

A microscopic image of axons, showing a dense network of blue, fibrous structures. The axons are arranged in a somewhat curved, parallel pattern, with many fine, branching fibers. The background is dark, making the blue axons stand out.

EMBO
Workshop

Axons 2021:

Structure and function

04 – 07 October 2021 | Virtual

Abstract Book

v. 1

Wed-PO-2-03

Morphometric comparison of neurite outgrowth in rat DRG cultures treated with DCPIB and BpV

Kamal AlJamal ¹, Gordana Stojadinovic ², Miroslav Zivic ¹, Natasa Todorovic ²

¹ *University of Belgrade - Faculty of Biology, Belgrade, Serbia*

² *Institute for Biological Research "Siniša Stankovic" , University of Belgrade, Belgrade, Serbia*

The chloride channel VRAC has been implicated in apoptosis, proliferation, transport of organic acids and regulatory volume decrease. Regulated by RAS/cGMP and by the ionic strength, it mediates efflux of anions (Cl⁻, glutamate, glutathione, taurine, cGAMP). Knowing that chloride transport activity, mediated by NKCC, is indispensable for axon growth, we hypothesized that VRAC could have a role in neurite elongation mechanism. Experimental treatments compared to control were: VRAC selective inhibitor DCPIB (10 μM); BpV (phen) (80 nM), an inhibitor of PTEN. Axon growth was measured on cultured dissociated dorsal root ganglia neonatal neurons fixed 24h after treatment and immunostained with anti-neurofilament H (NF-H) phosphorylated antibody. FIJI Simple Neurite Tracer was used for morphometry of individual neurons. 24h post treatment, total neurite length compared to control was decreased by BpV (p=0.0058) and to a lesser extent by DCPIB. In all measured parameters related to the degree of branching, BpV group neurons had small dispersion of values. In addition, DCPIB treated cells had larger fractal dimension (<0.05) than control neurons. The effect of DCPIB is mostly related to an increase in Fractal Dimension parameter, according to Principal Component Analysis. The increase in the number of primary branches per increment of cable length is significantly smaller in DCPIB group than in control and BpV -treated neurons. The DCPIB effect demonstrates that an anion current, presumably VRAC, has a role in the process of proper branching and directional elongation of neonatal DRG neurites.