

Original Article

A novel device to create consistent deep dermal burns in a porcine model

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Abstract: We conducted this study to evaluate a novel device to create a consistent and reproducible deep partial thickness burn in a porcine model. A thermostatically controlled, heated aluminium disc device was fashioned by the Biomedical Department of our institution. Contact burns were made on the flank of two Great White pigs by applying the device heated to 92°C at intervals of 5, 10, 15 and 20 seconds to four separate test areas area of skin. Biopsies for histological analysis of burn depth were taken on day 0 at 10 minutes post burn and on day 8. Biopsies taken at day 0 revealed superficial to mid-dermal burns, with minimal dermal edema and necrosis. Those from day 8 showed mid to deep dermal edema and necrosis in all four test areas following a 20 second contact duration burn. The new contact burn device was able to create a consistent deep dermal burn after 20 seconds of contact. We anticipate that this new device could be used to investigate the development of hypertrophic scarring in a porcine model.

Keywords: Contact burn, porcine model, partial thickness

Introduction

Hypertrophic scarring (HTS), which occur more commonly in children following a deep dermal burn injury, can be disfiguring and debilitating. Burns that do not heal within two weeks are usually treated with a split skin graft (SSG): the optimal timing for grafting a burn wound in order to minimise scarring remains debateable. A porcine model has the closest similarity to human skin compared to other animal models and has been used extensively for wound healing studies [1, 2].

Our institution has been investigating the duration of thermal contact required to produce consistent deep dermal contact burns in a porcine model. The device previously used was a cylindrical vessel made of Acetal plastic (Delrin®, DuPont, North Ryde, NSW, Australia), an inert, synthetic, non-conducting polymer. The base had a diameter of 50 mm and was covered by a piece of Latex glove, which is secured in place using an O-ring screw mechanism. Hot water at a temperature of 92°C was

added to the vessel from a kettle. A covering lid was then secured with a temperature monitoring device insitu. Using this device, the duration of contact required to create a deep dermal burn was found to be 20 seconds [3]. One limitation of this model in evaluating HTS was that the burn wounds created were of inconsistent depth.

The aim of this pilot study was to evaluate a novel device, developed in conjunction with our institutions Biomedical Department, to ensure a more consistent and reproducible deep dermal burn in our porcine model.

Materials and methods

Contact burn device

A novel contact burn device was developed in conjunction with the Biomedical Department of our institution (**Figure 1**). This consisted of a hand-held, insulated electrical device with an aluminium disc plate measuring 2.5 cm in diameter. The inbuilt thermostat was set to 92°C.

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Figure 1. New contact burn device.



Figure 3. Punch biopsy of burn site.



Figure 2. Induction of burn in porcine model.



Figure 4. Full thickness injury following 20 second contact burn.

Animal model

Two female Great White piglets, approximately 8 weeks of age, were housed in the vivarium for one week to acclimatise. They were fed on a standard diet and were fasted overnight before the procedure. The study was approved by our institution's Ethics Committee.

General anesthesia and monitoring

Light anesthesia with Zoletil (1.1 mg/kg, IM) and Isoflurane by mask was used. Animal care staff monitored closely for the symptoms of pain and provided additional analgesia of Buprenorphine 0.005-0.02 mg/kg IM or IV as required. For the duration of each experiment, warming mats were used to prevent hypothermia and reduce recovery time following the anesthesia. Pulse oximeter, blood pressure, respiratory rate, heart rate and body temperature were observed. Immediately following the second biopsy, Zoletil 4.4 mg/kg, followed by IV pentobarbitone to effect (absence of heart beat, respiration and pupillary dilation), was used to euthanize the animals.

Burn induction

Once the animal was adequately anesthetised, the skin was shaved, washed and dried. The intended test areas were marked out by tracing around the rim of the aluminium plate device with a surgical marking pen. Each area exposed to thermal injury was $\pi r^2 = 3.1412 \times 0.0125 \text{ m} \times 0.0125 \text{ m} = 0.00049 \text{ m}^2$. The total body surface area (TBSA) exposed to thermal injury in each pig was $8 \times 0.00049 = 0.0039 \text{ m}^2$.

The skin was moistened slightly with sterile saline to ensure optimal thermal coupling. Skin was prepared with Povidone-iodine. To reduce the risk of subsequent infection, Cefepime, a fourth generation cephalosporin, was adminis-

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Table 1. Day 8 biopsies

Contact Duration (seconds)	Depth of Dermal Necrosis	Depth of Dermal Oedema
5	mid	mid
10	mid*	mid
15	mid-deep	mid-deep
20	deep*	deep

*Summary of day 8 skin biopsy histological assessment of burn depth.

tered intramuscularly at a dose of 50 mg/kg. Standardised dermal burns were produced by applying the device perpendicular to the pig flank and without using pressure at intervals of 5, 10, 15 and 20 seconds to four separate test areas area of skin 2.5 cm in diameter (**Figure 2**). Four burns, separated by 4 cm from the boundaries of each burn to minimise any physiological or inflammatory interaction, were created on each flank. Different burn durations were randomly allocated to different sites in each pig to minimise possible bias from body sites healing differently. Core temperatures were recorded continuously during procedure and photographs were taken for independent clinician review.

