

Comparison of Healing Effects of Propolis to Silver Sulfadiazine on Full Thickness Skin Wounds in Rabbits

Yesari ERÖKSÜZ¹
İbrahim CANPOLAT²
Sibel SİLİCİ³

¹ University of Firat
Veterinary Medicine
Department of,
Patology
Elazığ-TURKEY

² University of Firat
Veterinary Medicine
Department of,
Surgery
Elazığ-TURKEY

³ University of Erciyes
Safiye Çıkırcıoğlu High
School, Department of
Animal Health and
Production
Kayseri-TÜRKİYE

Geliş Tarihi : 04.01.2008
Kabul Tarihi : 04.02.2008

Yazışma Adresi Correspondence

İbrahim CANPOLAT
Firat Üniversitesi
Veteriner Fakültesi,
Cerrahi Anabilim Dalı
23119
Elazığ-TÜRKİYE

icanpolat@firat.edu.tr

The effects of propolis and silver sulfadiazine (SS) on the wound healing were determined using qualitative and quantitative parameters and histopathological findings. A total of 30 full thickness skin wounds, 3 from each of 10 rabbits, were created on their dorsal aspects. Of these wounds, 10 were allocated to group 1 (propolis), other 10 group 2 (SS) and other 10 control. The skin wound in the 1, 2 and 3 groups were covered daily with 50% propolis cream, SS skin cream and bepanthane cream (control), respectively. Postoperatively the wound surfaces were macroscopically examined and the healing process and the rates of wound expansion, contraction and epithelialization processes were quantitatively analyzed. As a result, propolis was found in general to have a better wound healing feature than others.

Key words: Wound healing, propolis, silver sulfadiazine, rabbit.

Tavşanlarda Tam Kalınlıkta Deri Yaralarında Propolis ve Silver Sulfadiazine'in Yara İyileşmesi Üzerine Etkilerinin Karşılaştırılması

Propolis ve silver sulfadiazine'in yara iyileşmesi üzerine etkileri kalitatif ve kantitatif parametrelerle ile histopatolojik bulgular değerlendirilerek saptandı. On tavşanın sırt bölgesinde üçer tam kalınlıklı toplam 30 tane yara oluşturuldu. Yaraların 10 tanesi propolis, 10 tanesi silver sulfadiazine ve 10 tanesi kontrol olarak gruplandırıldı. Yaralara her gün propolis %50 kremi, SS ve bepanthane kremi (kontrol) uygulandı. Postoperatif dönemde yara genişlemesi, kontraksiyonu ve epitelizeasyon kantitatif olarak analiz edildi. Sonuç olarak propolisin diğer gruplara göre daha iyi bir yara iyileşmesi sağladığı saptandı.

Anahtar Kelimeler: Yara iyileşmesi, propolis, silver sulfadiazine, tavşan.

Introduction

Propolis (bee glue- resinous material) is a mixture containing a number of plant products including flavonoids and is deposited in beehives by bees. Propolis has attracted the interest of many investigators because of its anti-bacterial (1, 2), anti-viral (3), anti-inflammatory, and immuno-stimulatory (4, 5) effects. Additional studies have shown propolis to be tumoricidal, antifungal, antiamebic, and antipyretic (6, 7). It was found that propolis has therapeutic effect on wound healing. Silver sulfadiazine (SS) is the topical agent and is used universally today in wound healing. The purpose of this study was to compare healing rates of wounds treated with propolis or silver sulfadiazine in rabbit model.

Materials and Methods

The dorsal aspects of 5 male and 5 female rabbits were clipped and prepared for aseptic surgery. All rabbits were anesthetized with intramuscular administration of 10 mg/ kg xylazine hydrochloride (Rompun, Bayer) and 50 mg/ kg ketamine hydrochloride (Ketanes-Alke). On each animal, two cranially and one caudally located square (4 cm²) full-thickness skin wounds were created using a template prepared from an X-ray film. These wound sites were allocated to three groups and covered with 50% propolis cream (Turkish propolis), silver sulfadiazine skin cream and bepanthane cream (control), respectively. These applications were repeated every day. A wound site was considered macroscopically to be fully healed when its whole surface was covered with epithelium. The wound boundaries were traced on a transparent sheets were scanned and the areas of wound sites and epithelization fronts were measured with the help of the paintbrush (PB) computer program. The data obtained were calculated as described by Kilic et al. (8). Briefly; area (cm²) = P/K.M, where P is the value of a particular wound site obtained on the PB, K is a rate constant for expressing the value of the PB as cm², and M is magnitude of tracings after scanning. During the examination, the day wound contraction started, the rate of wound contraction, the day epithelization and the number of days in which wound fully completed were evaluated.

