A Novel, High-Throughput Electrophysiology Platform for Compound Screening with a Peripheral Nerve Microphysiological System

1025-B

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AxoSim Human Data, Faster.

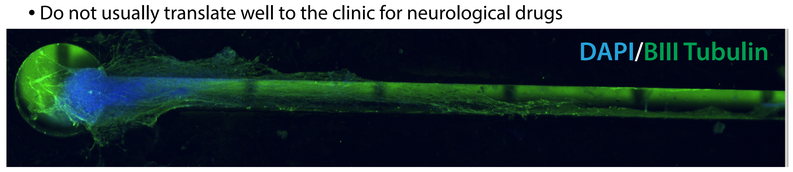
NerveSim® Powered by Nerve-On-A-Chip®

Overview

- AxoSim has developed a novel microphysiological system (MPS), the NerveSim®, to model peripheral nerves *in vitro* using rat- or human-derived cells
- The platform uses an embedded electrode array (EEA) to record functional electrophysiological signals from peripheral nerve cultures
- NerveSim® is a scalable, automated platform allowing measurement of multiple clinically relevant electrophysiological metrics such as nerve conduction velocity (NCV), peak response amplitude (AMP), and threshold stimulus strength (TSS)
- Quantification of these metrics enables compound screening for peripheral neurotoxicity, neuroprotection, and neurorehabilitation

Introduction

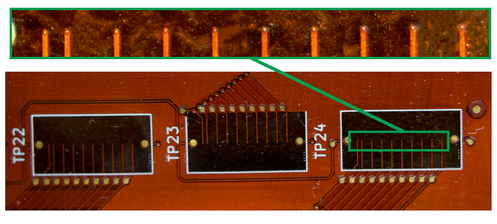
- The current neurological drug development pipeline is flawed
  - 94% of neurological drugs fail at or before clinical trials
  - Drug development requires ~$2.6 billion per drug
- Reliance on animal studies is the main problem
  - Expensive to gather functional data necessary for neurological drug testing
  - Difficult to scale or automate
  - Do not usually translate well to the clinic for neurological drugs



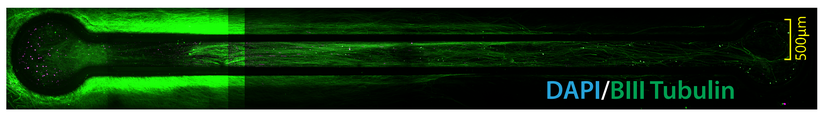
- AxoSim has developed the NerveSim® platform, a novel peripheral nerve MPS using an EEA, as a scalable alternative for the pharmaceutical industry to measure functional electrophysiology from either rat or human models
  - 3D tissue culture model to best mimic *in vivo* nerve growth
  - Designed to be scalable and automatable from the ground up
  - Collects clinically relevant electrophysiological metrics from primary rat or human induced pleuripotent stem cells (iPSCs) to maximize translation

Methods

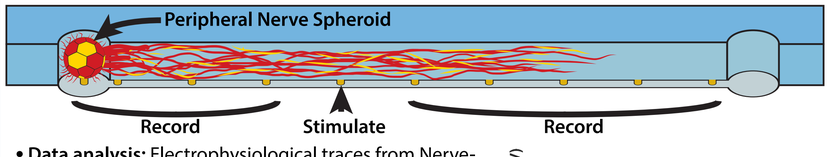
- **EEAs:** Custom 24-well tissue culture plate with 10 microelectrodes per well that are used for recording or current-based stimulation



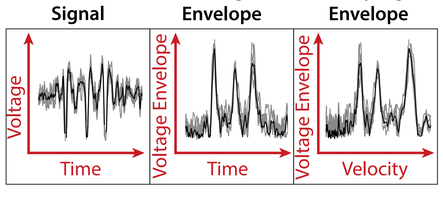
- **Microphysiological system:** Polymer construct guides axonal growth of peripheral nerve spheroids along electrode array to mimic nerve fiber tract. Spheroids grown in custom media formulation for 28-42 days to ensure dense axonal growth along electrodes

