ANAT 2160/BIOL 3430 Nervous tissue and cartilage laboratory module

1) Nervous tissue

Objectives:
1) understand the structure of neuronal somata (cell bodies) and cytoplasmic processes
2) identify somata and processes of neurons in other tissues in the body
3) identify cell types that support neurons in the central and peripheral nervous systems
4) understand the relationship between peripheral nerves and their connective tissue

Overview:
Neurons are the basic cellular building blocks of the nervous system, designed to communicate information from one part of the body to others. Nervous tissue, in addition to comprising the brain and spinal cord, is also a component of every other organ and system in the body. Neurons are supported by glia in the central nervous system and by satellite cells and Schwann cells in the periphery.

A) Neurons.

Neurons are among the largest cells in the body. Not only do they have a relatively large cell body (the soma), the volume making up the long cytoplasmic extensions of neurons (axons, dendrites) is also large. For instance, in an adult motor neuron with its soma in the spinal cord and an axon that innervates a toe muscle, that axon will be more than a meter long. All neurons are entirely surrounded by supporting cells, so there are many more support cells than there are neurons in the nervous system. In the slides below you will see some examples of neurons, their support cells and the connective tissue structure of peripheral nerves.

227, spinal cord, whole mount. Neurons, with their extensive processes, can be difficult to visualize in three dimensions in thin sections that are cut from nervous tissue. This sample was prepared from a small piece of the spinal cord that was flattened onto the glass slide by smearing the neural tissue. Because the tissue was not cut, the processes of some neurons were preserved and these can be followed for some distance from the soma. The slide was stained with a Nissl stain to show up the clumps of ribosomes (Nissl bodies, dark blue-purple granules) of the RER in the soma and the roots of the dendrites and axon. There is no RER in neuronal processes so Nissl bodies are not present there. The somata on this slide are those of large multipolar neurons. Some of these somata will have pale-staining nuclei with dark nucleoli. The many small, dark-staining nuclei surrounding the somata and processes of neurons on this slide are those of glia.

23, spinal cord section. This slide was made by cutting a thin cross-section from the spinal cord. Look for the somata of neurons in the ventral horn of the grey matter (the butterfly-shaped region in the middle of the section). Since the somata of neurons in the spinal cord are much larger than the thickness of the section, most of the processes of these neurons will be cut away leaving only fragments of the cell bodies and the roots of the processes. Look for neuronal somata that contain nuclei. Compare this view with the view in **slide 227** of the same neurons.

B) Glia.

CNS slide 61. This slide is found in the "Neuroanatomy" slide box; there is one of these boxes for every two lockers on each bench. The section is a piece of cerebral cortex from the brain that has been stained with a special stain, the Golgi stain, that causes approximately one in every hundred cells on the slide to be stained black. You can see the lightly stained nuclei of many other cells in the background that did not stain black with this technique. There will be many black-stained cellular fragments on this slide. Start from the outside edge of the tissue and move inward as you examine this section. Identify pyramidal neurons first (these have triangular somata with long slender processes and are plentiful), then try to find astrocytes. These cells will be larger than the pyramidal cells, with short, compact processes radiating in all directions from the soma. You will also see some blood vessels stained black (short curved lines or cylinders) and a great deal of artefactual staining. This slide is challenging and will take you some time to work through.

C) Peripheral nerve tissue

27, dorsal root ganglion. Ganglia are collections of neuronal somata in the body but outside the central nervous system; they are surrounded by a capsule of connective tissue, mostly Type I collagen fibres. The neurons in this ganglion are pseudounipolar (although you will not be able to see whole soma because the section is too thin) and are sensory, with dendrites bringing information from receptors in the body and axons that project into the spinal cord. Neuronal somata are the largest cells in the section, appearing round or oval with large nuclei; these cells tend to be stained more darkly than the surrounding tissue. Examine the slide by eye to identify an enlarged region of tissue; this is the ganglion, where many neuronal somata are located; the thinner portion of the section is a nerve connecting to the ganglion. Each neuron is surrounded by a layer of satellite cells (the small nuclei) which are functionally equivalent to astrocytes in the central nervous system. Note the large amount of connective tissue not only in the capsule, but within the ganglion and the nerve. What is one purpose of this extensive connective tissue?