The statistical comparison was made with ANOVA. The result were considered as significant $P \leq 0.05$. Ten and 20 days later, the rabbits were anaesthetized and the skin tissue biopsy samples were collected from rabbits for histopathological examinations. The samples were fixed in 10% neutral buffered formalin, and were cut into 5 μm sections and stained with hematoxylin and eosin (H&E), and examined microscopically (9).

Results

No mortality was seen in the animals during the study. Macroscopically, in all treatment groups, no difference was detected with regard to inflammatory response; heat, redness, and swelling. All wound sites was healed completely 17 days after surgery. The wound contraction and wound healing rates of propolis were higher than treatment and control groups (Figure 1), (Table 1).

Table 1. Changes in wound sizes of three groups during the healing periods (n=10).

Groups	Days of Operation		
	5	10	15
Propolis	3.5 ± 0.4^b	1.0 ± 0.2^b	0.4 ± 0.1^b
SS	3.7 ± 0.1^a	1.3 ± 0.1^a	0.6 ± 0.1^a
Control	3.8 ± 0.3^a	1.4 ± 0.2^a	0.6 ± 0.2^a

a,b,c : Aynı sütunda farklı harfleri içeren grup ortalamaları arası farklar önemlidir ($P < 0.05$)

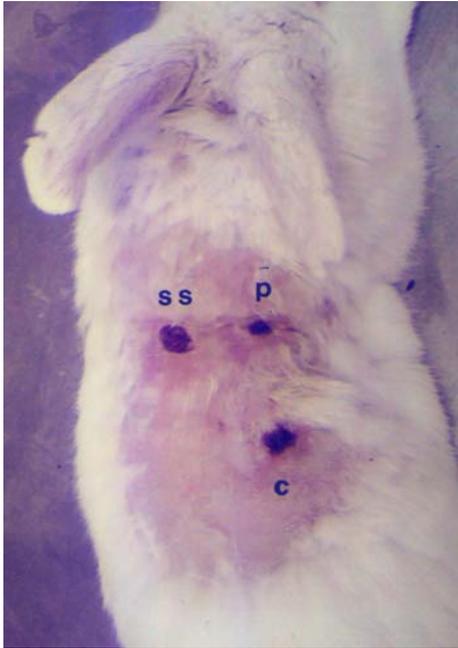


Figure 1. The wound healing rates of propolis (p) were higher than treatment (ss) and control (c) groups.

At the 10th day of experiment, histopathologically; there was inflammatory reaction including lymphocytes, macrophages and neutrophils and fibroblastic loose connective tissue in dermis of all groups examined. The severity of the reaction in propolis group was lower severe than SS and control group. In control group, inflammatory cells were abundant and completely filled the gap of wound. Connective tissue between the wound margins and surface debris were also more voluminous in control and SS groups than propolis group. At the 20th day of experiment, the epithelial and dermal healing was completed in propolis group and better than both control and SS groups. Both dermis and epidermis were completely regenerated in this group (Figure 2). The appearance of the skin was completely normal except for the decreased epidermal thickness. In SS and control groups, the surface was covered by scab and epidermal regeneration was not completed (Figure 3). In control group; dermal and epidermal regeneration was also retarded as compared to SS and propolis groups (Figure 4).

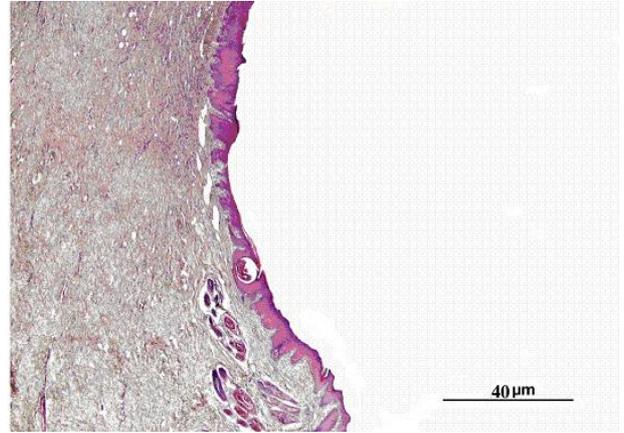


Figure 2. Complete dermal and epidermal regeneration characterized by full thickness of epithelial covering and non-vascular scar in dermis, in propolis group, H&E.

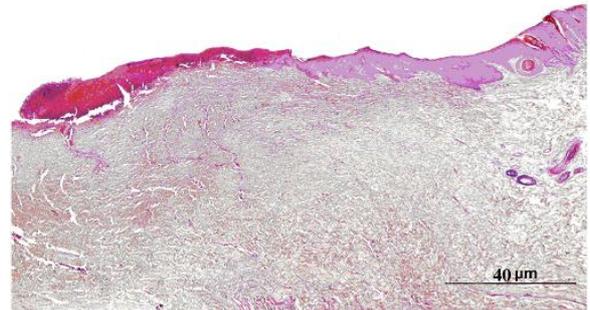


Figure 3. Complete dermal and incomplete epidermal regeneration with surface debris, in SS group, H&E.

