

Lecture 38 – Development II -- Scheiffele

Synaptogenesis

I. PNS: Neuromuscular Junction

The neuromuscular junction is a well studied model of synapse formation. Before a motor neuron contacts a skeletal muscle cell, the transmitter-containing synaptic vesicles are diffusely distributed in the motor axon and the muscle cell has receptors for the transmitter (AChRs) scattered all over its surface. Contact causes the AChRs to become highly concentrated on the crests of the postsynaptic junctional folds and the synaptic vesicles to become concentrated at presynaptic active zones directly opposite the junctional folds. This matching permits high fidelity transmission.

The pre- and postsynaptic sides are linked through the basal lamina. Regeneration experiments have shown that components of the basal lamina instruct the organization of the nerve terminal. A functional analysis of basal lamina proteins has yielded several of the key players that mediate the formation of pre- and postsynaptic specializations.

A. Postsynaptic specialization

The contact of the motor axon with the muscle cell triggers three processes that give rise to the large clusters of AChRs underneath synaptic boutons.

1. Clustering of preexisting AChRs (translocation)

Agrin released from the motor axon binds on the surface of the muscle cell to a receptor containing **MuSK**, a protein-tyrosine kinase. Through an as yet undeciphered signaling pathway, this causes the dispersed AChRs to cluster at the sites of developing boutons. Perhaps AChRs diffusing in the plane of the sarcolemma get trapped at developing clusters. **Rapsyn** binds to the cytoplasmic domain of the AChR and is essential for clustering.

Mouse knockouts lacking agrin, MuSK or rapsyn have few or no ACh clusters, supporting the above scheme. Agrin and MuSK knockouts also show excessive motor axon branch growth on the surface of the muscle, suggesting that agrin additionally is involved in retrograde signaling to the motor axon.

2. Reduction of synthesis of AChRs in extrasynaptic areas

An individual muscle cell has numerous nuclei distributed along its length. Depolarization of the muscle cell triggered by transmitter release from the motor axon causes nuclei only in extrasynaptic areas to decrease their transcription of genes coding for the subunits of the AChR by the following signaling pathway:

$$\uparrow V_m \rightarrow \uparrow [Ca^{++}]_i \rightarrow \uparrow PKC \rightarrow \downarrow \text{myogenin, MyoD} \rightarrow \downarrow \text{AChR transcription}$$

3. Increase of synthesis of AChRs in synaptic areas

Neuregulin released by the motor axon binds to the **erbB** receptor (another protein-tyrosine kinase) on the muscle cell and causes nuclei only in synaptic areas to increase

their transcription of genes coding for the subunits of the AChR by the following signaling pathway:

Neuregulin-erbB → ↑ras → ↑raf → ↑MAP kinase → phosphorylated transcription factors → ↑AChR transcription

Reduction of neuregulin in genetically engineered mouse results in NMJs with reduced mepp size.

B. Presynaptic specialization

Laminin 11 is in the basal lamina only in the synaptic cleft, not in extrasynaptic areas. The NMJ in laminin 11 knockout mouse shows diminishing transmission because the terminal Schwann cell progressively invades the synaptic cleft. Laminin 11 may prevent the entry of the Schwann cell into the cleft.

In vitro experiments suggest that **agrin** deposited in the basal lamina might signal the motor axons to stop growing and might induce the formation of presynaptic specializations.

II. CNS Synapses

Structure and function of synapses in the central nervous system is more complex and less well understood than that of neuromuscular junctions.

There are several **principle analogies** between synapse formation in the CNS and in the PNS which include:

- bi-directional signaling
- clustering of neurotransmitter receptors
- synaptic vesicles have similar components
- synapse elimination during development

However, there are also very significant structural and molecular differences in CNS synapses:

- no basal lamina but direct pre- to postsynaptic interactions
- no junctional folds but dendritic spines
- multiple innervation is common
- difference in neurotransmitters:
 - excitatory synapses commonly use glutamate
 - inhibitory synapses commonly use GABA (g-aminobutyric acid) or glycine
- different neurotransmitter receptors

Relevant reading: chapter 55 in “Principles”