Cellular Neurobiology
BIPN140

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http://classes.biology.ucsd.edu/bipn140.FA16/

The place to go for 1) the syllabus, 2) lecture outlines, 3) ppts, 4) problem sets, 5) original research articles, 6) section times and places, 6) office hours, 7) previous exams.
Frequency Coding of Information in the Nervous System

Adrian and Zotterman, 1926

Frequency of action potentials

Weight hanging from muscle
Diversity of Morphology

(A) Neurons in mesencephalic nucleus of cranial nerve V

(B) Retinal bipolar cell

(C) Retinal ganglion cell

(D) Retinal amacrine cell

*NEUROSCIENCE, Fourth Edition, Figure 1.2 (Part 1)*
Figure 1.2 Some nerve cell morphologies found in the human nervous system (Part 2)

(E) Cortical pyramidal cell

(F) Cerebellar Purkinje cells

**NEUROSCIENCE, Fourth Edition, Figure 1.2 (Part 2)**
Figure 1.3 The major light and electron microscopical features of neurons (Part 1)
Figure 1.6 Visualizing nerve cells and their connections

Studying structure

(A) Golgi stain/ cortical neurons
(B) Golgi stain/ Purkinje neurons
(C) Dye injection/ retinal neurons
(D) HRP (enzyme) injection/ autonomic neuron

(E) Cresyl violet/RNA/ cortical neurons
(F) Nissl stain/RNA/ cortical neurons
(G) Nissl stain/RNA/ olfactory bulb

NEUROSCIENCE, Fourth Edition, Figure 1.6
Figure 1.12 Use of genetic engineering to reveal pathways within the nervous system

- **Promoter**
- **Reporter gene** e.g. GFP, Green Fluorescent Protein

- **Cell body**
- **Sensory endings in skin**
- **Peripheral axon**
- **Dorsal root ganglion**
- **Central axon**
- **Axon terminals in spinal cord**
- **Spinal cord segment**
Figure 1.7 A simple reflex circuit, the knee-jerk response

Studying function

1. Hammer tap stretches tendon, which, in turn, stretches sensory receptors in leg extensor muscle

2. (A) Sensory neuron synapses with and excites motor neuron in the spinal cord
   (B) Sensory neuron also excites spinal interneuron
   (C) Interneuron synapse inhibits motor neuron to flexor muscles

3. (A) Motor neuron conducts action potential to synapses on extensor muscle fibers, causing contraction
   (B) Flexor muscle relaxes because the activity of its motor neurons has been inhibited

4. Leg extends
Figure 1.8 Relative frequency of action potentials in different components of the myotatic reflex
Figure 1.9 Intracellularly recorded responses underlying the myotatic reflex
Figure 1.13 Single-unit electrophysiological recording from cortical pyramidal neuron

(A) Somatic sensory cortex

(B) Receptive field (center)
Receptive field (surround)

- Touch in the center of receptive field increases cell firing
- Touch in the surround of receptive field decreases cell firing
- Touch outside of receptive field has no effect

Activity of cortical neuron

Period of stimulation
Figure 1.4  Distinctive arrangement of cytoskeletal elements in neurons (Part 1)

(A) tau, binds microtubules

(B) actin

(C) epithelial cell

(D) glial cell

actin

actin

Figure 1.4 Distinctive arrangement of cytoskeletal elements in neurons (Part 2)
Axoplasmic transport

A. *Definition*: movement of materials inside the axon, both away from and toward the cell body

B. *Rates*:• slow/ 1mm/day orthograde (anterograde)
  • fast/  400 mm/day orthograde (anterograde)
  • intermediate/ 250-300 mm/day retrograde

C. *Detection*: • direct visualization
  • arterial cuff/ligature (thread)
  • radioactive tracers

D. *Mechanisms*: • slow requires cell body, involves bulk flow
  • fast and intermediate require microtubules
    kinesin - motor molecule - orthograde transport
    dynein - motor molecule - retrograde transport
    each links cargo to microtubules; ATP hydrolysis leads to movement
Figure 1.5 Varieties of neuroglial cells

(A) Astrocyte
(B) Oligodendrocyte
(C) Microglial cell

(D) Astrocytes/culture
(E) Oligos/culture
(F) PNS myelinated axons/nodes of Ranvier
(G) Microglia
A Critical Period for Social Experience–Dependent Oligodendrocyte Maturation and Myelination

Manabu Makinodan, Kenneth M. Rosen, Susumu Ito, Gabriel Corfas

Early social isolation results in adult behavioral and cognitive dysfunction that correlates with white matter alterations. However, how social deprivation influences myelination and the significance of these myelin defects in the adult remained undefined. We show that mice isolated for 2 weeks immediately after weaning have alterations in prefrontal cortex function and myelination that do not recover with reintroduction into a social environment. These alterations, which occur only during this critical period, are phenocopied by loss of oligodendrocyte ErbB3 receptors, and social isolation leads to reduced expression of the ErbB3 ligand neuregulin-1. These findings indicate that social experience regulates prefrontal cortex myelination through neuregulin-1/ErbB3 signaling and that this is essential for normal cognitive function, thus providing a cellular and molecular context to understand the consequences of social isolation.
What is myelin?
The wrapping of oligodendrocyte membrane around axons. It increases the speed of propagation of action potentials.

What are the authors’ big questions?
Does social experience regulate myelination? How?

Why are these interesting questions?
If we can answer them, then we have an insight into the mechanism by which social isolation retards anatomical and functional neural development in response to environmental experience!

