Immunohistochemical Characterization of Multiple Sclerosis Plaques in Human Brain

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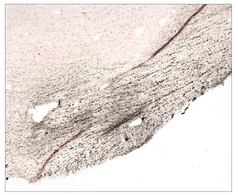
ABSTRACT

Multiple Sclerosis (MS) is a demyelinating disease with a complex pathological profile that includes myelin degeneration, neuronal damage, and immune cell infiltration in the areas containing plaques. We evaluated the pathology associated with MS in the brain of a 39 year old female whose cause of death was unrelated to the disease. In acute plaques the amino cupric silver method (de Olmos) revealed a dense core of degenerating nerve cells and fibers. Chronic lesions had little staining of cells or fibers and were devoid of staining by the Nissl counterstain, Neutral Red. Another silver stain, the silver nucleolar stain (AgNOR) was developed to reveal the nucleolar organizing regions in cancerous cells. We utilized the stain here to reveal the differences in interior cellularity between acute and chronic plaques. This is useful in getting accurate counts of the cell populations present in brain regions undergoing demyelination, and has proven to be a useful tool for stereological purposes. Weil-Myelin staining revealed roughly spherical plaques devoid of myelin staining. Nissl staining with Thionine distinguished acute and chronic lesions. Acute lesions appeared to be surrounded by a dense band of cells while the interior of the plaque had a normal distribution of cells. In chronic lesions the core was much lighter suggesting a loss of cells. The Perl's iron stain revealed a paucity of staining in acute lesions. Chronic lesions were surrounded by iron positive cells, some of which appeared to be phagocytic and filled with debris. Iba-1 immunoreactivity in acute plaques was observed both in the center of the plaque and in a dense ring of immunoreactive microglia surrounding the plaque. In chronic lesions the central immunoreactivity was diminished, but the ring of cells surrounding the plaque appeared thicker and more dense. Staining of near adjacent sets of serial sections reveals the chemoarchitechtural differences between acute and chronic states in MS lesions.

DEGENERATION

Axon Degeneration



Pons, AmCuAg, 4x

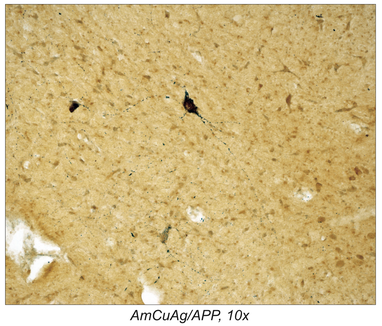
Optic Nerve, AmCuAg,10x

Positive staining in the optic nerve (right) and pons (left) are indicative of the vision and motor control symptoms associated with MS.

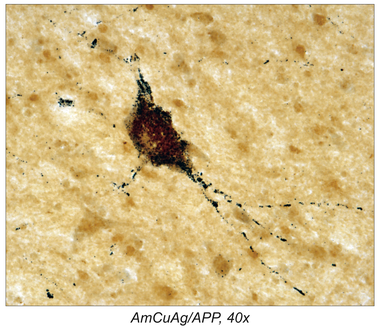
Acute lesion, AmCuAg, 20x

Degenerative profiles are shown here to be present in an acute MS lesion.

Cell Body Degeneration



AmCuAg/APP, 10x



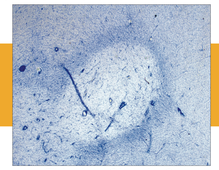
AmCuAg/APP, 40x

With this double stain, cell bodies and axons undergoing degeneration are stained black with the Amino Cupric Silver stain. Beta Amyloid precursor protein is stained brown within the cell body. The transporter mechanism that normally allows APP to be transported to the axons is blocked, leading to degeneration in the axons and a pooling of APP in the cell body.

