LESSON 2.4 WORKBOOK

What do our neurons need to work efficiently?

We’ve learned that axons can be very long, and that most of the proteins that make up their structures are made in the cell body. We’ve also learned that proteins don’t last forever, they wear out and need to be replaced – this includes all the channels and the Na⁺/K⁺ pump that’s so important for keeping the resting membrane potential stable, as well as everything that’s needed to convert the action potential into a chemical signal to cross to the other side of the synapse. How can our neurons keep up with this relentless demand? As you can imagine getting what’s needed to where it’s needed, when it’s needed is a task that requires a highly organized transport system that delivers cargo both into and out of the axon. When this transport goes wrong, it can be a big problem. This lesson will look at axonal transport.

Axon structure underlies axonal transport

We’ve used our big toe axons in several examples, but in fact axons can be anything from a few millimeters to several meters long. The shortest axons belong to the interneurons that connect local areas within the brain and can be less than 0.1 mm; the longest axons are the big toe axons that would be several meters long if you were a giraffe. But all axons, short or long, have the same internal structure thanks to their cyto (from the word for cell) – skeleton.

The cytoskeleton is conveniently organized parallel to the length of the axon so it can provide the tracks that transport cargo to and from the presynaptic terminal just like a railroad. The axon’s cytoskeleton has three main protein components — microtubules, neurofilaments, and actin — each of which play different roles. Neurofilaments are like filler that hold the mature axon rigid and determine its diameter. Actin is important when the axon is growing in the embryo and keep it flexible and able to move. Microtubules are important for axonal transport, so we’ll focus on them here.
DEFINITIONS OF TERMS

Anterograde transport – movement of materials from cell body to axon terminals.

Minus-end – end of microtubules oriented toward cell body.

Minus-end directed motors – molecular motors that travel toward the minus-end of microtubules, and thus carry cargo from the axon terminal to the cell body.

Plus-end — end of microtubules oriented toward axon terminal.

Plus-end directed motors – molecular motors that travel toward the plus-end of microtubules, and thus carry cargo from the cell body to the axon terminal.

Retrograde transport – movement of materials from axon terminals to the cell body.

For a complete list of defined terms, see the Glossary.

Microtubules provide the tracks on which cargo is transported

Microtubules form the actual ‘tracks’ that move cargo to and from the axon terminal. How do they do this? They are hollow cylindrical polymers that are made from two different forms of the tubulin protein called alpha and beta tubulin. These two tubulin isoforms polymerize together to form a hollow tube that is polarized, meaning that one end is different from the other. The two different ends are called the plus-end and the minus-end. In axons, all microtubules are oriented in the same way, with the plus-end toward the presynaptic terminal, and the minus-end toward the cell body (Figure 22). Motors can attach to the microtubules. If they are plus-end directed motors, they will move toward the presynaptic terminal. If they are minus-end directed motors, they will move towards the cell body. (Figure 23). We’ll talk more about these motors in a minute. Cargo can attach directly to these motors and be transported from the cell body into the axon and the presynaptic terminal (anterograde transport) or from the presynaptic terminal and axon back to the cell body (retrograde transport).

How does transport work?

Scientists knew that most protein synthesis occurred in the cell body, and they deduced that there must be a system that can transport these items efficiently to where they were needed. Fortunately, major technological advances in microscopy occurred precisely at the time we began to question exactly how materials are transported in the axon. One of the previous problems had been in observing very small structures. New advances with lenses and computing allowed scientists to detect structures moving down the axons. Using the newly developed tools that allowed us to see the inside of living axons, scientists could observe the movement of materials in axons and then figure out exactly how transport works.

What kind of motors carry cargo to the axon terminal? What motors carry cargo to the cell body?

Why do neurons need such an elaborate transport system?
LESSON READING

Proteins are not transported down the axon individually. Instead the cell body organizes them into structures (organelles) like mitochondria or the spherical membrane vesicles that contain neurotransmitters.

You can watch a video showing vesicles moving down an axon online — see this unit on the student website or click the link below. In this case the particles have been tagged with a fluorescent marker that allows them to be detected with a fluorescent microscope. The upper image shows the fluorescent signal and the middle image shows the actual picture of the vesicles. In the bottom image they have been merged.

