Homeopathy (2012) 101, 147–153
© 2012 The Faculty of Homeopathy
http://dx.doi.org/10.1016/j.homp.2012.05.006, available online at http://www.sciencedirect.com

ORIGINAL PAPER

Effects of microcurrent application alone or in combination with topical *Hypericum perforatum* L. and *Arnica montana* L. on surgically induced wound healing in Wistar rats

Fabiene CB Castro[1], Amanda Magre[1], Ricardo Cherpinski[1], Paulo M Zelante[1], Lia MG Neves[2], Marcelo AM Esquisatto[2], Fernanda AS Mendonça[2] and Gláucia MT Santos[2,*]

[1]*Herminio Ometto University Center, Araras, SP, Brazil*
[2]*Graduate Program in Biomedical Sciences, Herminio Ometto University Center, Araras, SP, Brazil*

Objectives: **This study evaluated the wound healing activity of microcurrent application alone or in combination with topical *Hypericum perforatum* L. and *Arnica montana* L. on skin surgical incision surgically induced on the back of Wistar rats.**
Design: **The animals were randomly divided into six groups: (1) no intervention (control group); (2) microcurrent application (10 μA/2 min); (3) topical application of gel containing *H. perforatum*; (4) topical application of *H. perforatum* gel and microcurrent (10 μA/2 min); (5) topical application of gel containing *A. montana*; (6) topical application of *A. montana* gel and microcurrent (10 μA/2 min). Tissue samples were obtained on the 2nd, 6th and 10th days after injury and submitted to structural and morphometric analysis.**
Results and conclusion: **Differences in wound healing were observed between treatments when compared to the control group. Microcurrent application alone or combined with *H. perforatum* gel or *A. montana* gel exerted significant effects on wound healing in this experimental model in all of the study parameters (*P* < 0.05) when compared to the control group with positive effects seen regarding newly formed tissue, number of newly formed blood vessels and percentage of mature collagen fibers. The morphometric data confirmed the structural findings. In conclusion, application of *H. perforatum* or *A. montana* was effective on experimental wound healing when compared to control, but significant differences in the parameters studied were only observed when these treatments were combined with microcurrent application.** *Homeopathy* (2012) **101**, 147–153.

Keywords: *Arnica*; *Hypericum*; Microcurrent; Wistar rat; Wound healing

Introduction

Tissue injuries trigger a cascade of events, including an inflammatory response and tissue formation and remodeling, which lead to at least partial reconstruction of the wounded tissue. Wound healing is a dynamic process in which a variety of extracellular matrix components act in concert to reestablish the integrity of injured tissue. As a consequence, the requirements for wound dressing can change as the healing progresses.[1]

Technological advances have permitted the emergence of a wide variety of wound healing treatments. The application of low amperage electrical stimuli modifies the healing process in living organisms, especially factors that delay or impair this process.[2] Animal studies using a variety of wound models and electrical stimulation protocols have reported an enhancement in some aspects of wound healing.[3–5] In this respect, microcurrent stimulation at 20 μA, which induces the flow of electrons into the skin

*Correspondence: Gláucia MT Santos, Av. Dr. Maximiliano Baruto, 500 Jd. Universitário 13607–339, Araras, São Paulo, Brazil.
E-mail: glauciasantos@uniararas.br
Received 24 February 2012; revised 16 May 2012; accepted 22 May 2012

and subcutaneous injury, has been shown to affect wound healing.[6] Other studies demonstrate concomitant increases in the number of fibroblasts within the wound, and an increase in collagen production.[7,8] Exogenous application of electrical stimulation has also been used in various *in vitro* and *in vivo* investigations on cellular activities in order to elucidate the effects of physiological levels of electricity on tissues.[3−5,9,10] The relationship between endogenous electrical activities and tissue injury and repair has been investigated in various areas of clinical practice.[11]

Another field of interest for the treatment of tissue injuries is the use of herbal and alternative remedies, which is increasing worldwide.[12−14] Alternative treatments other than the usual allopathic therapy have been used, including homeopathic treatment.[15,16] The principle of homeopathy involves the application of minimal doses since a dose-effect related response has been expected as usually observed for allopathic medications.[17]