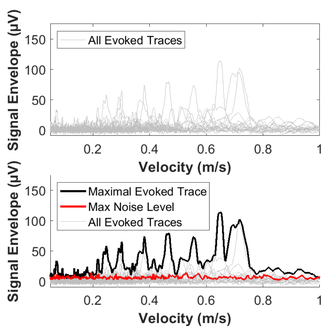


- **Compound dosing:** Positive control neurotoxic compounds are applied to mature NerveSim® cultures for 7-14 days, measuring electrophysiology before, during, and after dosing
- **Electrophysiology:** NerveSim® samples were stimulated with current-based electrical stimulation in parallel at multiple distal electrodes while recording on the remaining electrodes. Each stimulation used a stimulation current ramp (1 to 64 µA) to generate compound action potentials (CAPs) on nearby electrodes

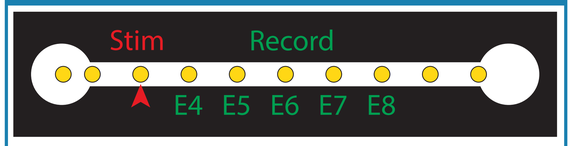


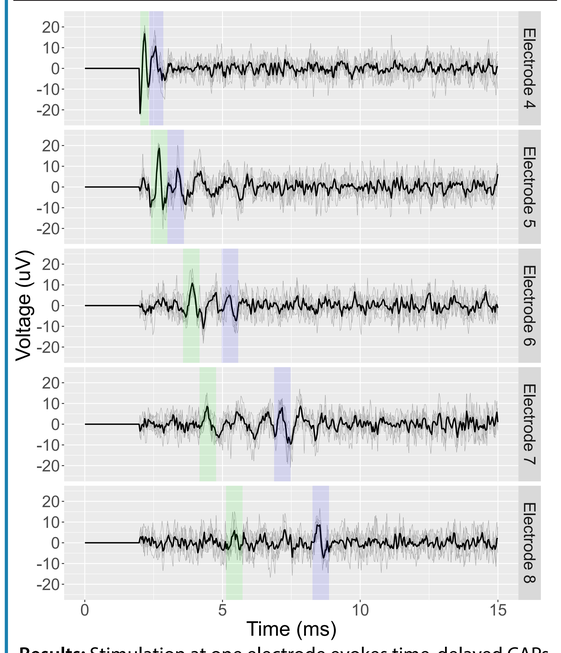
- **Data analysis:** Electrophysiological traces from NerveSim® were processed and then converted to the velocity domain. Population level responses for each well were obtained using a maximal velocity projection (MVP)





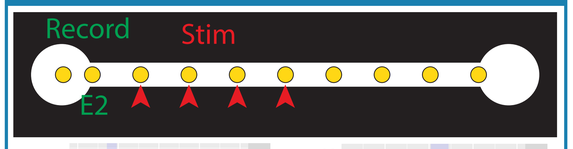
Microelectrode Recordings

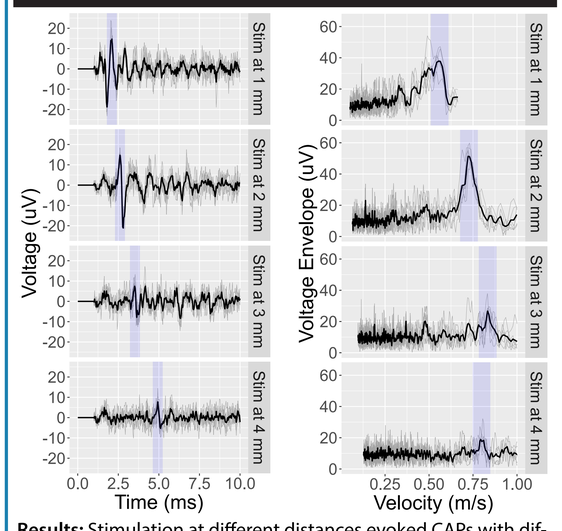




Results: Stimulation at one electrode evokes time-delayed CAPs on nearby electrodes as the signals travel at conduction velocity

