visualized Experiments : JoVE*, MyJove Corporation, 12 June 2009, https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2796662/.

[3] Al-Majed, A A, et al. “Brief Electrical Stimulation Promotes the Speed and Accuracy of Motor Axonal Regeneration.” *The Journal of Neuroscience : the Official Journal of the Society for Neuroscience*, U.S. National Library of Medicine, 2000, https://pubmed.ncbi.nlm.nih.gov/10729340/.

[4]Singh, Bhagat, et al. “Accelerated Axon Outgrowth, Guidance, and Target Reinnervation across Nerve Transection Gaps Following a Brief Electrical Stimulation Paradigm.” *Journal of Neurosurgery*, U.S. National Library of Medicine, 2012, https://pubmed.ncbi.nlm.nih.gov/22149377/.

[5] Mavrogenis, Andreas F, et al. “Current Treatment Concepts for Neuromas-in-Continuity.” *Injury*, U.S. National Library of Medicine, 2008, https://pubmed.ncbi.nlm.nih.gov/18715561/#:~:text=A%20neuroma%2Din%2Dcontinuity%20is,that%20no%20longer%20functions%20properly.

[6] Shapira, Yuval. “Brief Electrical Stimulation Promotes Nerve Regeneration Following Experimental in-Continuity Nerve Injury.” *Neurosurgery*, U.S. National Library of Medicine, 2019, https://pubmed.ncbi.nlm.nih.gov/29893910/.