LABORATORY 10 - NEURAL TISSUE

In this course, the study of nervous tissue will be limited primarily to features of the peripheral nervous system (PNS). In virtually every slide there will be some portion of this system that should be recognized. The central nervous system (CNS) as such will be studied in the neuroscience course.

OBJECTIVES:

LIGHT MICROSCOPY: Recognize neuron and its characteristics including axon, dendrites and cell body. In any section, identify large and small bundles of peripheral nerves, their composition, and their connective tissue coverings. Recognize arrangement of neuron cell bodies into various types of ganglia.

ELECTRON MICROSCOPY: Recognize neuron cell body and its characteristics. Recognize peripheral nerve bundles and the details of the association of axons and Schwann cells and the connective tissues that are associated with nerve bundles.

ASSIGNMENT FOR TODAY'S LABORATORY

GLASS SLIDES:

SL181 (Spinal cord) Multipolar neurons
SL 44 (Unsectioned nerve fibers) Peripheral nerve fibers
SL 45A (Spinal cord) Neuron cell bodies and fibers (and SL 45B)
SL168 (Spinal cord) Myelin stained
SL 12 (Brachial plexus) Cross section of large nerve trunk
SL 46 (Sciatic nerve) Longitudinal section of large nerve
SL 16, 23, 24, 47 Peripheral nerves in various organs
SL 49 Cranial nerve ganglion
SL 50 Spinal ganglion (dorsal root ganglion)
SL 51 Autonomic ganglion from sympathetic chain
SL 53 (Colon) Parasympathetic ganglion
SL 14 (Jejunum) Parasympathetic ganglion
SL 60 (Muscle) Neuromuscular spindle

ELECTRON MICROGRAPHS - See text and atlas

Neurons J. 9-23 to 9-30, W. 7.3
Neurons and nerve fibers J. 9-5; W. 7.5 to 7.7, 7.18

POSTED ELECTRON MICROGRAPHS

#16 Peripheral Nerve
Lab 10 Posted EMs

HISTOLOGY IMAGE REVIEW - available on computers in HSL

Chapter 2. Muscle Tissue
Frames: 429-501 (Disregard references that are specifically for CNS.)

SUPPLEMENTARY ELECTRON MICROGRAPHS

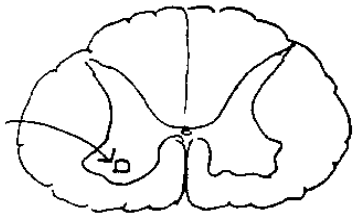
Rhodin, J. A.G., An Atlas of Histology
Copies of this text are on reserve in the HSL.
Nervous tissue, p. 156 – 176

I. NEURON (general features). (J. pp. 163-165).

A. Nerve cell body - The neurons we will examine are located in the spinal cord as illustrated below.

1. SL 181 - Spinal cord (scan, low, med) - cresyl violet. (J. 9-21; W. 1.7c). Study this slide for the characteristic features of a typical multipolar neuron cell body (the largest cells in this tissue). The neurons are large and each has a large nucleus (in green circle and prominent nucleolus (blue arrow), Nissl substance (RER) (green arrow), dendrites, axon hillock and axon (red arrow high, structures identified). Nissl substance is absent from the axon hillock. Since there is only one axon for each cell and it is stained palely, the axon hillock is difficult to identify. In order to find an axon hillock and axon, you will have to look at several cells. In addition to the neuron cell body in this region observe numerous small nuclei surrounded by little or no visible cytoplasm. These are nuclei of several types of supportive cells of the nervous system that are called collectively, neuroglia. The neuroglia of the central nervous system will be considered in the Neurosciences course.

2. Electron microscope (J. 9-5, diagram; W. 7.3) Note organelles.



B. Nerve fibers.

1. SL 44 - This is a preparation of unsectioned nerve fibers that have been teased apart, fixed and stained with osmic acid (osmium tetroxide) (W. 7.7). The black stained material is myelin that may have shrunken during processing. Observe the axon (also called axis cylinder) surrounded by the myelin sheath and nodes of Ranvier (red arrow) found between the Schwann cells* that lie in tandem along an axon. Do not confuse folds in the myelin or breaks in the fiber with nodes of Ranvier; locate nodes specifically. Although the nodes lack myelin, the plasma membranes of adjacent Schwann cells are in contact. Fibers of different sizes surrounded by varying amounts of myelin can be detected.

*In some texts the older term neurilemma may be used to describe the Schwann cell sheath.

2. SL 45A and SL 45B (This slide is included only in even numbered desks. Virtual slides A and B are similar, but took up the stain differently) The spinal cord was sectioned and then stained with silver (W. 1.21) (scan, med 1, med 2, high). Note neuron cell bodies in the "gray matter" (diagram and image above). In addition, there are dark fibers oriented in different directions scattered throughout the gray matter. These fibers are both axons and dendrites. Neurofibrils, cytoskeletal elements within the cell body (blue circle) and processes (red arrows) react with silver staining. (Although all cells contain these processes not every cell will stain when using this technique. It is not known why only certain cells are stained.)

3. SL 168 - Spinal cord, myelin stain. (scan, high) Compare to SL 45A and SL 45B. In this section the myelin sheath has been stained rather than neurofibrils. Tracts of nerves passing up and down the spinal cord are located in the periphery (white matter). These nerve fibers are cut in cross section so that the myelin sheath appears as a ring around each myelinated axon. Together this slide and SL 45A and SL 45B demonstrate the main histological features of the neuron and illustrate some of the basic techniques used in the anatomic study of the nervous system. Nerve endings and specialized receptors are not studied at this time.