Assessment

Punch biopsy (4 mm diameter) of full thickness skin from the burn site was performed after burn induction (**Figure 3**). Specimens were transported in formalin and routinely processed in the pathology department. Standard staining methods included haematoxylin and eosin, PAS and Masson's trichrome stains. Depth of injury was assessed on the basis of the depth of microvascular occlusion, collagen discolouration, intercollagen basophilic material, and necrosis of endothelial, epithelial and mesenchymal cells. Depth of injury was expressed as either superficial/epidermal, superficial dermal, mid-dermal, deep-dermal or full thickness. A second set of biopsies was performed on day 8 after injury to assess burn depth progression. The pathologist assessing the specimen was blinded to the duration of the contact burn.

Results

No clinical evidence of infection was observed in any of the test areas and there were no adverse events or morbidity. The device was easy to use and appeared to give burns of con-

sistent size and depth. Photographs were objectively assessed and test areas with 20 s contact duration were noted to have the appearance of a full thickness burn (**Figure 4**).

Biopsies taken were sent for full histological assessment. The biopsies samples taken at day 8 showed a distinct pattern at specific time points. The 5 s and 10 s samples all showed features of mid dermal necrosis and oedema. Despite site randomization, all specimens at 15 s had mid dermal necrosis and oedema and 20 s specimens had deep dermal necrosis and oedema (**Table 1**).

Discussion

Hypertrophic scarring represents the unsightly raised and often debilitating endpoint of burns wound healing. At present, there remains comparatively limited evidence regarding the optimal timing for split skin grafting and the effect that this has on scar outcome. Multiple studies have shown that burn wounds which take longer than 21 days or longer to heal have more than a 70% risk of developing HTS [4, 5]. Cubison *et al* [6] have previously reported the relationship between healing time and the development of HTS. They found that there was a higher incidence in HTS in burns grafted at day 10-14 (33%) compared to those conservatively managed (2%), but similar rates of HTS for burns grafted at day 15-21. This led them to conclude that surgery should be reserved for scald burns taking longer than 21 days to heal. The difficulty in investigating this objectively in children lies in the fact that multiple tissue samples need to be obtained and patients would also need to be randomised into a non-operative group. Additional evidence in relation to the timing of grafting and its impact on HTS from an appropriate animal model might provide clinicians with more information to design an appropriate clinical trial.

The porcine model has been used extensively for wound healing studies [7] and others have reported it to be an ideal model for HTS [8]. Its relative similarity to human skin compared to other animal models makes it the ideal choice. HTS in porcine models has been reported in two breeds of pig, namely the Red Duroc pig and the Large White pig. Our group at CHW has successfully used a modification to Jandera *et al.*'s model [9] to conduct a series of Burns First

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Aid Treatment (BFAT) experiments over the past four years in the Large White pig. Whilst this breed has been shown to develop consistent scarring following a contact burn injury, burn depth remained inconsistent [10]. The aim of our pilot study was to modify the current protocol to create a deep partial thickness burn (PTB) wound with a new burn contact device in order to create a consistent and reproducible hypertrophic scar.

Our results showed that contact duration of 20 seconds was necessary to create a deep dermal burn, with deep dermal oedema and necrosis seen in these test areas. The variable findings seen in biopsies taken at day 0 (10 minutes post burn induction) highlight the progression of burn depth with time.

In order to make a more consistent burn, the new device was designed with a small, flat aluminium disc plate. The device was lightweight and easy to operate. Based on Stahl's measurement, the calculation of the body surface area of a piglet [11] is $0.11 \times \text{wt}$ (in Kg). Using our new aluminium contact burn device, the total body surface area exposed to thermal injury in each pig was 0.0039 m^2 . This technique ensured that the total area exposed to thermal injury is less than 5% TBSA of each piglet. By using antimicrobial prophylaxis we were able to reduce the incidence of infection as another variable which may affect burn wound depth and scarring.

We were able to create a consistent, reproducible deep dermal burn with a novel aluminium disc device heated to 92°C at 20 seconds contact duration. This can be used to create the ideal porcine model to investigate HTS. The progression of burn depth with time was substantiated by comparing Day 0 and Day 8 biopsy results. Following on from our pilot studies, we hope to explore the temporal relationship between timing of excision and skin grafting of the burn to the development of HTS.

Disclosure of conflict of interest

None.

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