228, single axons, teased. To make this slide, a short segment of a large nerve was dissected with fine pins to separate out strands containing only a few axons or an individual axon. These were then stained with osmium, a heavy metal that stains lipid brown or black. The osmium was thus taken up by the myelin in the Schwann cell sheaths around each axon. Examine the slide to find thin dark strands, then look at these at high magnification to see the stained myelin sheaths. You will not be able to see the axons within the myelin since these will have deteriorated during the staining process. Where the cytoplasm of one Schwann cell ends and another begins along the length of an axon, you will see a break in the myelin; this is a node of Ranvier, and conduction along the axon jumps quickly from node to node.

99, 224 peripheral nerve cross and longitudinal sections. These slides have two pieces of tissue each, taken from whole peripheral nerves. In both slides, one section was cut along the long axis of the nerve and the other was cut in cross-section. The cross-section of the nerve contains all levels of connective tissue; identify the epineurium, the

perineurium, fascicles, and the endoneurium. Can you see any small blood vessels in the endoneurium? What is special about the perineurium? The axons themselves have deteriorated due to the histological procedure used to prepare the slide, but you can see the small, round profiles where the myelin in the Schwann cells surrounding the axons has taken up the stain. Some of the axons may have been cut obliquely in this piece of tissue, so their profiles will appear oval. In the longitudinal section, many of the axons within the fascicles of this nerve follow a wavy path. Identify fascicles, epineurium, perineurium and endoneurium. In the longitudinal section on slide 99. try to find some examples of nodes of Ranvier.

2) Cartilage

Objectives
1) Identify the cartilage matrix, and distinguish chondrocytes in the matrix.
2) Differentiate the perichondrium (where present) from the cartilage matrix proper and identify cell types present in the perichondrium.
3) Recognize the visible histological characteristics of the three types of cartilage present in the body.

Overview.
This special connective tissue provides flexible support and reinforcement for softer tissues in the body (i.e. reinforces larger airways in the respiratory system, supports ear and nose soft tissue, cushions and lubricates joints). The type of cartilage is determined by the type of fibres in the matrix, but all cartilage has the same type of chondrocyte, in holes (lacunae) in the matrix. Are there blood vessels in cartilage?

A) Hyaline cartilage: characterized by the uniform appearance of the matrix, which often stains bluish-purple. What type of fibre is present in hyaline cartilage? Only the shrunken remains of chondrocytes are present in the lacunae. In life, these cells have lipid and glycogen droplets in their cytoplasm. The cells would fill all the space available in the lacune, but cell shape has been distorted by processing the tissue to make the slide.
56, trachea. This specimen is a transverse section through the trachea of a pig, and contains sections of one or more of the circumferentially oriented rings of hyaline cartilage that help to keep the airway open. Note the lack of features in the matrix between the lacunae. There is a perichondrium around this cartilage; what cell and fibre types are present here?
1**29, developing finger bones**. This slide shows a longitudinal section of the phalanges (finger bones) of a developing digit from a fetus. The long bones are dumb-bell shaped with knobs of hyaline cartilage at each end. As the fetus develops this cartilage will be replaced with bone, leaving just a thin cartilagenous layer covering the ends of the bones that move against each other in the joints. These layers of cartilage form the articulating surfaces. Note that, even at this early developmental stage, the cartilage in the joints does not have any perichondrium, a condition maintained throughout life for joint cartilage. What are the functions of hyaline cartilage in this type of joint?

B) Elastic cartilage: this type of cartilage is found where tissue deformation and flexibility without damage are required, such as in the pinna of the ear, the nose and the epiglottis. There are some Type I collagen fibres present in the matrix of this cartilage, but the majority of fibres are elastin.
28, 29, epiglottis. These slides were made from sections of tissue cut longitudinally from the epiglottis; the free tip of the valve is at the thin end of the sections. **Slide 28** was stained to show the nuclei and cytoplasm of cells (purple), and Type I collagen fibres (pink); the elastin fibres are not stained here. On this slide, identify the cartilagenous core of the epiglottis, the epithelium covering both sides of the section, and the perichondrium. What cells can you see in the perichondrial region? Can you identify

chondrocytes in the matrix? **Slide 29** was stained to demonstrate the elastic fibres (dark stain) in the cartilage matrix. Are these fibres present with the same density throughout the matrix?

C) Fibrocartilage. The predominant fibre type in the matrix of this cartilage is Type I collagen, although there is also some Type II collagen present (unstained).
18, knee meniscus cartilage. The meniscus cartilage is a wedge-shaped piece of tissue within the knee joint that acts to cushion impact loading and give lateral stability to this joint. The chondrocytes may be difficult to see, owing to the intensely stained and numerous collagen fibres. Look for the nuclei of these cells at high magnification. How are the collagen fibres oriented in this specimen?