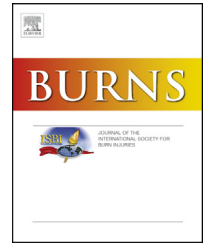


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Integrity of airway epithelium in pediatric burn autopsies: Association with age and extent of burn injury

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ABSTRACT

This study examines the structural integrity of the airway epithelium in autopsy tissues from pediatric burn subjects.

Methods: A semi-quantitative score for the degree of airway epithelial integrity was made for seventy- two pediatric burn autopsies. Multivariate ordinal logistic regression was performed to identify relationships between epithelial integrity and conditions related to tissue fixation, time of death after injury, age, total body surface area burn (TBSA), extent of 3rd degree burn, presence of inhalation injury, ventilator days and pneumonia.

Results: No significant difference in epithelial integrity scores was identified between burn only cases and those with inhalation injury. Significant correlations with bronchiolar epithelial integrity scores were identified for age, $p = 0.02$, and percent 3rd degree burn, $p = 0.02$. There was no significant relationship between epithelial integrity and time between death and autopsy, $p > 0.44$.

Conclusions: Airway epithelial loss seen in autopsy tissue is not simply an artifact of tissue fixation. The degree of compromise correlates most strongly with age and degree of burn. Further studies are needed to identify physiological or critical care factors following burn injury that contribute to compromise in the structural and functional properties of the airway epithelium.

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1. Introduction

Although pneumonia is a major cause of morbidity and mortality of burn subjects, there is limited understanding of how burn trauma and factors in critical care may alter the airway epithelium and its properties of innate defense. Intuitively, necrosis or detachment of airway epithelium would interfere with mucociliary clearance of bacteria, and this important barrier against infection would be compromised. Inhalation of toxic smoke produced inside a burning building certainly has an injurious effect on the airway epithelium, but the possible contributions of other factors in epithelial loss or damage are not clear. In a past study of sixty-two autopsy cases, we identified extensive distal migration of upper airway mucus into the small airways and parenchyma in scald only victims, suggesting dysregulation in the process of mucociliary clearance [1]. In a more recent studies of autopsy tissue, we found that regardless of the presence of inhalation injury, small airways are extensively obstructed and that 36% of the cases showed bacterial invasion of the airway mucosa [2]. Together, these studies suggest compromise in airway epithelial function following burn injury. However, our ability to study airway damage in burn subjects using autopsy tissue is limited, as postmortem detachment of the airway epithelium from the basement membrane is common and this factor must be considered in microscopic analysis.

The purpose of this study was to assess the structural integrity of the airway epithelium in autopsy tissue in relation to several measurable factors that included the time between death and autopsy, also known as the postmortem interval in which autolysis may occur, the percentage of total body surface area (TBSA) occupied by 3rd degree burns, the presence of inhalation injury, the time between injury and death, days after injury, number of days with ventilatory support and presence of pneumonia.

2. Materials and methods

Approval for the analysis of autopsy material from these patients was obtained from the University of Texas Medical Branch Institutional Review Board. Lung tissue samples from 72 autopsies performed between 2000 and 2012 were included in the study. From three to seven histological sections of lung tissue were examined from each case. All tissue sections were examined by one person experienced in the study of airway epithelial structure (RAC) without knowledge of any information related to the autopsy case. In cases where there was total absence of an epithelium, the airway was identified by the pattern of basal cells in addition to the presence of surrounding smooth muscle and vasculature, appropriate for the luminal size of the airway. Following scoring for epithelial loss, a pathologist (HKH), reviewed the slides and confirmed the pattern of airway preservation. For each case, scores were assigned for bronchial and bronchiolar epithelial integrity. A score of 0 indicated that airway epithelium was absent in all sections examined, 1 = an epithelium was present but rarely seen, 2 = the presence of an epithelium either attached or

detached but remaining within the airway cross section was commonly seen, and 3 = an epithelium, either still attached to the basement membrane or within the airway lumen, was consistently seen for all airway cross-sections. Following scoring, information related to the patient's age, gender, and race, along with conditions related to the injury and death, TBSA burned, percent third degree burn, presence of an inhalation injury, days after injury at the time of death, number of ventilatory days and time between death and autopsy were tabulated with their related airway epithelial scores. The presence of inhalation injury was recorded when this diagnosis was included in the patient's chart prior to death, and was based on conventional clinical criteria including bronchoscopic findings of soot, hyperemia and epithelial sloughing in the upper airways [3,4]. Additionally, for each case, the presence of pneumonia at death was extracted from the autopsy record. Of the 72 cases reviewed, bronchial airways, identified as airways with cartilage or mucosal glands, could not be identified in sections from ten cases.

For statistical analysis, nonparametric, Mann-Whitney rank sum tests were used to assess differences in integrity scores and case demographics between burn only and subjects with an inhalation injury using SigmaPlot software (Systat, San Jose, CA).

Multivariate ordinal logistic regression, based upon the proportional odds model with equidistant intervals, was used to model the associations of bronchial and bronchiolar scores with age, total body surface area burn, percentage of 3rd degree burn, presence of inhalation injury, presence of pneumonia, days between injury and death, number of ventilatory days and hours between death and autopsy. Age, days after injury, and hours from death to autopsy were log (base 2) transformed for better centering and to simplify interpretation. The Pearson correlation between age and third degree burn percentage was assessed as an indication of their independence. Multivariate analyses were performed using R statistical software [5] with ordinal logistic regression performed using the "ordinal" package [6]. All statistical tests assumed a 95% level of confidence.

3. Results

3.1. Case demographics

Fig. 1A and B are micrographs illustrating examples of the absence and presence of an epithelium in a bronchus and bronchiole, respectively. Following scoring of airway integrity, the specifics of each autopsy case were identified. Case demographics and issues related to the burn and inhalation injury, ventilatory days, postmortem interval and pneumonia are shown in Table 1.

3.2. Comparison of cases with inhalation injury to those with burns alone

Table 2 shows the comparison in the subjects with burns alone and those with both burn and inhalation injury. Mean values and standard deviations (SD) associated with TBSA, percent

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