

PROPATH

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Immunohistochemistry

Ubiquitin Immunohistochemistry in Alzheimer's Disease

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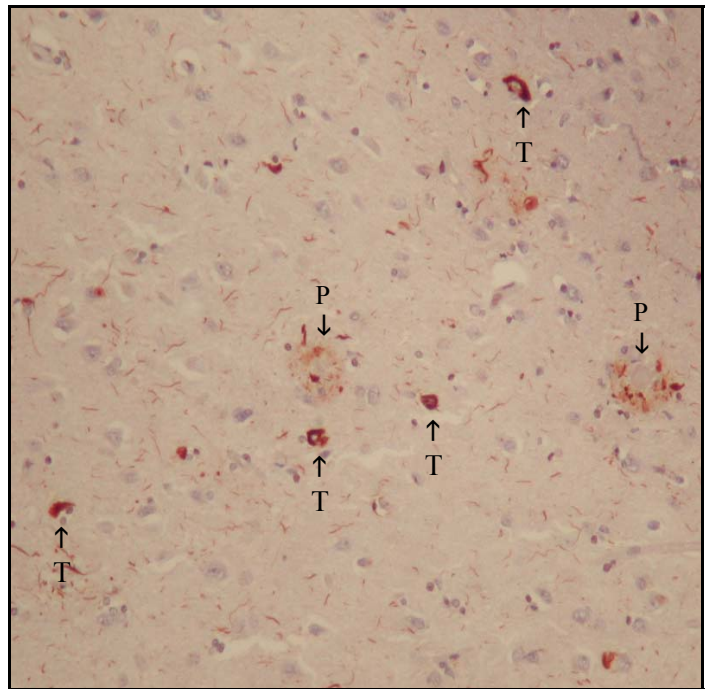
by Rodney T. Miller, M.D., Director of Immunohistochemistry

Alzheimer's disease is a devastating illness that exacts a tremendous emotional and economic toll on its victims and their families. Because some cases of Alzheimer's disease have been found to have genetic predispositions, the accurate diagnosis of Alzheimer's disease in a post-mortem brain specimen can be of great importance to family members of the victim.

For community hospital pathologists (or immunohistochemists like myself) who do not have extensive training in neuropathology (and forgot what little they learned during residency training), rendering a confident diagnosis of Alzheimer's disease can be challenging for several reasons. In addition to unfamiliarity with normal and abnormal microscopic anatomy of brain tissue, the pathologist may not have ready access to standard special stains that have been classically used in the examination of brains from suspected Alzheimer's patients. Some of these standard special stains employed to highlight the neurofibrillary tangles and neuritic plaques (such as the Bielschowsky stain) involve silver impregnation techniques that can be technically quite demanding, and as such are often not readily available in many community hospital settings.

Fortunately, a paper published in 2000 by Chu and associates (Reference 1) establishes the utility of immunohistochemical staining for ubiquitin as an extremely useful, simple, and economical tool for assisting in the accurate recognition of Alzheimer's disease, even in the hands of inexperienced trainee pathologists (and presumably also by neuropathologically challenged immunohistochemists and community hospital pathologists).

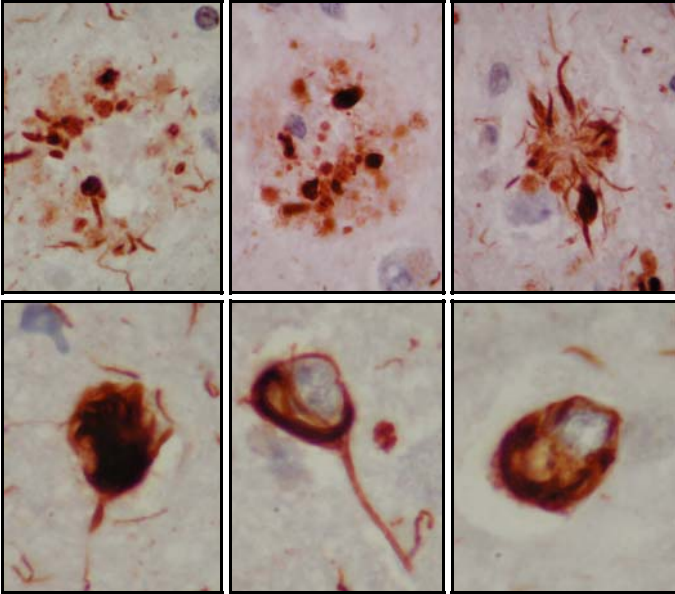
Ubiquitin is a protein that is present in all cells of the body, and it performs an essential function by tagging for destruction damaged proteins or other proteins that are no longer needed by the cell. Ubiquitin has the ability to form covalent bonds with these "condemned" proteins, and the resulting ubiquitin-protein conjugates are usually



This low power photograph of ubiquitin immunostained brain from a patient with Alzheimer's disease allows the neuritic plaques (P) and neurofibrillary tangles (T) to be readily recognized.

highly unstable and rapidly degraded by ubiquitin-dependent proteases that are associated with proteosomes. Proteosomes are large barrel-shaped enzyme complexes that take in ubiquitin-tagged proteins in one end, and digest the tagged proteins into smaller peptides or single amino acids which are released at the opposite end, so that they are available as building blocks needed for production of new proteins by the cells.

In Alzheimer's disease, ubiquitin accumulates in both neuritic plaques and neurofibrillary tangles, making it an excellent target for immunohistochemical localization. In their study, Chu et al studied brain tissue from 16 patients who demonstrated the full range of neuropathologic changes in Alzheimer's disease, including some non-demented age-matched controls. The cases were immu-



Ubiquitin stains highlight neuritic plaques (top row, 400X) and neurofibrillary tangles (bottom row, 1000X) in Alzheimer's brain.

nostained for ubiquitin, and were evaluated in a blinded fashion by 4 different reviewers, including an experienced neuropathologist, a neuropathology fellow, a senior AP/CP pathology resident, and a first-year pathology resident with brief exposure to neuropathology on the autopsy service. The cases were then categorized using guidelines from the Consortium to Establish a Registry for Alzheimer's Disease (CERAD), which separates cases into the categories of "Definite Alzheimer's Disease, Probable Alzheimer's Disease, Possible Alzheimer's Disease, and Non-Alzheimer's Disease".

The ubiquitin immunostains highlighted plaques and tangles extremely well, and allowed all observers (including those with little experience in neuropathology) to accurately categorize the cases. The overall sensitivity of the ubiquitin immunostains was 92.5%, with a specificity of 97%, positive predictive value of 98%, and negative predictive value of 89%.

The investigators also found that ubiquitin highlighted Lewy bodies, making them far easier to identify than with H&E and standard special stains. Obviously this finding indicates that ubiquitin immunostains are also useful in the diagnosis of Parkinson's disease, which is reported to coexist in about 20% of patient's with pathologically confirmed Alzheimer's Disease.

Ubiquitin is now available in the ProPath Immunohistochemistry Laboratory to assist in the accurate diagnosis

of potential Alzheimer's Disease and Parkinson's/Lewy Body neuropathology. (With the benefit of this simple immunostain, it may not take as many "brains" to diagnose Alzheimer's Disease as in the past!)

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