Capsaicin on the viability of random-pattern skin flaps in rats[1]

Capsaicina na viabilidade de retalhos isquêmicos randômicos em ratos

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ABSTRACT
Purpose: To evaluate the effects of capsaicin on the viability of ischemic random-pattern skin flaps in rats. **Methods**: Forty EPM1-Wistar rats were randomized into two groups of 20 animals each, the capsaicin group and the control group. A random-pattern skin flap measuring 10 x 4cm was raised and a plastic barrier was placed between the flap and the donor site. After the surgical procedure, the control group was treated with an inert vehicle in the form of a cream applied uniformly to a rayon bandage which, in turn, was applied to the surface of the skin flap. The capsaicin group was treated in the same way, but in this case capsaicin was added to the cream. This procedure was repeated for two consecutive days. **Results**: There was a significantly smaller amount of flap necrosis in the capsaicin group (35.07%) than in the control group (44.75%) (p=0.035). **Conclusion**: Topical administration of capsaicin improved the viability of ischemic random-pattern skin flaps in rats.
Key words: Capsaicin. Surgical Flaps. Neuropeptides. Necrosis. Rats.

RESUMO
Objetivo: Avaliar os efeitos da capsaicina na viabilidade de retalhos isquêmicos randômicos em ratos. **Métodos**: Quarenta ratos EPM1-Wistar foram distribuídos ao acaso em dois grupos de 20 animais cada, um grupo capsaicina e um grupo controle. Um retalho isquêmico randômico medindo 10 x 4cm foi elevado e uma barreira plástica foi colocada entre o retalho e a área doadora. Após o procedimento cirúrgico, o grupo controle foi tratado com um veículo inerte sob a forma creme aplicado uniformemente sobre uma atadura de *rayon*, que, por sua vez, foi aplicada à superfície do retalho. O grupo capsaicina foi tratado da mesma forma, porém a capsaicina foi adicionada ao creme. Este procedimento foi repetido por dois dias consecutivos. **Resultados**: Houve uma quantidade significativamente menor da necrose do retalho no grupo capsaicina (35,07%) comparado ao grupo controle (44,75%) (p=0,035). **Conclusão**: A administração tópica da capsaicina melhorou a viabilidade de retalhos isquêmicos randômicos em ratos.
Descritores: Capsaicina. Retalhos Cirúrgicos. Neuropeptídeos. Necrose. Ratos.

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Introduction

Necrosis of skin flaps may be attributed to extrinsic and intrinsic factors. Among the intrinsic factors, the best known is the inadequate vascular perfusion of the flap[1,2]. In many studies, drugs such as antiadrenergics[3,4], vasodilators[5,6], antispasmodics[7], anticoagulants and calcium-channel blockers[8] have been used to improve flap viability. However, undesirable side effects, high drug prices, restricted availability or the need for a long-term treatment during the preoperative period make some drugs impractical for clinical use[8].

In other studies, researchers tried to improve the viability of skin flaps by topically applying medicines, seeking to increase local action and minimize systemic action, thereby reducing side effects[6,9]. Among these drugs is capsaicin[9-11], a substance that, in general, promotes vasodilatation and inhibits platelet aggregation[12,13].

In this context, capsaicin has the potential to be administered topically in the postoperative period, and may improve the viability of ischemic skin flaps by transdermal action. However, in these studies[10,11] the skin flap remained covered with bandage containing capsaicin throughout the treatment period,

preventing the continuous evaluation of the signs of ischemia and necrosis development. Also in most studies the capsaicin treatment usually varies from 6 to 7 days in the postoperative period[9-11]. These limitations make it potentially difficult to determine the applicability of this method to clinical practice. Thus, the present study was designed to investigate the usefulness of a different method of capsaicin application, with the overall goal of optimizing the technique so capsaicin may be used clinically to treat ischemic skin flaps in the future. Therefore, the purpose of this study was to evaluate the effects of capsaicin on the viability of ischemic random-pattern skin flaps in rats.

Methods

Forty adult male EPM1-Wistar rats (*Rattus norvegicus albinus*), weighing between 230 and 380 g, were used in the study. The animals were housed in individual cages on a 12:12 hour light-dark cycle, and fed standard rat chow and water *ad libitum*. This research was approved by the Research Ethics Committee at the UNIFESP/EPM, Sao Paulo, Brazil, under the process number 0381/03.

The animals were randomly assigned by lottery to either the capsaicin or control groups of 20 animals each.

The rats were anesthetized intraperitoneally with tiletamine hydrochloride (25 mg/kg) and zolazepam hydrochloride (25 mg/kg) for surgery, and prior to the application of capsaicin on the two consecutive days. Following anesthesia, each rat was placed on a flat surface with legs extended, the back was shaved, and the skin flap was outlined. After, a 10 x 4-cm, cranially-based dorsal skin flap was raised[14]; its cranial limit was determined by a transverse line at the level of the angles of the scapulae. The skin flap was dissected to the deep fascia and a plastic barrier was placed between the flap and its bed (donor site)[15,16]. The flap was returned to its original position and sutured in place with simple interrupted 4-0 monofilament nylon sutures[17]. A solution containing 1g of capsaicin powder (dissolved in 10 ml of 70 % ethanol) was added to a cream, consisting of cetostearyl alcohol (Lanette® wax cream) and mineral oil, to a concentration of 0.2% (200 mg of capsaicin per 100 g of cream). After surgery, 2g of cream without capsaicin (control group) or 2g of the same cream with 0.2% capsaicin (capsaicin group) was uniformly applied to a rayon bandage. Following, the bandage was carefully positioned on the skin flap (Figure 1) in order to prevent its contact with sutures on the flap edges. After 35 minutes, the rayon bandage was removed and the animal was returned to the cage. This application was repeated for two consecutive days in all animals of both groups.