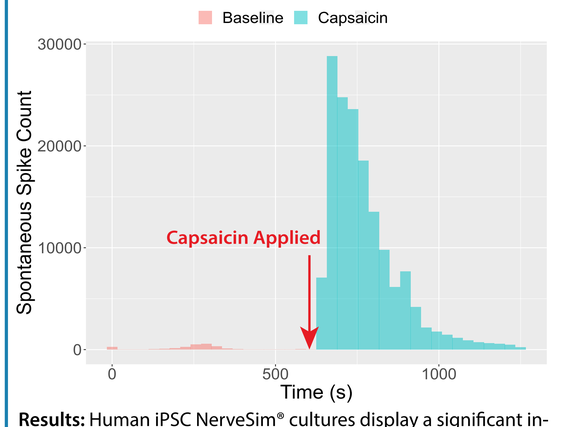
Spatial Electrophysiology





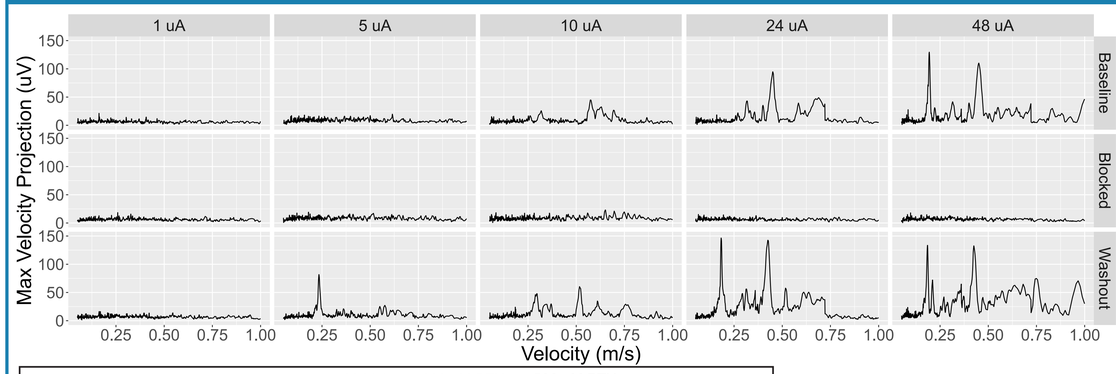
Results: Stimulation at different distances evoked CAPs with different time delays but with similar conduction velocities

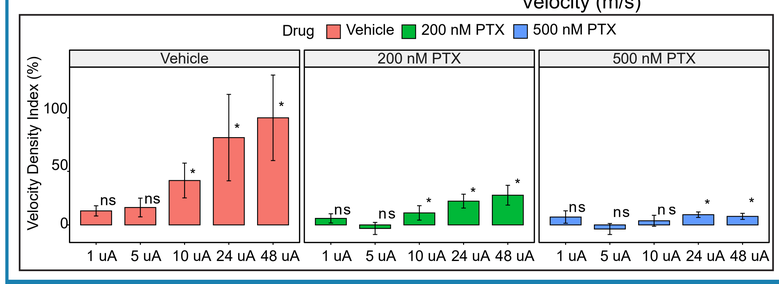
Spontaneous Activity



Results: Human iPSC NerveSim® cultures display a significant increase in spontaneous activity with the application of 1 µM Capsaicin. Increasing spontaneous electrophysiological activity has been associated with activation of a pain phenotype

