H4 Usability of Immunohistochemistry in Forensic Pathology

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After attending this presentation, attendees will better understand the usability of Immunohistochemistry (IHC) in forensic pathology.

This presentation will impact the forensic science community by providing information concerning how decomposition and the time between death and the autopsy affect the usability of diagnostic IHC in forensic samples.

IHC is an important diagnostic tool in surgical pathology but has limited use in forensic pathology. It is the general opinion that proteins degrade rapidly in devitalized tissues, decreasing the usefulness of IHC. The purpose of this study was to examine how the time since death affects the ability to carry out IHC analyses on forensic tissue samples.

Cases for this study were selected on the basis of the elapsed time from death to autopsy. The cases were allocated to one of the four groups (A to D) defined as: (1) Group A included cases autopsied shortly after death (one to three days) with no macroscopic signs of body decay; (2) Group B cases were autopsied three to seven days after death with signs of body decomposition such as green discoloration of the skin, marbling of the vasculature, swelling, gaseous distention, and skin slippage; (3) Group C cases were autopsied 7-14 days after death with marked swelling, black discoloration of the skin, moist and gas-ridden soft tissue, and some skeletonization and mummification of the bodies; and, (4) Group D were autopsied more than two weeks after death and the bodies typically displayed skeletonization and mummification. All the bodies (groups A-D) were found inside buildings, which in Denmark are normally heated to room temperature.

Formalin-fixed paraffin-embedded tissue samples were collected from 37 bodies and consisted of: (1) 37 samples of liver tissue — group A (10), B (10), C (8), and D (9); (2) 35 samples of lung tissue — group A (10), B (8), C (9), and D (8); (3) 16 samples of brain tissue — group A (10), B (3), and C (3); and, (4) 35 samples of muscle tissue — group A (10), B (7), C (10), and D (8). Tissue Microarrays (TMA) were constructed and IHCs were performed using an automated stainer.

The quality of TMAs and the degree of histologic changes following autolysis were evaluated using light microscopy of hematoxylin-eosin stained sections. The degree of autolysis of lung and liver samples were scored from grade 1 to grade 5 according to the degree of nuclear destruction: normal nucleus (grade 1); karryolysis and pyknosis (grade 2); karyorrhexis (grade 3); no nucleus visible (grade 4); and, no cell visible (grade 5).

The sections were stained with anti-KL1, anti-S100, anti-vimentin, and anti-CD45 antibodies, which are commonly used as a primary antibody panel in cancer diagnostic. The evaluating scoring protocol was positive (moderate or strong nuclear and cytoplasmic staining of more than 90% of cells) and negative (no staining or very weak staining in less than 10% of cells).

A good correlation between the postmortem interval and the degree of histological changes was found in all tissue samples (lung, liver, brain, and skeletal muscle). Strong positive staining of bile duct epithelium using anti-KL1 antibody was found in eight of ten samples (80%) of group A and no samples of group B-D. The staining of glial cells and myelin in brains with anti-S100 antibody was found in ten of ten samples (100%) of group A, three of three samples (100%) of group B, and three of three samples (100%) of group C. The staining of lymphocytes and kupffer cells with anti-CD45 antibodies was found in 36 of 38 (95%) samples of group A, 25 of 28 samples (89%) of group B, 21 of 30 samples (70%) of group C, and 15 of 25 samples (60%) of group D. The staining of endothelium with anti-vimentin antibodies was found in 36 of 40 samples (90%) of group A, 6 of 28 samples (21%) of group B, 1 of 32 samples (3%) of group C, and no positive staining in group D.

The data showed that IHC staining can be used, with some limitations, in forensic tissue samples. The IHC staining of bile duct epithelium in liver with anti-KL1 antibodies was positive in 80% of samples within three days after death, while the staining of lymphocytes with anti-CD45, myelin, and glia with anti-S100 antibodies, and endothelium with anti-vimentin antibodies were positive for an even longer time after death.

Validation, Immunohistochemistry, Tissue Decomposition