G79 Histologic Diagnosis of Amniotic Fluid Embolism: Providing Context Through Immunohistochemistry

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After attending this presentation, attendees will understand that a substantial amount of cytokeratin-positive cellular material is consistently present in the vasculature of lung sections obtained at autopsy from non-gravid women, complicating the utility of keratin immunohistochemistry in the evaluation of cases of suspected amniotic fluid embolism. This cytokeratin-positive material is likely an autopsy artifact, as corroborated by the presence of TTF-1-positive cells within vascular spaces in the same lung sections. The caliber of vessel in which cytokeratin-positive material is found may help to identify true circulating keratin.

This presentation will impact the forensic science community by using immunohistochemistry to characterize the intravascular cellular material in postmortem lung specimens from non-gravid women in order to provide the appropriate context in which to assess the same immunohistochemical stains when they are employed in the evaluation of suspected cases of amniotic fluid embolism.

Amniotic fluid embolism (AFE) is among the most common natural causes of maternal death in the United States, yet AFE remains an enigmatic condition that is difficult to diagnose, the identification or confirmation of which often rests on the autopsy pathologist. The microscopic examination of multiple lung sections is essential when evaluating for AFE, with the identification of squamous cells, keratin debris, mucus, and other presumably fetal cellular debris, usually in the lungs, widely considered diagnostic in the appropriate clinical setting. Identifying these cellular elements, in particular circulating squamous cells or keratin, can be challenging despite extensive tissue sampling and thorough microscopic examination.

The difficulty in finding circulating keratinocytes is compounded by other cellular debris that may mimic their appearance, such as sloughed endothelial cells and pneumocytes. Immunohistochemistry, in particular cytokeratin AE1/AE3, has been advocated as a means to identify circulating keratinocytes. However, cytokeratin immunostains are not specific for fetal keratinocytes, and the immunohistochemical profile of intra-vascular cellular material in autopsy lung specimens from women who are not pregnant has not been formally described. To this end, three immunohistochemical stains–cytokeratin AE1/AE3, Thyroid Transcription Factor-1 (TTF-1) and CD34–were used to characterize the intravascular cellular debris in postmortem lung sections from non- gravid women in order to provide the appropriate context in which to interpret such stains in the evaluation of suspected cases of AFE.

Fourteen cases of women who died without penetrating injuries or identifiable peri-mortem needle punctures, who were not pregnant, and who were not decomposed at the time of autopsy were selected. Lung tissue was fixed in formalin and embedded in paraffin as part of the routine histologic sampling of each autopsy. Hematoxylin and eosin (H&E), TTF-1, cytokeratin AE1/AE3 and CD34 stains were performed on sections of each block of lung tissue. For purpose of comparison, H&E stains and the same three immunohistochemical stains were also performed on blocks of lung tissue from a known case of unequivocal amniotic fluid embolism and a case of a deceased neonate with abundant intra-alveolar amniotic fluid. The H&E sections were evaluated for the presence of intra-vascular material consistent with or resembling squamous cells or keratin debris. The immunostains were evaluated for the presence or absence of positive-staining intra-vascular cellular material.

All fourteen lung sections from non-gravid women contained elongate cellular material and debris by H&E staining, most of which appeared to be sloughed endothelial cells and only superficially resembled epidermal squamous cells and keratin when compared to the known AFE case and the neonatal lung. Rarely, fragments of bronchial epithelium were located in intra-vascular spaces. In both the known AFE and the neonatal lung sections, keratin characterized by distinct basophilic, “glassy” flakes of material, often in aggregates, was easily identifiable by H&E staining. No such material was identified in the lungs of the non-gravid women. All fourteen lung sections also contained intra-vascular keratin-positive cellular material, usually in great abundance. This material consisted of round cells and debris, some of which was reminiscent of keratin. However, the keratin-positive material in these lung sections was present only in larger caliber vascular spaces and not in capillaries and arterioles. By contrast, the keratin- positive material in the known AFE case was present in both large and small caliber vessels, including capillaries and arterioles. Eleven of fourteen cases had TTF-1 positive cellular material in intra-vascular spaces, although always a small amount and consisting only of round cells. All fourteen cases had abundant intra-vascular CD-34 positive material, consisting of elongate cells and debris.

Most intra-vascular cellular material that even superficially resembled circulating squamous cells and keratin in the lung sections from fourteen non-gravid women was sloughed endothelium as confirmed by

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CD34 immunostaining. The cytokeratin-positive intra- vascular cellular material in the lungs of the non-gravid women most likely represented respiratory elements trans-located into the vascular spaces as an autopsy artifact, an etiology corroborated by the presence of TTF-1-positive cells within vascular spaces. The consistent abundance of intra-vascular cytokeratin-positive cellular material emphasizes the need for caution interpreting cytokeratin stains when evaluating autopsy lung sections for amniotic fluid emboli. The caliber of vessel which contains cytokeratin-positive material may help to differentiate true circulating keratin from an autopsy artifact, as only the known AFE case had keratin in capillaries and arterioles.

Amniotic Fluid, Immunohistochemistry, Maternal Mortality

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