Hypericum perforatum L. (family Hypericaceae), popularly known as St. John's wort, has long been used both orally and topically for wound healing in the folk medicine of various countries.[18] Extracts of *H. perforatum* are widely used to treat depression[19] and for centuries have been used as a medicinal herb. The current popularity of *Hypericum* comes from recent clinical trials and reviews on its clinical efficacy and pharmacological potential. *Hypericum* possesses a unique pharmacology in which it displays the pharmacology of many classes of antidepressants and new mechanisms not typical of standard antidepressants.[20] Moreover, it has been proposed to have antibacterial and antiviral effects and to exert anti-inflammatory and analgesic activity. *H. perforatum* extract contains flavonoids and phenolic acids, which demonstrated a free radical scavenging activity, exerting very efficient anti-inflammatory effects in animal model of acute inflammation.[21,22] Oral administration of *Hypericum* tincture has shown to increase the repair of incisions and excision in experimental studies on rats[23] and, in clinical studies, Samadi et al[24] demonstrated that *H. perforatum* facilitates cesarean wound healing.

Arnica montana is an ornamental perennial herbaceous plant with a creeping rhizome, characteristic of the family Asteraceae. Arnica is the classic homeopathic remedy for trauma of various kinds and the plant is widely known for its anti-inflammatory action.[25] The Homeopathic Pharmacy lists as its first indication 'trauma' and even recommends it as first-aid treatment.[26] It has also been used for myocarditis, cardiac insufficiency, arteriosclerosis, angina pectoris, and has demonstrated significant decreases in postoperative swelling and hematomas.[26−29]

In the present study, we investigated the effect of homeopathic concentrations of *Hypericum* and *Arnica* since they have been widely used in folk medicine.[30,31] The objective of this study was to evaluate the effects of microcurrent application alone or in combination with the topical administration of a gel containing *H. perforatum* L. or *A. Montana* L. on the healing of skin wounds surgically induced in Wistar rats.

Material and methods

The gels containing *H. perforatum* L. (3dH − 10%) and *A. montana* L. (3dH − 10%) were obtained from Botica Natural (pharmacy for compounding, homeopathy and natural products − pharmaceutical officer Dr José Carlos de Almeida − CRF: 87-221/Águas de Lindóia/SP). The gel consisted of 10% Natrosol, 0.2% Nipagin, 5% propylene glycol, and water qsp. The choice of these preparations was due to popular use in the treatment of wounds and trauma and also by the presence of antioxidant compounds important in tissue repair.[32]

Animals

Male Wistar rats (*Rattus norvegicus*) weighing 250−350 g, obtained from Prof. Dr Luiz Edmundo de Magalhães Experimental Animal Center, Uniararas, were housed individually in cages at a constant temperature (23 ± 2°C). The rats had free access to food and water and were maintained under a 12-h light/dark cycle. The average weight and behavior of the animals did not differ significantly between the beginning and the end of the study. The ***n*** for each experimental group was determined as a consequence of number of groups in order to minimize their suffering. This protocol was approved by the Ethics Committee of UNIARARAS (number 088/2006). The study followed the international guidelines on animal experimentation and biodiversity.[33−35]

Linear incision wound model

Each animal's back was depilated 48 h before surgical intervention. After local asepsis with 0.4% chlorhexidine digluconate, the animals were anesthetized by intraperitoneal injection of xylazine hydrochloride (20 mg/kg body weight) and ketamine hydrochloride (50 mg/kg). After the position was marked with a dermographic pen and pachymeter, a 2-cm long and 0.2-cm deep surgical incision was made with a sterile surgical blade through the full thickness of the skin in the craniocaudal direction in all animals as described by Mendonça et al.[3] The incision was not sutured. The animals received Sodium Dipyrone oral administration (drops/kg) in the first 24 h after induction of the wounds.

The animals were divided into seven groups of nine animals each and submitted to the following treatments: (1) simulated intervention (control group − C); (2) topical application of gel (N); (3) microcurrent application (10 μA/2 min) (MC); (4) topical application of gel containing *H. perforatum* (Hyp); (5) topical application of *H. perforatum* gel and microcurrent (10 μA/2 min) (Hyp + MC); (6) topical application of gel containing *A. montana* (Arn); (7) topical application of *A. montana* gel and microcurrent (10 μA/2 min) (Arn + MC).

A transcutaneous electrical stimulator (Physiotonus Microcurrent, Bioset, Rio Claro, São Paulo, Brazil) was used for electrical stimulation. The device was set to microgalvanic-continuous mode using an intensity of