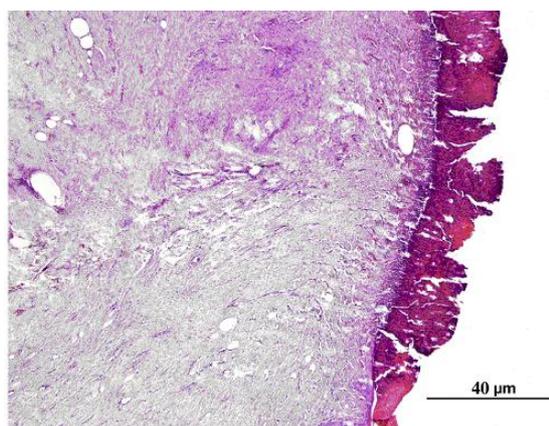


Figure 4. Incomplete dermal and epidermal regeneration characterized by capillary proliferation and crust formation, in control group, H&E.

Discussion

The wound healing process involves many complex factors. These may be classified as local factors, systemic factors and organ and species variability in

References

1. Castaldo S, Capasso F. Propolis, an old remedy used in modern medicine. *Fitoterapia* 2002; 73: 1-6.
2. Kartal M, Yıldız S, Kaya S. ve ark. Antimicrobial activity of propolis samples from two different regions of Anatolia. *J. Ethnopharmacol* 2003; 86: 69-73.
3. Kooa H, Gomesa BP, Rosalen PL et al. In vitro antimicrobial activity of propolis and Arnica montana against oral pathogens. *Arch Oral Biol* 2000; 45: 141-148.
4. Borrelli F, Maffia P, Pinto L, et al. Phytochemical compounds involved in the antiinflammatory effect of propolis extract. *Fitoterapia* 2002; 73: 53-63.
5. Khayyal MT, El-Ghazaly MA, El-Khatib AS Mechanism involved in the antiinflammatory effect of propolis extract. *Drug Exp Clin Res* 1993; 19: 197-203.
6. Grunberger D, Ganarjee R, Eisinger K et al. Preferential cytotoxicity on tumor cells by caffeic acid phenethyl ester isolated from Propolis. *Experientia* 1980; 44: 230-232.
7. Park YK, Koo MH, Abreu JA et al. Antimicrobial activity of Propolis on oral microorganisms. *Curr Microbiol* 1998; 36: 24-28.
8. Kılıç, S, Timurkaan, N, Unsaldı, S ve ark. Comparison of Effects of Some Wound Healing Materials on Full Thickness Skin Wounds in Rabbits. *Turkish Journal of Veterinary and Animal Sciences* 2002; 26: 263-272.
9. Luna LG: *Manuel of Histologic Staining Methods of Armed Forces Institute of Pathology*. McGraw-Hill Book Company, New-york, 222, 226, 1968.
10. Subrahmanyam M A prospective randomised clinical and histological study of superficial burn wound healing with honey and silver sulfadiazine. *Burns* 1998; 24: 157-161.
11. Nagaia T, Sakai M, Inoue R et al. Antioxidative activities of some commercially honeys, royal jelly, and propolis. *Food Chem* 2001; 75: 237-240.
12. Gracia CG An open study comparing topical silver sulfadiazine and topical silver sulfadiazine-cerium nitrate in the treatment of moderate and severe burns. *Burns* 2001; 27: 67-74.
13. Gregory RS, Piccolo N, Piccolo MT et al. Comparison of Propolis Skin Cream to Silver Sulfadiazine: A Naturopathic Alternative to Antibiotics in Treatment of Minor Burns. *J Altern Complem Med* 2002; 8: 77-83.

response to injury. Topical medications should provide a specific desired effect during the appropriate stage healing (8). In this study, we adopted an experimental design including macroscopical and histological evaluations of the wound healing process. Propolis is a natural drug that has been employed extensively since ancient times (1, 5, 10, 11). Khayyal et al. (5), reported that aqueous propolis extract possesses significant anti-inflammatory properties and has successfully reduced oedema in both acute and chronic models of inflammation.

The propolis skin cream appears to have a beneficial effects on healing burn wounds (10, 12, 13). It was also reported that propolis has a therapeutic effect on wound healing, inflammations of the skin and other skin lesions (1, 4). In this study, all wound sites was healed in 17 days after surgery. The wound contraction and wound healing rates of propolis were higher than other groups.

According to the results of the present study, including macroscopical, qualitative, and microscopical analyses propolis has a higher healing performance than other groups.

