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A new model for the standardization of experimental burn wounds

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ABSTRACT

Background: Burns are common and recurrent events treated by physicians on a daily basis at most emergency rooms around the world. There is a constant need to understand the physiopathology of burns, so as to minimize their devastating results. The objective of the present report is to describe a burn apparatus in association with an innovative method of animal fixation, as to produce burns of varying sizes and depths.

Methods: Rats were subjected to burns of 60 °C, 70 °C, and 80 °C for 10 s and after 3 days half of the rats in each group were killed and the resulting lesions were analyzed using histological techniques. In the other half of the rats the wound was measured weakly until complete re-epithelialization.

Results: All burns were easily visible and the histological feature for the 60 °C burn was a superficial second-degree burn (28% of the dermis), for 70 °C we observed a deep second-degree burn (72% of the dermis), and in the 80 °C group, a third degree-burn was present (100% of the dermis).

Conclusions: This is a safe, reliable, easy to construct and use model that has the ability to produce a regular and uniform reproducible burn due to precise temperature control associated with standardized animal positioning.

1. Introduction

Burns are among the most common injuries in modern life [1]. In the United States alone, every year 450,000 patients receive medical treatment related to burns, with over 40,000 hospital admissions and 3500 deaths due to burns [2].

Initial studies concerning burns were focused on improving the overall survival rate. These works provided a better understanding of the physiopathology of burns and led to enhanced resuscitation techniques that drastically reduced the number of deaths of burn victims during the early phases of injury. In a second phase, the focus of investigation shifted toward minimizing morbidity and improving the quality of life.

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Abbreviations: C, Celsius; V, Volts.

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Nowadays, one of the primary approaches concerns scar quality, especially when the burn does not affect a large body surface [3]. Therefore, in experimental models, the burn areas must be small enough not to have a systemic repercussion, but yet large enough to permit adequate observation and sampling.

An experimental model is essential to test therapies before clinical use. In order to evaluate the effectiveness of burn wound treatment, however, it is extremely important to be able to create predictable and uniform burns, where most of the confounding variables can be eliminated. Burn depth, defined by three different elements – temperature, time of exposure, and contact pressure – is the primary determinant of prognosis [4–7].

The most frequent models are the contact burn and the scalding burn [1,5,8–12]. The contact burn is normally induced with the use of a metal bar that is heated in a temperature-controlled water bath and applied to the skin for a pre-determined amount of time [5,8,9]. In contrast, the scalding model usually employs a template with an aperture through which part of the body is immersed in a water bath with a controlled temperature for a specific period of time [1,10–12].

We devised a refined temperature-controlled apparatus that can be easily assembled and operated, and that is capable of producing different size burns. Associated with a method of fixation and stabilization of the skin, the apparatus allows pressing down perpendicular to the skin surface while maintaining a constant pressure.

The objective of the present study was to describe a technique for inducing a burn of standard and constant extent and depth.

2. Materials and methods

2.1. *Animals*

The study was approved by the Ethics Committee on Animal Research of the Biology Institute Roberto Alcantara Gomes, Rio de Janeiro State University, Brazil (protocol number 056/2012). All procedures rigorously followed current guidelines for animal experimentation [13].

Thirty-six male Wistar rats (275–300 g) were used. The animals were housed in appropriate cages, one animal to a cage, under conditions of controlled temperature and humidity, on a 12 h light/12 h dark cycle. Free access to water and standard laboratory chow was allowed.

Two days prior to the burn creation the animal dorsum was shaved with electric clippers and depilated with a commercial depilatory cream (VEET(TM) – Rickitt Benckiser Colombia SA; Cali, Colombia).

After a 6-h fast, the animals were anesthetized intramuscularly with 80 mg/kg ketamine (Agener União; Embu-Guaçu, Brazil) and 12 mg/kg xylazine (Agener União; Embu-Guaçu, Brazil). The entire procedure was performed under aseptic conditions.

After sedation was achieved, the animal was laid on its side with a 2-cm high Styrofoam board adjacent to it. The skin was stretched and fixed with four hypodermic needles (1.25 cm × 26G) (Solidor; Barueri, Brazil) so that that the head of the burn apparatus would not only fit completely but would also permit a complete and perpendicular contact of the weighted apparatus – 1 kg of lead (Fig. 1).

2.2. *Burn apparatus*

The head of a high power commercial soldering iron (Weller 80 Watts – Cooper Hand Tools; Sorocaba, Brazil) was removed and replaced with one of three cylindrical aluminum heads of different diameters: 19, 23, or 32 mm.

A small hole was drilled 3 mm above the contact surface, and a Type K temperature probe (Minipa; São Paulo, Brazil) was inserted and attached to a digital thermometer (Minipa MT-401A; São Paulo, Brazil) (Fig. 2).

Soldering iron works as a resistance. The insertion of a variable autotransformer (JNG TDGC2 0,5KvA; Yueqing, China) between the power outlet and the soldering iron allowed varying the output voltage for a steady AC input voltage. This permits the regulation of the voltage to the soldering iron, leading to a precise control of its temperature. Since the variable autotransformer has an analog dial, we used a digital voltmeter (Minipa ET-1002; São Paulo, Brazil) to determine the exact output. During the pretest phase, we determined that to obtain the expected temperatures – 60 °C, 70 °C, and 80 °C – it was necessary to set the output on the voltage autotransformer to 55 V, 60 V, and 65 V, respectively, while the time needed to stabilize the temperature was 20 min.

2.3. *Burns*

For the present study we opted for the aluminum cylinder head measuring 23 mm in diameter. Using Meeh's formula ($A = 10 \times W^{2/3}$, where: A = area in cm^2, 10 is a constant and W = weight in grams) to calculate the surface area of the animal, the burn represented about 1% of the total surface area [14]. The device was heated to 60 °C, 70 °C, or 80 °C. When the cylinder reached the desired temperature, we waited 5 s for stabilization before applying it to the skin for 10 s. We used 12 animals for each temperature and it was necessary to wait 3 min between animals for the cylinder to recover the desired temperature.

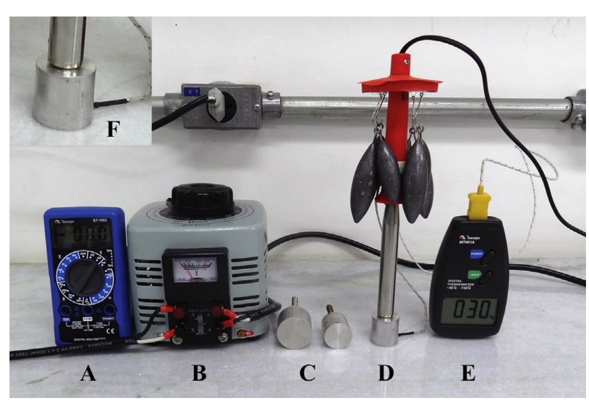

Fig. 1 – The burn apparatus: A – digital voltmeter; B – variable autotransformer; C – aluminum cylindrical heads of different diameters; D – high power commercial soldering iron; E – digital thermometer; F – close up of the type K temperature probe that was inserted into the soldering head.