II. PERIPHERAL NERVES AND THEIR ASSOCIATED CELLS AND TISSUE

The following slides demonstrate the variation in structure and size of peripheral nerve bundles as they are combined to form the various nerves of the body.

A. SL 12 - Brachial plexus. Large nerves (nerve trunk) in this section (J. 9-32, 9-34; W. 7.13, 7.14, 7.15, 7.16) are cut primarily in cross section and stained with H and E. Identify the following structures in several locations:

1. The connective tissue associated with peripheral nerves has distinct regions and characteristics. Recognize and distinguish between the epineurium, perineurium and endoneurium (scan, low, high; blue arrows, epineurium; red arrows, perineurium; green arrows, endoneurium).

2. Blood vessels within these sheaths

3. Within the individual nerve fibers (seen mainly in cross section) identify the following:
   a. Myelin sheath - the lipid content of myelin is dissolved out of the section because organic solvents are used to prepare the tissue. However, the residual proteins often form an eosinophilic network in the site where myelin was present. This artifact of tissue preparation is called "neurokeratin" and is typical of H & E sections (med, high, doughnut between blue circles).
   b. Axon (axis cylinder) observed as a central structure within smaller blue circle. In some nerve fibers the axon may be absent or stained very lightly.
   c. Unmyelinated fibers are present, but cannot be distinguished.

4. SL 46 - Sciatic nerve - longitudinal section (low, high)
   H and E Note all features listed under slides 44 (previous page) and 12.

B. Smaller nerves found in the connective tissues around or within the walls of organs (W. 7.19). Examination of the following selection of slides will provide you with practice in identifying nerves:

SL 23 (scan, high), SL 24, SL 47 - Locate nerves (e.g., blue arrows, and SL 24), identify as many features as possible, smallest nerves have only perineurium (epineurium by some authors) and endoneurium.

C. Electron microscope - (J. 9-29, 9-30, 9-33; W. 7.5 to 7.7, 7.18). Study enough electron micrographs from other texts or atlases to be able to identify elements of a nerve and distinguish between myelinated and nonmyelinated fibers.

III. GANGLIA (nerve cell bodies outside the central nervous system)

A. Cranial or spinal (dorsal root) ganglia with unipolar ganglion cells.

1. SL 49 - (J. 9-3, 4; W. 7.20). Semilunar ganglion of the fifth cranial nerve. H and E. Analyze the structure of this cranial ganglion and find the following:
   a. Outer connective tissue capsule
   b. Nerve bundles
   c. Neuron cell bodies (within blue circles)
   d. Satellite cells (amphicytes, capsule cells, glia)

2. SL 50 - Spinal ganglion (dorsal root ganglion). H and E. Observe the same organization that is found in the cranial ganglia (low, med).

B. Bipolar neurons are found in specific sensory areas and will be studied with the organs concerned.

C. Autonomic ganglia (sympathetic and parasympathetic), small multipolar cells.

1. SL 51, Vertebral ganglion (sympathetic trunk). (W. 7.18).
   a. The multipolarity of the neurons may be difficult to determine.

   b. Some cells may be binucleate and/or have clumps of yellow pigment in them. The nuclei are eccentrically located within the cells.

2. SL 53, SL 14 (W. 7.21) - An example of groups of cells of parasympathetic ganglia lying in an organ. Note two layers of smooth muscle at the periphery of this ring-shaped organ (colon) (scan). Locate neurons (blue arrows) lying between the muscle layers and also isolated neurons in the wide layer of loose irregular connective tissue.

IV. NERVE TERMINATIONS

In your textbook study the typical structure of the synapse. Pay close attention to the ultrastructure of the synapse. Also study the structure of the motor end plate and compare it to the synapses between neurons. (J. 9-7 to 9-10, 10-18; W. 7.8-7.12).

A. (Optional) - SL 60, Neuromuscular spindle, (within blue circle) (J. p. 204 and Figure 10-19; W. 7.33) - As a review, look at this cross section of skeletal muscle. Locate the neuromuscular spindle. This is the neuromuscular device within a muscle that "senses" the length and sets the tone of the muscle reflexively. Read about its organization in the text. Briefly, what roles do the neuromuscular spindle and motor end plate play in the contraction of a muscle?

Other sensory endings will be observed in later sections.

REVIEW

1. Contrast the ultrastructure of myelinated and nonmyelinated axons.

2. What happens to the distal part of a peripheral axon when it is damaged or cut? What evidence of this damage occurs in the cell body?

3. Briefly compare the structure of the connective tissue sheaths of a muscle with those of a peripheral nerve.

4. Compare the terminations of somatic and visceral efferent fibers.

OBJECTIVES FOR LABORATORY 10: NEURAL TISSUE

1. Using the light microscope or digital slides, identify:

    - In spinal cord gray matter (note several stains were used)
        - Nerve cell body
            - Nucleus
                - Nucleolus
            - Nissl substance (RER)
            - Axon
                - Axon hillock
            - Dendrites
    - In spinal cord white matter (note several stains were used)
        - Axons and dendrites (indistinguishable)
    - In prepared nerve specimen
        - Myelinated axon
        - Node of Ranvier
    - Peripheral nerve (in any orientation)
        - Connective tissue of peripheral nerve
            - Epineurium
            - Perineurium
                - Fascicle
            - Endoderm
        - Axon
        - Myelin
        - Schwann cell nuclei
    - Ganglia
        - Neuronal cell bodies
        - Satellite cells
        - Bundles of axons and dendrites

2. On electron micrographs, identify:

    - Peripheral nerve
        - Connective tissue
            - Epineurium (magnification may be too high to see this layer)
            - Perineurium
                - Fascicle
            - Endoneurium
    - Myelin
    - Schwann cell
        - External lamina
    - Axon
        - Myelinated
        - Ensheathed (unmyelinated)