Video: Axonal Transport

We knew that cargo had to get to both the pre-synaptic terminal and back from it to the cell body. From many different kinds of experiments that used fluorescent and radioactive chemicals to tag transported proteins, we found out there are basically two different ways to transport material in the axon — fast and slow. Fast axonal transport is used to get materials where they are needed quickly — to the presynaptic terminal to be used in neurotransmission across the synapse (anterograde transport), or back to the cell body for recycling (retrograde transport). It’s fast because it uses a lot of energy, and depends on the microtubule motors we talked about before (Figure 24). In contrast, slow axonal transport is used to rebuild the axonal cytoskeleton itself — the microtubules, neurofilaments and actin polymers. Frankly, we are still unsure about how slow transport works and it is an active area of research. It’s important though because a number of diseases affect slow transport and have serious consequences.

Fast transport – anterograde and retrograde

Anterograde transport

The presynaptic terminal needs a constant supply of membrane vesicles, because as we shall see in the next unit, it packages neurotransmitters into the vesicles so it can control their release into the synapse. It (and the axon) also requires a constant supply of energy in the form of ATP, which is delivered by mitochondria. Hence vesicles and mitochondria need to be transported rapidly and on time, so the axon has developed a sophisticated mechanism that directs them where they need to be.
LESSON READING

Recall that fast transport uses microtubules and motors, and that anterograde transport (meaning going to the presynaptic terminal) uses plus-end directed motors. These motors work by attaching to and traveling along microtubules toward their plus-end which, again, is oriented towards the presynaptic terminal. So, the motors use the polarity of the microtubules as a cellular GPS. These plus-end directed motors are a family of proteins called kinesins (Figure 25). In the cell body, kinesins, which actually look exactly like legs and feet, attach to the cargo being transported down the axon. The kinesin molecule then walks down the microtubule toward the plus-end at the presynaptic terminal, carrying the cargo to its destination. Kinesin uses energy in the form of ATP to move itself and its cargo down the microtubule. In fact it hydrolyzes one molecule of ATP for each 8 nanometer (nm) step it takes down the microtubule. The vast amounts of energy needed to make this trip are supplied by the mitochondria, so its clear that mitochondria will be needed in the axon as well as the presynaptic terminal.

You can watch a short video online showing how kinesin ‘walks’ down the microtubule towards the plus end. The video was made using the actual structure of the kinesin molecule, and what is known about how its shape changes as it hydrolyzes ATP and attaches to microtubules. The cargo would attach to the two kinesin ‘arms’ that are sticking up.

Video: Kinesin Transport Protein

Retrograde transport

Rapid retrograde transport returns materials from the presynaptic terminal or the axon to the cell body for recycling. As in fast anterograde transport, cargo moves along microtubules attached to a motor protein. In this case the motor is the minus-end directed motor protein, dynein. Dynein works much the same way as kinesin, except it carries substances toward the cell body (Figure 26).

The combination of the polarity of the microtubules, and the specificity of the motors for the microtubules and their cargo, ensures that cargo gets to the right place at the right time – this is crucial if the axon terminal is going to function properly. While we’ve told a simplified story here, keep in mind that the kinesins are actual a family of proteins with more than 40 different kinesin proteins, each specific for a particular cargo or particular destination.

What motor protein is responsible for getting cargo from the cell body to the axon terminal? How does it work?

What motor protein is responsible for getting cargo to the cell body from the axon terminal? How does it work?

DEFINITIONS OF TERMS

Kinesin – plus-end directed motor that carries cargo from the cell body to the axon terminal along microtubules.

Dynein – minus-end directed motor that carries cargo from the axon terminal to the cell body along microtubules.

Retrograde transport – movement of materials from axon terminals to the cell body.

For a complete list of defined terms, see the Glossary.
Imagine you have nerve damage that affects your axonal transport systems, what symptoms would you have? Think about this question in the light of what you know about the roles of axonal transport, and how neurons function to transmit signals.

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If kinesin uses one molecule of ATP for each 8 nm step, how much ATP is used to move one synaptic vesicle from a neuronal cell body at the base of your spine to your big toe? (Assume that the distance from your spine to big toe is 3 feet (or 1 meter).