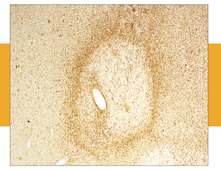
COMPARISONS

Chronic vs. Acute Lesions

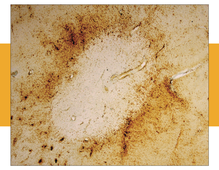
Chronic



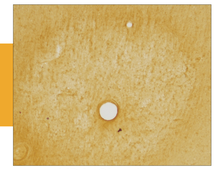
Thionine, Chronic, 2x



Iba1, Chronic, 2x

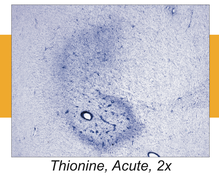


Perls, Chronic, 2x

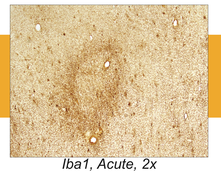


MDA, Chronic, 2x

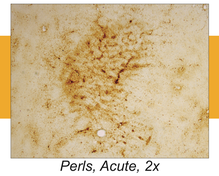
Acute



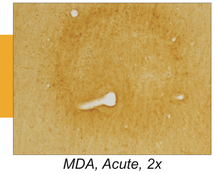
Thionine, Acute, 2x



Iba1, Acute, 2x



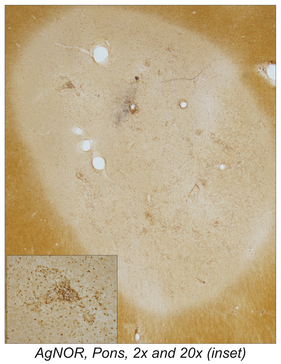
Perls, Acute, 2x



MDA, Acute, 2x

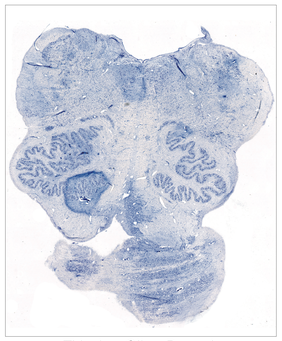
There are some basic differences in the pathophysiology of MS lesions in the chronic versus acute state. Acute lesions tend to exhibit a higher cellularity in the interior of the lesion, while chronic lesions show a marked reduction of cellularity. Both types of lesions are shown to posses a distinctive "cuffing" of cells around their perimeter. Histological methods have shown these cells to include astrocytes, microglia, and macrophages.

Pons and Inferior Olivary Nucleus



AgNOR, Pons, 2x and 20x (inset)

AmCuAg; Olive, Pons, 4x



Thionine; Olive, Pons, 4x

Weil-Myelin; Olive, Pons, 4x

Pons and Olivary Function

Olive: Fibers leaving the inferior olive project to the cerebellum, playing a role in muscle coordination and movement. Loss of myelin or cell bodies in the inferior olive has been shown to have a negative impact on completing detailed or technical motor skills.

Pons: The Pons, meaning "bridge" in Latin, connects the brainstem and medulla to the brain. Some axonal pathways that pass through pons are responsible for bladder control, equilibrium, eye movement, facial sensation, and posture. Loss of these pathways can result in some traditional MS symptoms such as enuresis, vertigo, ataxia, nystagmus, and muscle spasms.

By comparing near adjacent sections with different stains, one can get a general histological picture of the damage associated with MS. Cell bodies and axons undergoing degeneration are revealed with the Amino Cupric Silver stain. The dense "cuffing" and gliosis that occurs in MS lesions are shown in the Thionine Nissl stain. The Weil-Myelin stain depicts lesioned areas that have sustained damage to the myelin sheath. The silver nucleolar stain (AgNOR) is useful for getting a cell count both in the interior and around the edge of the lesion.

CONCLUSIONS

Anatomical comparison of near adjacent tissue sections with different IHC and classical stains is important in determining the progression and composition of disease states. In a general comparison of chronic and acute MS lesions some comparisons can be made as seen in the "Lesions" portion of this poster. The lack of cellularity and myelin in chronic lesions leads one to believe that lesions in this state are perhaps beyond immune repair. The density of staining in acute lesions might suggest that the immune response could be stopped or reversed, leading to remyelination and cellular repair. Further investigation into autoimmune components of MS may point to possible new pathways into the remediation of MS and its symptoms.