Protein tyrosine phosphatase

Protein tyrosine phosphatases are a group of enzymes that remove phosphate groups from phosphorylated tyrosine residues on proteins. Protein tyrosine (pTyr) phosphorylation is a common post-translational modification that can create novel recognition motifs for protein interactions and cellular localization, affect protein stability, and regulate enzyme activity. As a consequence, maintaining an appropriate level of protein tyrosine phosphorylation is essential for many cellular functions. Tyrosine-specific protein phosphatases (PTPase; EC 3.1.3.48) catalyse the removal of a phosphate group attached to a tyrosine residue, using a cysteinyl-phosphate enzyme intermediate. These enzymes are key regulatory components in signal transduction pathways (such as the MAP kinase pathway) and cell cycle control, and are important in the control of cell growth, proliferation, differentiation, transformation, and synaptic strengthening.

PTPs are known to be signaling molecules that regulate a variety of cellular processes including cell growth, differentiation, mitotic cycle, and oncogenic transformation. This PTP contains an extracellular region, a single transmembrane segment and two tandem intracytoplasmic catalytic domains, and thus represents a receptor-type PTP. The extracellular region of this protein is composed of multiple Ig-like and fibronectin type III-like domains. Studies of the similar gene in mice suggested that this PTP may be involved in cell-cell interaction, primary axonogenesis, and axon guidance during embryogenesis. This PTP has been also implicated in the molecular control of adult nerve repair. Four alternatively spliced transcript variants, which encode distinct proteins, have been reported.

In vitro studies showed that PTPσ was upregulated in growth cones in a chondroitin sulfate proteoglycans (CSPGs) rich gradient. Furthermore, although PTPσ is normally distributed throughout neurons when CSPGs were absent, PTPσ levels were increased in the growth cones when CSPGs were present. This marked rise in growth cone PTPσ levels coincided with growth cone overadherence to proteoglycans; the PTPσ-CSPG interaction caused beaded growth cone structure and prevented further axonal growth. These revelations further illuminate the ability of PTPσ to inhibit regeneration of nerve fiber because upregulated PTPσ interacted with the CSPGs to stabilize dystrophic growth cones [1)].

To counter the PTPσ-CSPG–mediated inhibition of growth cone regeneration, Dr Jerry Silver’s team developed a PTPσ wedge domain mimetic, intracellular sigma peptide (ISP), that binds human PTPσ and is conjugated to a TAT domain for membrane permeability. ISP was developed to mimic and act as a PTPσ wedge, thereby inhibiting the function of mature and immature PTPσ proteins expressed in regenerating axons exposed to proteoglycans. In vitro ISP treatment promoted sensory neuron growth cone extension and Axon regeneration through a CSPG gradient in a dose-dependent fashion; ISP doses above the optimal 2.5 mmol/L reduced cell attachment.

Time-lapse imaging illustrated that a CSPG gradient collapsed growth into “beads” (A), whereas the cones continued to reform with ISP treatment (B) and cross the CSPG gradient (C). This observed remarkable regeneration in the presence of scar tissue resembles the effects of chondroitinase ABC, an enzyme known to catalyze CSPGs and to improve Axon regeneration. ISP treatment for control adult PTPσ-null neurons did not significantly increase axonal crossings through the CSPG gradient.

n vivo studies of chronically injured rats with a contusive SCI were conducted to understand the extent to which ISP aids Axon regeneration. Tissue data revealed that ISP inactivation of PTPσ did not result in significant neuroprotection; no difference in lesion size and white matter sparing was observed compared with vehicle-treated controls.

The absence of neuroprotection limited the ability of ISP to improve neuronal recovery. Interestingly,

despite the absence of neuroprotection, systemic intraperitoneal ISP injections led to significantly more reformed serotonergic (5-HT) fibers.

5-HT fibers are associated with movement and micturition, correlating with the significantly higher functional recovery and voiding frequency observed in ISP-treated rats. Furthermore, in locomotor recovery assessment, ISP-treated rats scored significantly higher in open-field Basso-Beattie-Bresnahan functional recovery tests compared with vehicle controls. The ISP peptide, conjugated to FITC, was detected in intact nervous system tissue after systemic injections, proving its ability to cross the blood-brain barrier. Understanding the therapeutic potential of ISP in the chronic stages of SCI injury is important to fully realize its capacity to support functional recovery.

This systemic delivery of ISP to prevent PTPσ-mediated inhibition around the glial scar is potentially powerful for improving regeneration after SCI from trauma. Inhibiting the disabling CSPG scar barrier with ISP can help solve a significant obstacle in Axon regeneration by improving growth cone motility after injury. One significant limitation that remains for therapeutic strategies involving PTPσ inhibition is the absence of neuroprotection, because the damaged ascending and descending spinal pathways remain at risk for secondary damage after trauma. ISP treatment is therefore only part of the answer. Combining ISP with neuroprotective treatments could further minimize secondary damage and amplify the regenerative capacity of injured axons after SCI. It is envisioned that local ISP release from conduits in nerve grafts may be useful to prevent retraction of axonal growth cones and to enhance bypassing the CSPG scar to cross the graft interface. Further research to compare systemic and locally sustained ISP release will be essential to determine the optimal therapeutic timing, route and dosing, and related nerve graft therapies.

This study raises many questions: How is neuroplasticity involved? Why is ISP-mediated PTPσ inhibition correlated with 5-HT fiber regeneration? Is this a backup regenerative pathway in the post-SCI environment? How can ISP work beyond contusive injury models? Answering these and other relevant questions is key for potential translation of this novel ISP strategy beyond animal models to human clinical trials [2].

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2)
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