8 - Experimental model for transcutaneous electrical nerve stimulation on ischemic random skin flap in rats

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ABSTRACT - The objective of this paper was to develop an experimental model to be used in the study of Transcutaneous Electrical Nerve Stimulation (TENS) on viability of random skin flap in rats. The sample was 15 Wistar-EPM rats. The random skin flap measured 10x4 cm and a plastic barrier was placed between the flap and the donor site. The animals were submitted to TENS for 1 hour immediately after the surgery and on the two subsequent days. On the seventh postoperative day, the percentage of necrotic area was measured and calculated. The experimental model proved to be reliable to be used in the study of effects of Transcutaneous Electrical Nerve Stimulation in random skin flap in rats.

KEY WORDS - Electrical stimulation. Skin flap. Necrosis.

Introduction

Skin flaps are largely used in all fields of plastic surgery, especially in reconstructive surgery. They have been utilized for centuries and, during this time, one preoccupation has been to develop techniques to provide more assurance in skin flap realization. The research of skin flaps survival mechanisms and your possible curative factors have been motive for publications. But in spite of all of these studies related with random skin flap survival, little progress was gotten in this field in the last 50 years.

It is know that one of main complications that occur in the creation of the flap is ischemia, which, in a large number of cases, develops to tissue necrosis taking a failure in proposed treatment. Due to that complication, a considerable amount of research has been done with the aim of improving the blood flow in flaps, decreasing ischemic conditions and preventing necrosis.

In the literature, there have been publications about several types of drugs, such as vasodilators, calcium channel blockers, prostaglandin inhibitors, anticoagulants, antiadrenergics and antioxidants. However, many

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of those present undesirable adverse effects, which makes their use in clinical practice unviable. Therefore, a new research field using nonpharmacological agents such as acupuncture and electroacupuncture[^16[^11], pulsed electromagnetic energy[^12], low-power lasers[^13[^14[^15]], polarized low frequency electrical currents[^16[^15]] and nonpolarized currents[^18[^19]] has emerged.

Among these resources, the transcutaneous electrical nerve stimulation has deserved detach due to its low cost and application facility, begining to be studied as possible method in the treatment of ischemic skin flaps.

**Proposition**

Due to the characteristics of this kind of stimulation, associated with the methodological deficiency of few experimental studies performed in animals with relation of current parameters and operation procedure, the purpose of this study was to develop an experimental model to be used in the study of Transcutaneous Electrical Nerve Stimulation (TENS) on viability of random skin flap in rats.

**Method description**

The study was approved by Comission of Ethics in Research of Universidade Federal de São Paulo – Escola Paulista de Medicina going in the current apt legislation.

**Animals**

The present study used 15 rats (Rattus norvegicus: var. albinus, Rodentia, Mammalia), adults, males, of lineage Wistar EPM – 1, weighing 260 to 330 g.

**Electrostimulator**

The device used in this study was the Orion Tens® [Orion Aparelhos para Fisioterapia LTDA; serie number 00849], digital e controlled by microprocessor. The emitted pulses are rectangulars, biphasis and simmetricals. With electrical stimulator, it was used a cable, two silicone electrodes (4,2 x 1,5 cm), gel and adhesive tape for fix the electrodes.

**Operation technique**

In animals, it was realized a random skin flap with cranial base, measuring 10 cm length and 4 cm width. The animals were anesthetized with sodium thiopental (50 mg/kg) intraperitoneally during the operatory procedure and during electrostimulation sessions. After anesthesia induction, the rats were positioned in a plane surface with members extended and it was performed a digital tricotomy in their backs.

Then it was done the planning of the flap through a plastic mould [film F-1 (poliester + polietilene)], cut out in pattern sizes (10x4 cm) in the backs of animals, taking as limits the inferior angles of the scapulae and the superior bones of pelvis (Figure 1). The random skin flap with cranial base was cut by scalpel and elevated through deep fascia, including the superficial fascia, panniculus camosus, the subcutaneous tissue and skin[^18[^9]] (Figure 2). After flap elevation a plastic barrier (film F1), with same dimensions (10x4 cm), was placed between it and the donnor site[^7]. The suture was realized with simple nylon 4-0 stitches (Figure 3).[^9]
After operatory procedure, the animals were kept anesthetized and subjected to Transcutaneous Electrical Nerve Stimulation (TENS) for 1 hour and, on the two subsequent days, in determined time, with high intensity (± 15 mA) and high frequency (80 Hz). The electrodes were placed on the base of the flap, where the first was positioned thorough of incision beginning and the second apart 1.5 cm from first electrode, in direction of a distal portion of the flap (Figure 4). The electrical pulses had a duration of 200 microseconds. After the electrical stimulation sessions, the animals got back to their respective cage and received commercial ration and water *ad libitum*.
FIGURE 4 - Electrodes were placed at the base of the flap for the transcutaneous electrical nerve stimulation.

Method for estimate percentage of necrotic area in distal portion of flaps

The percentage of skin flap necrosis area was calculated on the seventh postoperative day via the paper template method. The limit between viable tissue characterized by soft skin, rosy, warm and with hair and necrotic tissue (stiff skin, dark, cool and without hair) was demarcated in the animals. A mould of entire flap was drawn and cut in transparent paper, being checked in a precision balance (+/- 0.001g error). It was cut from this fragment just the correspondent area to flap necrosis that was also checked. After that, it was used the following formula:

\[
\text{percentage of necrosis area of the flap} = \frac{\text{weight of paper template of flap necrosis}}{\text{weight of paper template of total area of flap}} \times 100
\]

Perspectives

The experimental model proved to be reliable to be used in the study of effects of transcutaneous electrical nerve stimulation in random skin flap in rats.

References

RESUMO - Este artigo propõe o desenvolvimento de um modelo experimental para verificar o efeito da Estimulação Elétrica Nervosa Transcutânea (TENS) na viabilidade do retalho cutâneo randômico em ratos. A amostra constituiu-se de 15 ratos, da linhagem Wistar-EPM. O retalho cutâneo randômico foi realizado com dimensões de 10x4 cm e uma barreira plástica foi interposta entre o mesmo e o leito doador. Os animais foram submetidos à TENS por 1 hora imediatamente após a operação e nos outros dois dias subsequentes. No sétimo dia pós-operatório foram calculadas as porcentagens de área de necrose. O modelo experimental mostrou-se factível para estudo dos efeitos da Estimulação Elétrica Nervosa Transcutânea em retalho cutâneo randômico em ratos.


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