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Development of a long-term ovine model of cutaneous burn and smoke inhalation injury and the effects of early excision and skin autografting

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ABSTRACT

Smoke inhalation injury frequently increases the risk of pneumonia and mortality in burn patients. The pathophysiology of acute lung injury secondary to burn and smoke inhalation is well studied, but long-term pulmonary function, especially the process of lung tissue healing following burn and smoke inhalation, has not been fully investigated. By contrast, early burn excision has become the standard of care in the management of major burn injury. While many clinical studies and small-animal experiments support the concept of early burn wound excision, and show improved survival and infectious outcomes, we have developed a new chronic ovine model of burn and smoke inhalation injury with early excision and skin grafting that can be used to investigate lung pathophysiology over a period of 3 weeks.

Materials and methods: Eighteen female sheep were surgically prepared for this study under isoflurane anesthesia. The animals were divided into three groups: an Early Excision group (20% TBSA, third-degree cutaneous burn and 36 breaths of cotton smoke followed by early excision and skin autografting at 24 h after injury, $n = 6$), a Control group (20% TBSA, third-degree cutaneous burn and 36 breaths of cotton smoke without early excision, $n = 6$) and a Sham group (no injury, no early excision, $n = 6$). After induced injury, all sheep were placed on a ventilator and fluid-resuscitated with Lactated Ringers solution (4 mL/% TBS/kg). At 24 h post-injury, early excision was carried out to fascia, and skin grafting with meshed autografts (20/1000 in., 1:4 ratio) was performed under isoflurane anesthesia. At 48 h post-injury, weaning from ventilator was begun if PaO_2/FiO_2 was above 250 and sheep were monitored for 3 weeks.

Results: At 96 h post-injury, all animals were weaned from ventilator. There are no significant differences in PaO_2/FiO_2 between Early Excision and Control groups at any points. All animals were survived for 3 weeks without infectious complication in Early Excision and Sham groups, whereas two out of six animals in the Control group had abscess in lung. The percentage of the wound healed surviving area (mean ± SD) was 74.7 ± 7.8% on 17 days post-surgery in the Early Excision group. Lung wet-to-dry weight ratio (mean ± SD) was

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significantly increased in the Early Excision group vs. Sham group ($p < 0.05$). The calculated net fluid balance significantly increased in the early excision compared to those seen in the Sham and Control groups. Plasma protein, oncotic pressure, hematocrit of % baseline, hemoglobin of % baseline, white blood cell and neutrophil were significantly decreased in the Early Excision group vs. Control group.

Conclusions: The early excision model closely resembles practice in a clinical setting and allows long-term observations of pulmonary function following burn and smoke inhalation injury. Further studies are warranted to assess lung tissue scarring and measuring collagen deposition, lung compliance and diffusion capacity.

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1. Introduction

The short-term pathophysiology of acute lung injury secondary to burn and smoke inhalation has been studied extensively [1–4] but there are few studies of long-term pulmonary pathophysiology following burn and smoke inhalation [5]. The purpose of present study was to develop an animal model of smoke inhalation injury and cutaneous burn to document the long-term effect on the pulmonary parenchyma. In order to study the long-term effect of injury, the animals need to survive over 2 weeks with appropriate treatment. Early burn excision and skin grafting have become the standard of care in the management of major burns [6]. Many clinical studies and small-animal experiments support the concept of early burn wound excision and show decreased operative blood loss, length of hospitalization, and incidence of infection compared with late excision [7–12]. The effects of early excision and skin grafting have not, to our knowledge, been studied well in a large-animal model. We hypothesized that long-term model of smoke inhalation injury and cutaneous burn could be produced if early burn excision and autografting were utilized.

In the present study, we have developed a new ovine model of smoke inhalation injury and cutaneous burn with early excision and skin autografting to investigate chronically in lung tissue. We also demonstrated the effects of the early excision and skin autografting in our model with inhalation injury.

2. Materials and methods

This study was approved by the Animal Care and Use Committee of the University of Texas Medical Branch (Galveston, TX, USA) and conducted in compliance with the guidelines of the National Institutes of Health and the American Physiological Society for the care and use of laboratory animals.

2.1. *Surgical preparation*

Eighteen female sheep were surgically prepared for this study under isoflurane anesthesia. The mean animal weight (mean ± SD) was 34 ± 4.6 kg. The right femoral artery was canulated with Silastic catheter (Intracath; 16 gauge, 24 in.; Becton Dickinson Vascular Access, Sandy, UT, USA). A thermodilution catheter (Swan–Ganz model 131F7, Baxter, Edwards Critical-Care Division, Irvine, CA, USA) was introduced through the right external jugular vein into the pulmonary artery. Through the left fifth intercostal space, a catheter (Durastic silicone tubing DT08, 0.062-in. ID, 0.125-in. OD; Allied Biomedical, Paso Robles, CA, USA) was positioned in the left atrium. The animals were given 5–7 days to recover from the surgical procedure, with free access to food and water.

2.2. *Experimental protocol*

Before the experiment, the vascular catheters were connected to the monitoring devices, and maintenance fluid (Ringer lactate, 2 mL/kg) was started. After baseline measurements and sample collections were completed, the animals were randomized into three groups: Early Excision group (20% TBSA, third-degree cutaneous burn and 36 breaths of cotton smoke followed by early excision and skin autografting at 24 h after injury, $n = 6$), Control group (20% TBSA, third-degree cutaneous burn and 36 breaths of cotton smoke without early excision, $n = 6$) and Sham group (no injury, no early excision, $n = 6$).

Immediately after injury, anesthesia was discontinued. The animals were allowed to awaken but were maintained on mechanical ventilation (Servo Ventilator 900C, Siemens-Elema AB, Sweden) for at least a 48 h experimental period. This was continued until the weaning process was completed. Ventilation was performed with a positive end-expiratory pressure of 5 cmH_2O and a tidal volume of 15 mg/kg. During the first 3 h after injury, the inspiratory O_2 concentration was maintained at 100% to induce rapid clearance of carboxyhemoglobin after smoke inhalation. The ventilation was then adjusted according to blood gas analysis to maintain arterial O_2 saturation >90% and PCO_2 between 25 and 30 mmHg. At 48 h post-injury, weaning from ventilator was begun if PaO_2/FiO_2 was above 250. Animals were then monitored for 3 weeks. Fluid resuscitation was given during the first 48 h experimental period with Ringer's lactate solution following the Parkland formula (4 mL/% burned surface area/kg body weight for first 24 h and 2 mL/% burned surface area/kg body weight/day for the next 24 h). One-half of the volume for the first day was infused in the initial 8 h, and the remainder was infused in the next 16 h. From 48 h to 432 h, the animals received Ringer's lactate (2 mL/% burned surface area/kg body weight/day). For 96 h post-injury, animals were allowed free access to food but not to water, to accurately measure fluid intake. Free access to water was permitted after this period. A Foley catheter was

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