The Effect of Electric Fields on Axon Projection

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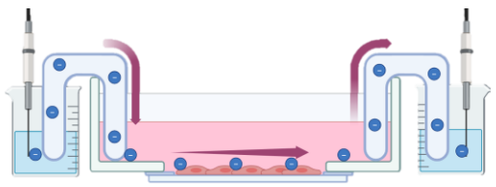
Background

Electric field effects on cell populations have been investigated since 1960. While studies have shown interesting results, no mechanism has been identified, and cells behave in unexpected ways with no proven hypothesis.

A neurons growth cone is made of positive leading F-actin bundles [1] so the hypothesis is that neurons will differentiate in the direction of an applied DC electric field.

Literature analysis

Traditional systems comprised of agar salt bridges to reduce electron degradation and metal ion interactions in the cell media.



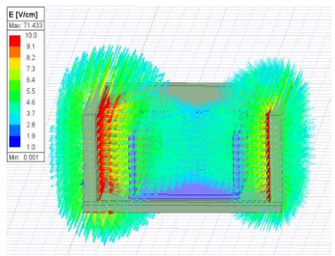
The current systems have not isolated the effects of electron interaction and electric field. DMEM, a major component of most cell media, has a dielectric constant of ~80 [2] and a conductivity of ~2mS/cm.

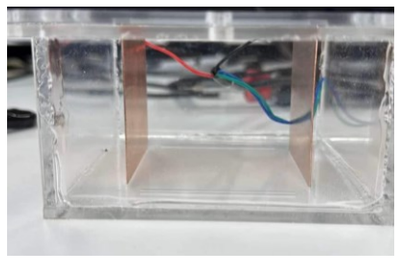
$$R = \frac{L}{\sigma \cdot A}, I = \frac{V}{R}$$

Using 3cm and a voltage of 30 volts, the current between the plates results in 20mA, which cannot be ignored.

Device design and fabrication

A parallel plate capacitor capable of growing cells was simulated and constructed to protect the circuit from the highly humid environment in which cells are grown.

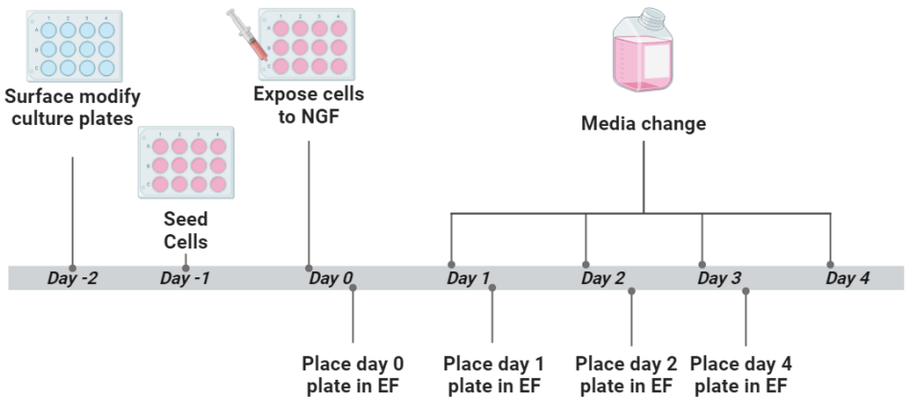




The system was powered by a 12v outlet, which was stepped up to 30 volts to power operational amplifiers driven by an Arduino.

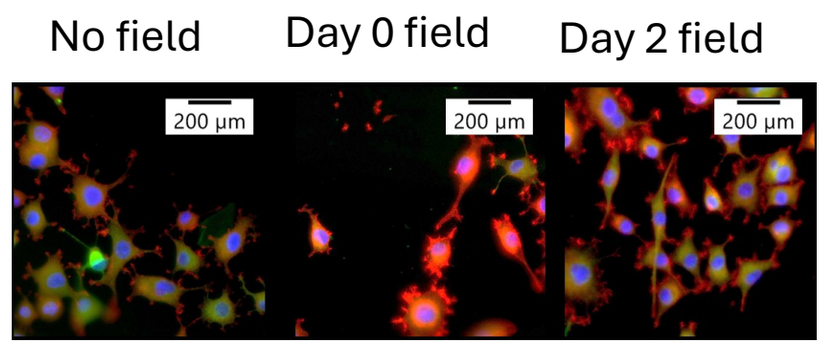
Methods: Cell culture

The differentiation process was optimised for PC12 rad pheochromocytoma to determine best NGF concentrations, cell density and surface treatment. The cells then followed the optimised process, with exposure to the field at various timepoints after initial exposure to NGF.



Results

The cells can successfully differentiate into neurons both in and out of the field. Morphological differences show that cells have reduced differentiation and trend towards the anode, showing projections in the direction of the electric field.



Discussion and Future Work

While there is an identified change in morphology due solely to the electric field, the mechanism has still not been determined. Many more conditions and factors must be considered to assess the underlying biological mechanics accurately. To move forward with this, the system should be redesigned to incorporate microfluidic systems to efficiently assess gene and protein changes in the cells under various electric field conditions. Similarly, multiple compact versions of the device can be created to be run in parallel to test various electric field stimulation conditions.

References

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[1] Cammarata, G. M., Bearce, E. A., & Lowery, L. A. (2016). Cytoskeletal social networking in the growth cone: How +TIPs mediate microtubule-actin cross-linking to drive axon outgrowth and guidance. Cytoskeleton, 73(9), 461–476. https://doi.org/10.1002/cm.21272

[2] Badea, I., Alwani, S., Kaur, R., Michel, D., Chitanda, J. M., Verrall, R., & Karunakaran, C. (2016). Lysine-functionalized nanodiamonds as gene carriers: development of stable colloidal dispersion for in vitro cellular uptake studies and siRNA delivery application. *International Journal of Nanomedicine*, 687. https://doi.org/10.2147/ijn.s92218

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