What was previously known?
Early social isolation causes behavioral and cognitive deficits in the adult that correlate with alterations in myelin.
Prefrontal cortex (PFC)-dependent behaviors and medial PFC oligodendrocytes are altered by juvenile isolation (day 21>day 63)

IS = isolation; RE = regular environment; EE = enriched environment
Figure 2

The effects of social isolation on oligodendrocytes and behaviors occur only during a **critical period** (day 21>day 35)

IS = isolation; RE = regular environment; EE = enriched environment
What we know so far

1. Social isolation leads to altered oligos and altered behavior, during a critical period.

2. NRG1 (ligand) – ErbB (receptor) signaling is important for myelin formation (previous research).

3. Social isolation leads to reduction in NRG1 (data in the paper, not presented today) and thus NRG1-ErbB signaling.

So..................

4. Does elimination of ErbB3 exclusively in oligos negatively affect myelin formation and behavior?
Elimination of ErbB3 exclusively in oligos negatively affects myelin formation and behavior.

We have a molecular explanation for cognitive changes following social Isolation!

Cre+ = ErbB3 eliminated
Next time: Electrical Signaling
Control of Local Protein Synthesis and Initial Events in Myelination by Action Potentials

Hiroaki Wake, Philip R. Lee, R. Douglas Fields*

Formation of myelin, the electrical insulation on axons produced by oligodendrocytes, is controlled by complex cell-cell signaling that regulates oligodendrocyte development and myelin formation on appropriate axons. If electrical activity could stimulate myelin induction, then neurodevelopment and the speed of information transmission through circuits could be modified by neural activity. We find that release of glutamate from synaptic vesicles along axons of mouse dorsal root ganglion neurons in culture promotes myelin induction by stimulating formation of cholesterol-rich signaling domains between oligodendrocytes and axons, and increasing local synthesis of the major protein in the myelin sheath, myelin basic protein, through Fyn kinase-dependent signaling. This axon-oligodendrocyte signaling would promote myelination of electrically active axons to regulate neural development and function according to environmental experience.
What is myelin?

The wrapping of oligodendrocyte membrane around axons. It increases the speed of propagation of action potentials.

What is the authors’ big question?

Do action potentials stimulate myelination?

Why is this an interesting question?

If the answer is “YES”, then axon - oligo signaling promotes anatomical and functional neural development according to environmental experience!

What was previously known?

Axons can release glutamate and ATP, and oligo progenitors are sensitive to them.
Figure 1

Release of synaptic vesicles from axons promotes myelination

A. Experimental procedure

- DRG = dorsal root ganglion neurons
- BnTX = botulinum toxin, blocks transmitter release
- OPC = oligo progenitor cells

On DRG

<table>
<thead>
<tr>
<th>Day 0</th>
<th>Day 1</th>
<th>Day 5</th>
<th>Day 21</th>
</tr>
</thead>
<tbody>
<tr>
<td>+BnTX or -BnTX or +TnTX</td>
<td>Plate OPC onto DRG</td>
<td>Electrical stimulation 5 hr</td>
<td>Fix the samples</td>
</tr>
</tbody>
</table>

B.

Green = myelin basic protein
Purple = neurofilament protein

10 µm

C.

Number of myelin segments/cell

- BnTX
- +BnTX
- +TnTX

*
Figure 2  Electrical activity in axons generates calcium elevations in the processes of OPCs by releasing glutamate
Figure 4  Action potentials induce local translation of myelin basic protein by release of glutamate

Kikume-MBP-3’ UTR is a DNA construct transfected into OPCs. MBP protein is green when expressed, turns red in UV, and then newly synthesized green MBP can be easily identified.
**Conclusion:**

Action potentials in DRG neurons regulate stimulate local protein synthesis of myelin basic protein (MBP) in processes of oligodendrocytes. Thus electrical activity may be important for modifying brain circuits according to experience. Myelination continues through the first three decades of life.
Activity-Dependent Transfer of Brain-Derived Neurotrophic Factor to Postsynaptic Neurons

Keigo Kohara, Akihiko Kitamura, Mieko Morishima, Tadaharu Tsumoto*

www.sciencemag.org  SCIENCE  VOL 291  23 MARCH 2001
What are neurotrophins?

Proteins synthesized by neurons that act on other neurons, stimulating growth, differentiation and survival. E.g. BDNF

How were they thought to act?

Released by postsynaptic neurons and acting on presynaptic neurons, i.e. target-derived factors

What did the authors find out?

BDNF is transferred from presynaptic to postsynaptic neurons; this process depends on electrical activity (action potentials)
Figure 1

Nuclear injection of DNA encoding BDNF-GFP, mouse cortical neuron \textit{in vitro}

![BDNF/GFP](image1.png) ![Anti-BDNF antibody](image2.png) ![Overlay](image3.png)
Figure 2  Movement of BDNF in the axon of a living neuron

Velocity 0.3 µm/sec = 25 mm/day
Figure 3

Transfer of BDNF-GFP from presynaptic to postsynaptic neuron

Injected DNA presynaptically encoding BDNF-GFP & DsRed

Note: DsRed protein does not cross; thus BDNF transfer is specific
Figure 4

Activity-dependent transfer of BDNF-GFP

A

+TTX (tetrodotoxin)

I

Fluorescence intensity (%)

Control  Contacted

K

+TTX

Control  Contacted
Conclusion:
BDNF is transferred from presynaptic to postsynaptic neurons; this process depends on electrical activity (action potentials)
Figure 1.4  Distinctive arrangement of cytoskeletal elements in neurons (Part 3)

Neuromuscular Junctions

(H) tubulin  (I) Axon

(AChR) Receptors

(J) Basal lamina  (K) Dystrophin/scaffold protein