We can all appreciate the beauty of the stars, planets and constellations on a clear night or by viewing the dazzling images taken with the Hubble Telescope. But in this short course, I hope to convince you that the cells of the brain rival the celestial bodies in their beauty as individual elements or collectively. However, unlike the stars overhead, the stars of the brain have only been visible since about the last quarter of the 1800s. And even then, only a privileged few have viewed them. The reason for this is that brain cells are transparent and require special anatomical methods to be seen. The chronological development of those methods, from the late 1800s to the present date, directly impacted our understanding of the normal and pathological functioning of the brain and so, that chronology provides a convenient means of organizing the material in this lecture series.

**Week 1**

**Brain Basics: Development and important features of the brain and spinal cord (CNS)**

- **Brain Tube** – In embryonic development, cells of the brain and spinal cord are born and then migrate away from the walls of a neural tube to mature. The spinal cord forms around the "tail" end of the tube. The "nose" end of the tube expands into chambers called ventricles and the brain forms around those ventricles. The neural tube is filled with cerebrospinal fluid (CSF). With the exception of a unique region in the temporal lobe, no new neurons are generated in the adult brain.

- **Brain Bag** - The brain and spinal cord are enclosed by membranes (*meninges*). The tough, outer membrane (dura) contains pain receptors and large veins and arteries. The thin, inner membrane is "shrink-wrapped" to the surface of the brain. The intermediate membrane forms a CSF-filled (subarachnoid) space with the inner membrane. Vessels supplying the brain travel in this space.

- **Brain Float** – Because the brain and spinal cord are completely immersed in CSF they become buoyant and buffered from impact. The external weight of the brain is about 1400 gms (3 lbs) but in the intact skull, the calculated weight is about 25-50 gms.

- **Brain Sewer** – CSF is constantly generated into the brain ventricles and at certain passageways it passes into subarachnoid space surrounding the brain. From those spaces, one-way valves allow CSF to flow into a large vein located between the brain hemispheres. So, the brain is constantly being flushed with CSF.

- **Brain Mush** – The structure of the brain depends entirely on being confined by the skull and meninges. Without these, the brain collapses. So, brains must be pickled (fixed) and sometimes embedded in a supportive matrix so that they can be studied.

- **Brain Components** - The two major cell classes of the brain are neurons and glia in about a 1:1 ratio. Glia are structural support and maintenance cells. They are smaller than neurons and they continue to be generated in the adult brain.

**Neuron Parts**

Neurons have 3 parts: **dendrites** and an **axon** for receiving and transmitting neural signals, respectively, and a **cell body**. The cell body contains DNA and RNA and the structures for protein synthesis and packaging for the whole cell. The outline below follows the development of the stains and imaging techniques for studying each part of the neuron.

**Gray matter** – Collections of nerve cell bodies (commonly referred to as nerve cells or neurons) and glia

- **The cerebral cortex** – Localization of function in the frontal lobe
  - John Hughlings Jackson’s observations of epileptic seizures (1864) in humans
  - Fritsch and Hitzig’s electrical stimulation (1870) of the dog motor cortex
  - Betz’s discovery (1874) of gigantic neurons in the motor cortex of dog and other mammals
  - Nissl’s stain for nerve cell bodies (1884) and Brodmann’s (1907) cytoarchitectural maps of cerebral cortical areas
  - Nissl’s studies of the nerve cell responses to injury
  - Nissl stain of other gray matter areas – LGN thalamic nucleus (visual relay nucleus), hippocampus, striate cortex (primary visual cortex), cerebellar cortex
Week 2
White matter – Collections of axons
- Weigert stain (1884) for fatty (myelin) sheath – demyelinating diseases
- Klüver-Barrera (1954) stain for cells and fibers
- Bielschowsky stain (1902) for normal neurofibrils within axons – neurofibrillary tangles, a characteristic of Alzheimer's disease
Dendrites (+ "nude" axons) – unmyelinated processes of neurons that display characteristic forms
- Golgi & Ramón y Cajal (1906 Nobelists) Masters of the “capricious” Golgi stain
- Ramón y Cajal - fundamentals of neural circuits, how the brain works
- Astrocyte glia: Cajal's gold sublimate stain

Week 3
Neuronal Connectivity – Experimental neuroanatomy and distributive processing
- Marchi osmium stain for degenerating myelinated fibers (1880)
- Nauta reduced silver stain (and variants) for degenerating axons (1954 – 1969)
- Intra-axonal tracing – Anterograde (cell body to axon terminal), retrograde, both
- Autoradiography (1972) – Anterograde labeling of axons using radioactive tracers detected with photographic methods and darkfield microscopy
- Axonal transport of plant, cholera toxin, and viral protein markers (1984 – present)
- Fluorescent dye markers (1986 – present)

Week 4
Ultrastructure of neurons and glia
- Electron microscopy and the *Fine Structure of the Nervous System*, Peters, Palay and Webster (1960)
- Ultrastructural counterparts of neuronal features seen (or not seen) with the light microscope
- Synapse
- Nissl stained structures – rough ER (Nissl bodies), nucleolus, nucleus
- Neurofibril counterparts – neuro(micro)tubules, neurofilaments
- Myelin sheath
- Dendritic spines
- Unmyelinated axons

Immunohistochemistry - Confocal laser scanning microscopy, antibodies and fluorescent dyes (1990 – present)
- Confocal laser scanning microscopy uses a focused laser beam to scan back and forth across a brain specimen at incremental depths. All scans of the laser beam at one depth are stored as one optical section, the beam is automatically refocused at the next depth and the process is repeated through the thickness of the item of interest. Multiple laser beams (3-5) of different wavelengths excite corresponding fluorescent dyes that are bound to different antibodies. Antibodies, in turn, bind to selected components of cells in the specimen. Software is used to view optically sectioned images individually or summed and reconstructed to provide images of labeled cells in all dimensions.
- New cells (cytogenesis) in the brains of adults
- Cell identities (phenotypes)–putative neurons and progenitor cells in the cerebral cortex, neurons of the hippocampal formation,
- Dynamic processes of development and cell fates
Overview