The percentage of skin flap necrosis was measured on the seventh postoperative day, using the paper template method[18] by an observer who was blind to the treatment condition.

Statistical analysis was performed using Student's t-test at a significance level of 5% ($p \leq 0.05$).

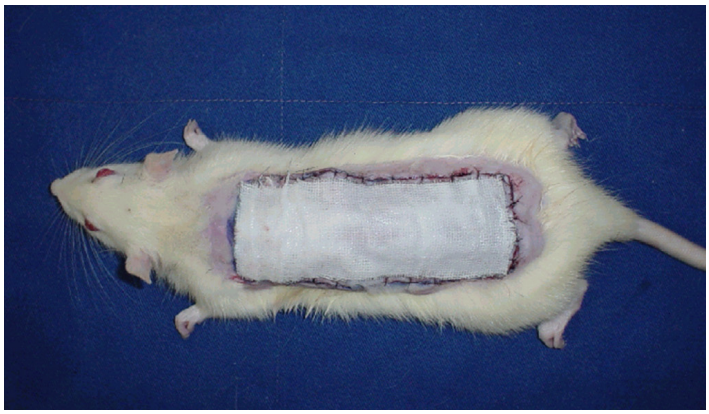

FIGURE 1 - Rayon bandage containing capsaicin or inert vehicle placed on the skin flap

Results

There was a significantly smaller amount of flap necrosis (p=0.035) in the capsaicin group than in the control group. The percentages of necrotic area (mean ± standard deviation) in the capsaicin and control groups were 35.07 ± 14.67% and 44.75 ± 12.81%, respectively (Figure 2).

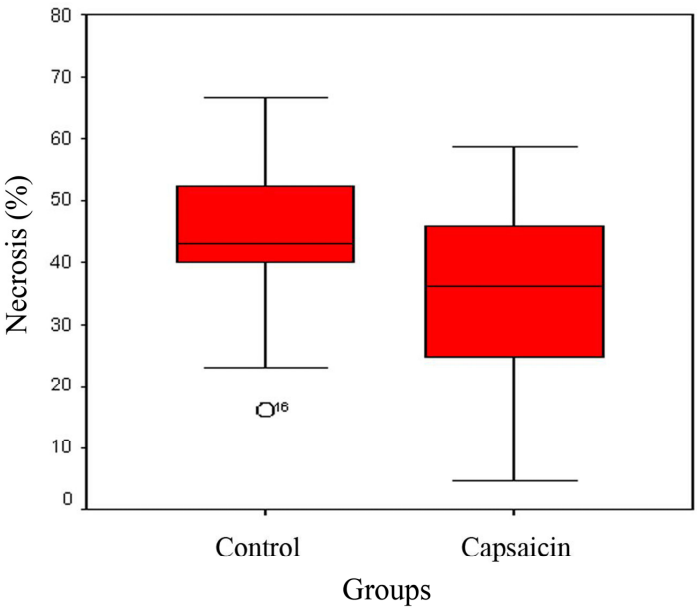

FIGURE 2 - Distribution of the percentage of necrotic area per group

Student's t-test
Control group x capsaicin group (p=0.035)

Discussion

Skin flap necrosis after reconstructive surgeries remains an important problem for plastic surgeons[17]. In view of the physiological mechanisms of ischemia that lead to flap necrosis and the effects of capsaicin, it is important to study this substance with the aim of improving the viability of ischemic random-pattern skin flaps. Iinuma and Sawada[10] attributed the improvement in skin flap viability after topical application of capsaicin to platelet disaggregation. On the other hand, Miyawaki *et al.*[9] concluded that the beneficial effect of capsaicin in the skin flap viability is due to vasodilation caused by the release of vasodilator neuropeptides, such as substance P (SP) and calcitonin gene-related peptide (CGRP), and increased neovascularization[4,19,20].

The amount of capsaicin and application time used in our study were based on the work by Wang *et al.*[21] Capsaicin was indirectly applied to the skin; it was first uniformly applied with a spatula to a rayon bandage (which was slightly smaller than the flap dimensions) to prevent manual contact that could alter the pH of the substance. The rayon bandage was removed 35 minutes after application to keep the bandage from slipping while the animal was caged and to prevent its removal by the animal, which would interrupt the treatment process and make our study different from other similar studies in the literature[10,11].

Many authors have shown that percutaneous absorption of capsaicin through animal and human skin occurs within 3 to 4 minutes after application, and produces maximal vasodilation at about 30 to 34 minutes[21-25].

The 3-day application period was chosen based on studies of transcutaneous electrical nerve stimulation[4,17], and iontophoresis of CGRP[26]. Although these modalities of treatment differ from that used in the present study, their mechanisms of action by which the viability of skin flaps is enhanced are similar to that of capsaicin, that is, vasodilation induced by neuropeptides release.

The mean percentage of necrotic area was 35.07% in the capsaicin group and 44.75% in the control group. There was a significant difference between groups ($p=0.035$), leading to the conclusion that capsaicin improved the ischemic condition of the random-pattern skin flaps, even if the postoperative treatment period (3 days) was shorter than in previous studies[9-11].

Capsaicin in concentrations of continuous topical administration causes depletion of neurotransmitters, leading to a reversible interruption of the conductivity of sensory nerve fibers. For this reason, capsaicin is widely used in the treatment of pain[13,25,27-29]. However, further studies on the application of capsaicin in humans are needed in order to adapt the model, and determine the concentration of capsaicin suitable for human use, since no study on this subject was found in the literature.

Conclusion

Topical administration of capsaicin improved the viability of ischemic random-pattern skin flaps in rats.

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