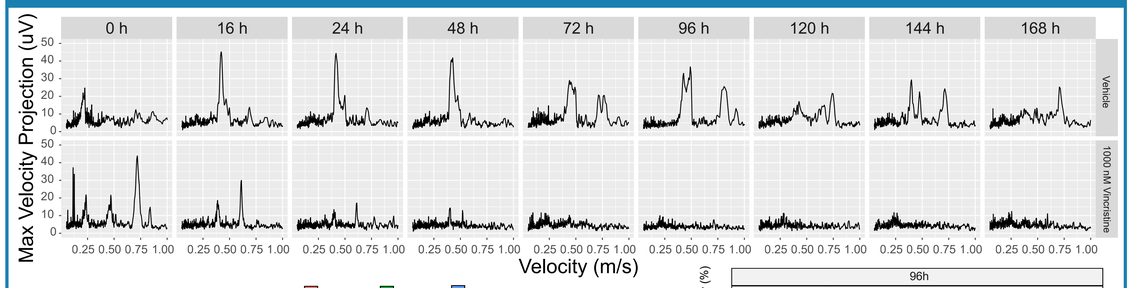
Threshold Stimulus Strength

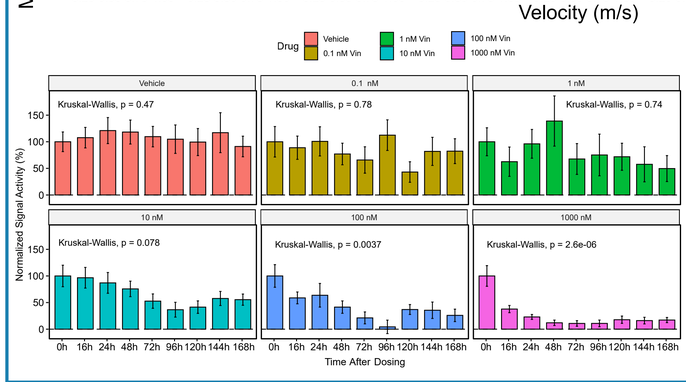


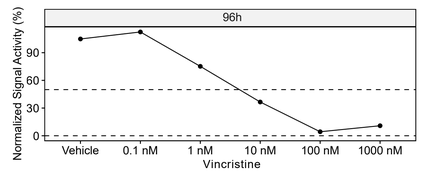


Top: Stimulation of human iPSC culture with a current ramp shows that 10 uA is the TSS at baseline. Application of a reversible blocker eliminates the response and it returns after washout more sensitvie with a TSS of 5 uA
Left: TSS from a rat culture dosed with paclitaxel (PTX) showing differences across dosing conditions

Longitudinal Electrophysiology

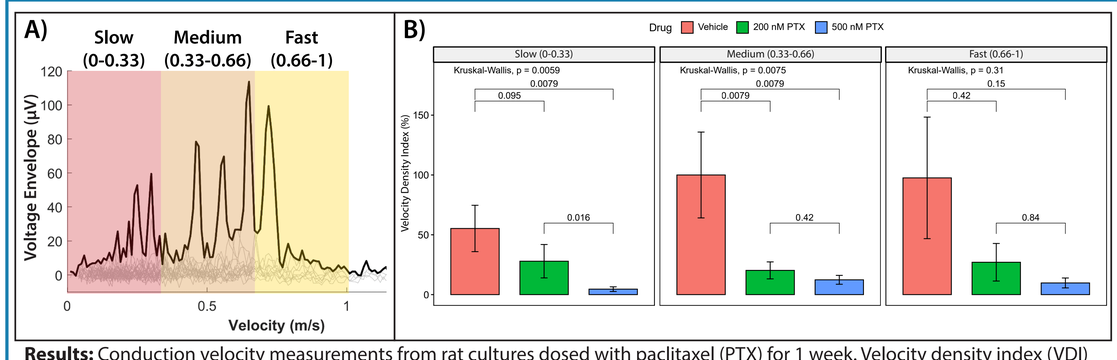






Top: MVP traces from a human NerveSim® dosed with a DMSO vehicle (top) and 1000 nM vincristine (bottom) over a 1 week period
Left: Total signal activity for 5 concentrations of vincristine and the DMSO vehicle over 1 week of time
Right: Dose response curve for vincrstine at 96 hours

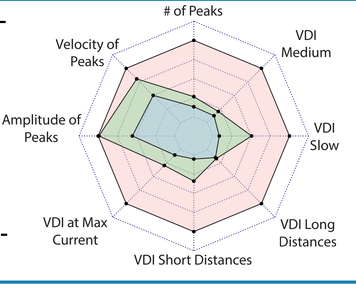
Neurotoxic Effects on Conduction Velocity



Results: Conduction velocity measurements from rat cultures dosed with paclitaxel (PTX) for 1 week. Velocity density index (VDI) at max current (48 µA) was separated into velocity bins (A) and compared across different dosing conditions (B). High doses of PTX caused a significant reduction in VDI across all velocities while low doses only caused a significant decline in medium speed responses

Conclusions

- The NerveSim® platform provides collection of data-rich electrophysiological metrics that can provide insights into neurotoxicity, neuroprotection, and neurorehabilitation
- Functional electrophysiological measurements provide the same quantitative metrics as clinical electrophysiology
- Future screening of multiple compounds with different mechanisms will generate a database for predicting compound mechanisms based on high-throughput electrophysiology



For more information: