*Harran Üniv Vet Fak Derg, 2020; 9 (2): 194-199* *Research Article*
**DOI:10.31196/huvfd.817720**

# Investigation of the Clinical Effectiveness of Polyhexanid/Polyhexamethylene Biguanide Wound Dressing in Chronic Cases

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**Geliş Tarihi: 28.08.2020** **Kabul Tarihi: 02.11.2020**

**Abstract:** Polyhexanid/polyhexamethylene biguanide is one of the most used wound antiseptics, with a high tissue compatibility and low cytotoxicity. In the present study, it was aimed to investigate the effectiveness of antimicrobial gel and spray that containing 0.1% PHMB, on wound healing in the treatment of patients who come to our clinic with chronic infected wound formation due to different reasons. The materials of the study consisted of 12 cats and 8 dogs with chronic, infected, ulcerative and suppurative wounds in various parts of the body, which were treated at the Surgery Clinic of SU Faculty of Veterinary Medicine. In the evaluation of the wound bed, the color, depth, shape, size of the wound, type and amount of the exudate, localization of the wound, and wound care conditions were taken into consideration. Surgical or mechanical superficial debridements of the wound were performed initially. Following this procedure, irrigation of the wound area was achieved with 0.9% saline. Protective dressing was applied to the wound area of all patients by applying antimicrobial (Actolind® w Gel, ACTO, Germany) spray containing 0.1% PHMB twice a day, and gel of the same product after 5 minutes. When the wound bed of the cases were evaluated on the 15th day, (100%) granulation and epithelization occurred in all the wounds of 19 patients, and only in one patient epithelization was observed. At the end of the 30th day, it was noted that granulation was completed in all cases except one case and passed to the epithelization stage. In all clinical cases, no complications were encountered during the treatment process. Clinical studies of polyhexanide/polyhexamethylene biguanide are promising. In this study, positive response was obtained in a short time. However, more experimentally controlled clinical and laboratory studies are required in order to improve 0.1% PHMB applications in chronic wound treatments.
***Keywords:*** *Antiseptic, chronic wound, infected wound, polyhexanide/polyhexamethylene biguanide.*

## Kronik Olgularda Poliheksanid/Polyheksametilen Biguanid Yara Örtüsünün Klinik Etkinliğinin Araştırılması

**Özet**: Poliheksanid/poliheksametilen biguanid (PHMB), doku uyumluluğu son derece yüksek, "sitotoksisite" derecesi düşük ve günümüzde en sık kullanılan yara antiseptiklerinden biridir. Sunulan çalışmada; farklı sebeplere bağlı olarak kronik enfekte yara oluşumu ile kliniğimize gelen olguların tedavisinde % 0,1 PHMB içeren antimikrobiyal jel ve spreyin yara iyileşmesi üzerine etkinliğini araştırmak amaçlanmıştır. Çalışmanın materyalini; S.Ü. Veteriner Fakültesi Cerrahi Kliniği'ne vücudun çeşitli bölgelerinde kronik, enfekte ve ülseratif yara şikayeti ile getirilen 12 kedi ve 8 köpek oluşturdu. Yara yatağının değerlendirilmesinde; yaranın rengi, derinliği, şekli, büyüklüğü, eksudat tipi ve miktarı, yaranın lokalizasyonu ve yara bakım şartları göz önünde bulunduruldu. Yaranın şirurjikal ya da mekanik yüzeysel debritmanı yapıldı. Bu işlemin ardından %0,9'luk serum fizyolojik ile yara bölgesinin irrigasyonu sağlandı. Bütün hastaların yara bölgesine günde iki kez olmak üzere önce %0,1 PHMB içeren antimikrobiyal (Actolind® w Solution, ACTO Pharma, Germany) sprey, 5 dakika sonra aynı ürünün jeli uygulanarak koruyucu pansuman yapıldı. Yara yatağı 15. günde değerlendirildiğinde 19 hastanın yaralarında (%100) granülasyon ve epitelizasyonun oluştuğu, bu süre içerisinde sadece 1 hastada epitelizasyon gözlendi. 30. gün sonunda 1 olgu dışında bütün olgularda granulasyonun tamamlanıp epitelizasyon aşamasına geçtiği kaydedildi. Klinik olguların tamamında tedavi sürecinde herhangi bir komplikasyonla karşılaşılmadı. Poliheksanid/poliheksametilen biguanid'in klinik çalışmaları umut vericidir. Bu çalışmaya dahil olan olgularda da kısa süre içinde olumlu geri dönüş alınmıştır. Fakat, kronik yara tedavilerinde %0,1'lik PHMB uygulamalarının geliştirilebilmesi için daha fazla kontrollü deneysel ve klinik çalışmalar ile detaylı laboratuvar değerlendirmelerine ihtiyaç vardır.
***Anahtar Kelimeler:*** *Antiseptik, enfekte yara, kronik yara, poliheksanid/poliheksametilen biguanid.*

## Introduction

The wound is the loss of anatomical and functional continuity of living tissue with impaired skin integrity. In veterinary medicine, the wound generally occurs due to traffic accidents, falls, sharp and piercing tools, gun injuries and bite (Murphree, 2017). Normal wound healing usually follows a series of well-managed repair processes. Complex biochemical pathways and cellular interactions allow hemostasis, inflammation, proliferation, and maturation phase to progress in

order (Bayındır, 2007). Many local and systemic factors that affect wound healing. Local factors are infection in the wound area, insufficient blood circulation, hypoxia, tissue necrosis, the presence of foreign particles, recurrent traumas, and mobility of the wound area. Systemic factors are diseases such as nutritional deficiency, diabetes, chronic renal failure, immunodeficiency, and corticosteroid use, age, and genetic structure of the patient (Hanna and Giacopelli, 1997; Robson et al., 2001). While many wounds heal without problems, with the prolongation of the tissue repair period, healing problems occur in many chronic wounds and open wounds are observed. The loss of the ability of a chronic wound to heal indicates a disruption somewhere in the natural stages of wound repair (Moore et al., 2006). The healing of open wounds often stops during the inflammatory phase or proliferative stage. The wound area that does not heal for a long time creates an excellent culture medium for microorganisms (Woo et al., 2007).

It appears that infection of the wound can cause pain and discomfort to the patient by prolongation of the inflammatory phase of wound healing and can lead to serious and potentially fatal systemic sepsis unless properly treated (Consensus Panel, 2010). The use of only systemic antibiotics in wound treatment is often insufficient for wound healing. Topical antimicrobial agents are generally alcohol-based, iodine-based, biguanides: chlorhexidine gluconate and polyhexanide/polyhexamethylene biguanide (PHMB), halophenols (chloroxilenol), bisphenols (triclosan), silver compounds, and oxygenated water. The use of topical antimicrobials in the treatment of wound infections has been considered as an alternative to systemic antibiotics to minimize parenteral antibiotic use (O'Dwyer and Demetriou, 2016). PHMB is an antiseptic that has been used for more than 60 years in various industries and relatively new to use in veterinary medicine. PHMB is highly effective as antimicrobial by destroying the outer and cytoplasmic membranes of bacteria (O'Dwyer and Demetriou, 2016). PHMB is one of the most used wound antiseptics, with high tissue compatibility and low cytotoxicity (Wehner et al., 2009). PHMB has a broad spectrum of activity against bacteria, viruses, and fungi (Moore and Gray, 2007) and is recommended as therapeutic alternatives to antibiotics (Hancock and Sahl, 2006).

This study aimed to investigate the effectiveness of antimicrobial gel and spray containing 0.1% PHMB on wound healing in the treatment of patients who were brought to our clinic with chronic infected wound formation due to different reasons.

## Materials and Methods

The materials of the study consisted of 12 cats and 8 dogs with chronic, exudative, and suppurative wounds in various parts of the body, which were treated at the Surgical Clinic of SU Faculty of Veterinary Medicine. There was no age, race, and gender limit in patients. After clinical examination and blood analysis of all patients, wound beds were evaluated. In the evaluation of the wound bed, the color, depth, shape, size of the wound, type and amount of the exudate, localization of the wound, and wound care conditions were taken into consideration. Firstly, the periwound area was shaved. Then, according to the condition of the wound, surgical or mechanical superficial debridements were performed. Following this procedure, irrigation of the wound area was applied with 0.9% saline. After irrigation, the wound was dried with a sterile gauze and an antimicrobial (Actolind® w Solution, ACTO Pharma, Germany) spray containing 0.1% PHMB was applied to the wound area twice a day, and protective dressing was applied by gel of the same product after 5 minutes. Four cases were monitored in the hospital. Some of the patients with ownership were brought to the hospital daily and followed up by dressing and bandaging.

The ways of using drugs were described to other clients, and they were called for control periodically. Also, cefazolin was injected to the cases (30 mg/kg, i.m., Iespor®, İbrahim Etem Ulagay İlaç Sanayi Türk A.Ş., Istanbul, Turkey), a parenteral broad-spectrum 1st generation cephalosporin group antibiotic for 7-14 days for antimicrobial therapy. During the treatment period, corticosteroids were not injected. The cases were kept under control until the wound healed. Clinical observations were performed on days 0, 15, and 30 day the type of the wound and its underlying etiology, localization of the wound, the type and color of the tissue in the wound bed, the state of infection/inflammation, the amount and color of the exudate, the wound edges and the condition and epithelization of the surrounding skin were recorded (Table 1). These evaluations were made as described by Gray et al., (2006).

***Tissue type in the wound bed****:* Necrotic (dead tissue), fibrin, exudative (purulent/purulent discharge), granulation tissue, epithelization tissue.

***Infection/inflammation status of the wound****:* Purulent and green exudation was considered as infective; when the exudate was stopped and the

wound began to turn red it was considered as inflammatory (Gray et al., 2006).

***Tissue color in the wound bed:*** Necrotic tissues were evaluated as black, fibrinous tissues yellow-yellowish, suppurative tissues with purulent discharge green, granulation tissues red, epithelization tissues pink and mixed color.

***Evaluation of exudate type and amount:*** İt was evaluated as; dry/less; no exudate (-), moderate/moist; excess exudate discharge (++) and excessive/wet; excessive exudate discharge (+++). The exudate type was assessed and recorded as serous, seromucous, serosanguineous, hemorrhagic, fibrinous, and purulent.

***Measuring wound surface and depth:*** The percentage of wound contraction for wounds surface were calculated and noted on days 0, 15, and 30 by the following formula, as described by Kumar et al. (2006): % wound contraction on day X = (area on day 0 - open area on day X/area on day 0) × 100. Although this method is the simplest, it should be known that it is not the exact result (Table 1).

## Results

**Tissue type in the wound bed:** When tissue types in the wound beds were evaluated, 19 of 20 cases (95%); necrotic, fibrinous, and suppurative appearance on day 0, were detected as necrotic and fibrinous in one case (5%). When the wound bed was evaluated on day 15 granulation and epithelization occurred in all of the wounds of 19 patients (100%), and granulation was observed only in one patient (Table 1). At the end of the 30th day, epithelization (keratinocyte) was recorded in all cases (100%).

**Infection findings:** When the status of the lesions was evaluated on day 0, 95% of the cases (19 cases) were infected, 5% (one case) were inflammatory; on day 15, 100% were inflammatory (Table 1) and on day 30 both findings were disappeared 100%.

**Discoloration in the wound:** On day 0, the color of the wound was detected as mixed in 35% of cases (in 7 cases), yellow-yellowish and green in 30% (in 6 cases), yellow-yellowish, green and red in 10% (2 cases), red in 5% (1 case), yellow-yellowish and red in 5% (1 case) and black, yellow, and red in 15% (3 cases). On day 15 the wound color was determined as red-pink in 95% (19 cases) and pink in 5% of the cases (1 case) (Table 1).

**Exudate type and amount findings:** The exudate type and amount are compared between days, on day 0, excessive exudate with fibrinous and purulent character in 35% (7 cases) of the cases, excess exudate with fibrinous and serosanguinous character in 10% (2 cases), in 35% of the cases (in 7 cases) excessive exudate with fibrinous purulent serosanguinous character, in 10% of cases excessive exudate with fibrinous purulent and hemorrhagic character, in 5% of the cases (1 case) excessive exudate with fibrinous, purulent, serous and hemorrhagic character, and 5% of the cases less exudate with hemorrhagic serosanguinous character was determined. While the exudate type was hemorrhagic serosanguineous in 90% of the cases on the 15th day, it was evaluated as hemorrhagic in 10% of the cases. During this period, excess exudate was detected in 25% of the cases (Table 1). On the 30th day of the evaluation, only 5% of cases had exudation and 95% of the cases did not have any exudation.

**Results of measuring wound surface and depth:** The total size of the wound was 592.05 $cm^2$ on day 0, 197.8 $cm^2$ on day 15, and 3.5 $cm^2$ on day 30. The reduction in wound size was 67% on day 15 and 98.2% on day 30. While the decrease in the size of the postoperative lesions (7 cases) was 58.88% on day 15 98.49% on day 30 the decrease in the wound size of 3 patients with suspected osteomyelitis was recorded as 55.84% on day 15 (Table 1) and 100% on day 30. In all clinical cases, no complications were encountered during the treatment process. All the cases recovered for a certain period; 19 cases in 30 days, 1 case in 45 days.

## Discussion

Non-healing wounds and wounds that have secondary infections, mostly caused by drug-resistant bacteria, are common problems for veterinarians. Normal wound healing occurs in three interrelated stages, such as hemostasis, proliferation, and maturation. However, ulcerative wounds and burns that develop on the skin for various reasons often delay the healing process. Therefore, both topical antimicrobials and systemic antibiotics are used to accelerate the healing of infected ulcerative wounds (O'Dwyer and Demetriou, 2016).

PHMB, which we discussed in this study, is a topical agent that its antiseptic/antimicrobial activity on wound care has been proven by experimental and clinical studies. The mechanism of action of PHMB is, initially to attach to the positively charged surface of the bacteria, and then penetrate to the internal cytoplasm of the bacteria through the cytoplasm membrane, leading to cell death by disrupting the integrity and permeability of the phospholipid structure (Lee et al., 2004).

**Table 1.** The condition of the tissues in the wound bed on the 0th, 15th and 30th day.

| 0th Day | | | | | | | 15th Day | | | | | | | 30th Day | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Case | Tissue | Infection/Inflammation | Exudate type and amount | The color of the tissues | The size of the wound | Epitelization | Case | Tissue | Infection/Inflammation | Exudate type and amount | The color of the tissues | The size of the wound | Epitelization | Case | Tissue | Infection/Infla | Exudate type and amount | The color of the tissues | The size of the wound | Epitelization |
| 1 | N, F, S | Infected | ++ F,P | Mix | 12.5 $cm^2$ | - | 1 | G,E | Inflammatory | + H,SS | R,P | 5.75$cm^2$ | +++ | 1 | E | | | P | | +++ |
| 2 | N,F,S | Infected | +++ F,P | Y, G | 10 $cm^2$ | - | 2 | G,E | Inflammatory | + H,SS | R,P | 2.25$cm^2$ | ++ | 2 | E | | | P | | +++ |
| 3 | N,F,S | Infected | ++ F,P | Y, G, R | 18 $cm^2$ | - | 3 | G,E | Inflammatory | + | P | - | +++ | 3 | E | | | P | | +++ |
| 4 | N,F,S | Infected | +++ F,P | B, Y,R | 21 $cm^2$ | - | 4 | G,E | Inflammatory | ++ H,SS | R,P | 12 $cm^2$ | + | 4 | E | | | P | | +++ |
| 5 | N,F | Inflammatory | ++ F,SS | B,Y, R | 23.5$cm^2$ | - | 5 | G,E | Inflammatory | ++ H,SS | R,P | 13.25$cm^2$ | + | 5 | E | | | P | | +++ |
| 6 | N,F,S | Infected | + H,SS | R | 2 $cm^2$ | - | 6 | E | Inflammatory | + | R,P | - | +++ | 6 | E | | | P | | +++ |
| 7 | N,F,S | Infected | +++ F, P, H | Mix | 12 $cm^2$ | - | 7 | G,E | Inflammatory | ++ H,SS | R,P | 8 $cm^2$ | + | 7 | E | | | K,P | 3.5 $cm^2$ | +++ |
| 8 | N,F,S | Infected | +++ F, P, SS | B,Y,G | 12.5 $cm^2$ | - | 8 | G,E | Inflammatory | ++ H,SS | R,P | 5.5 $cm^2$ | + | 8 | E | | | P | | +++ |
| 9 | N,F,S | Infected | +++ F,P,SS | Y,G | 8 $cm^2$ | - | 9 | G,E | Inflammatory | + H | R,P | - | ++ | 9 | E | | | P | | +++ |
| 10 | N,F,S | Infected | ++ + F,P,SS | Mix | 15 $cm^2$ | - | 10 | G,E | Inflammatory | + H,SS | R,P | 3.5 $cm^2$ | ++ | 10 | E | | | P | | +++ |
| 11 | N,F,S | Infected | ++ F, SS | Y, R | 6 $cm^2$ | - | 11 | G,E | Inflammatory | + H,SS | R,P | - | +++ | 11 | E | | | P | | +++ |
| 12 | N,F,S | Infected | +++ F,P,S,H | Mix | 24.8 $cm^2$ | - | 12 | G,E | Inflammatory | + H,SS | R,P | 8.25$cm^2$ | ++ | 12 | E | | | P | | +++ |
| 13 | N,F,S | Infected | +++ F,P | Y, G | 15 $cm^2$ | - | 13 | G,E | Inflammatory | ++ H,SS | R,P | 3.75$cm^2$ | + | 13 | E | | | P | | +++ |
| 14 | N,F,S | Infected | +++ F,P,SS | Mix | 139 $cm^2$ | - | 14 | G,E | Inflammatory | + H,SS | R,P | 52.5 cm | ++ | 14 | E | | | P | | +++ |
| 15 | N,F,S | Infected | +++ F,P,SS | Y,G | 13 $cm^2$ | - | 15 | G,E | Inflammatory | + H,SS | R,P | - | +++ | 15 | E | | | P | | +++ |
| 16 | N,F,S | Infected | + ++ F,P,SS | Y,G | 42 $cm^2$ | - | 16 | G,E | Inflammatory | + H,SS | R,P | 12cm | ++ | 16 | E | | | P | | +++ |
| 17 | N,F,S | Infected | +++ F,P,SS | Mix | 96 $cm^2$ | - | 17 | G,E | Inflammatory | + H,SS | R,P | 32 $cm^2$ | ++ | 17 | E | | | P | | +++ |
| 18 | N,F,S | Infected | ++ F, P | Y, G | 65 $cm^2$ | - | 18 | G,E | Inflammatory | + H,SS | R,P | 26.05$cm^2$ | ++ | 18 | E | | | P | | +++ |
| 19 | N,F,S | Infected | +++ F, P, H | Y, G, R | 18 $cm^2$ | - | 19 | G,E | Inflammatory | + H,SS | R,P | 3 $cm^2$ | ++ | 19 | E | | | K,P | | +++ |
| 20 | N,F,S | Infected | ++ F, P, | Mix | 38.5 $cm^2$ | - | 20 | G,E | Inflammatory | + H,SS | R,P | 5$cm^2$ | ++ | 20 | E | | | P | | +++ |

Table 1. N: necrotic, F: fibrinous, S: suppurative, G: granulation tissue, E: epithelialization tissue. The color of the tissues in the wound bed is; R: red, epithelialization tissues P: pink Mix: mixed color. Exudate type and amount was evaluated as dry/less; no exudate (-), moderate / moist; excess exudate discharge (++) and excessive/wet; excessive exudate discharge (+++). The exudate types. S: serous, SM: sero-mucous, SS: serosanguineous H: hemorrhagic, F: fibrinous and P: purulent.

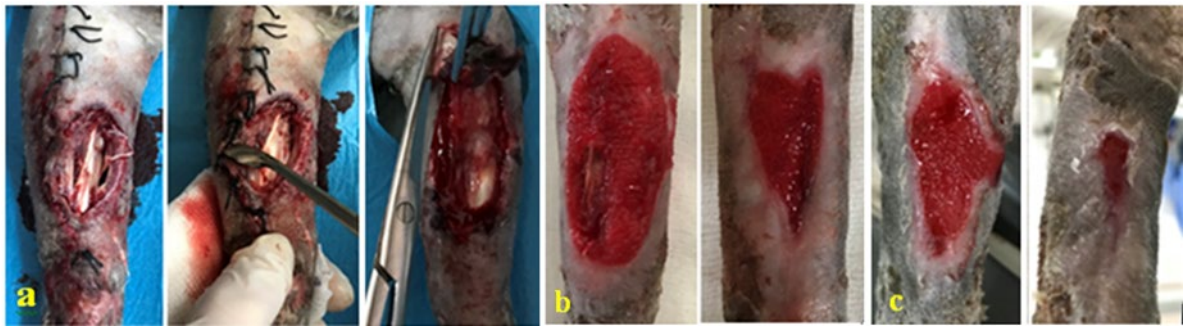


**Figure 1.** (Case 5) Postoperatively infected, necrotic and exudative wound in this case of fragmented, closed fracture at radio-ulna diaphysis of a cat, a) debridement of the wound with sharp tool and wet-dry dressing on day 0, b) Medial and lateral views of the wound healing by wet-dry dressing with PHMB on day 15; c) Medial and lateral views of the wound on the 30th day of treatment.

Cazzaniga et al. (2002) used a 0.2% PHMB impregnated gauze in the wound created by *Pseudomonas aeruginosa* in a pig model. In treated wounds with 0.2% PHMB-gauze, bacterial colonization is significantly reduced or eliminated after 72 hours. In the study, they note that the biguanide impregnated dressing can be used to prevent the proliferation of bacterial pathogens and can be particularly useful in preventing bacteria from entering open, effusive wounds. In this clinical study, showing the same results as the study mentioned above, no symptoms of infection and exudate were observed in all wounds on day 15 of 0.1% PHMB treatment in 95% of the patients who were healed. In addition, cessation of exudation in the wounds on day 15 was evaluated as evidence that the microbial environment in the wound disappeared and PHMB treatment showed close to 100% success in chronic wound healing.

As is known from previous studies, the presence of necrotic or devitalized tissues on the wound surface prevents the evaluation of the destructed tissue area and reduces the penetration of local antiseptic agents (Hendrickson, 2012; Weir et al., 2018). For this reason, topical PHMB antiseptic was applied after the superficial mechanical debridements of the necrotic tissues were provided in the lesions of all cases included in the study. However, unlike other antiseptics, PHMB's antimicrobial activity is not impaired in wound fluid, or with a high amount of blood or albumin. This feature increases the importance of PHMB in clinical use. Also, after the PHMB is applied to the area, it binds to the cellular surfaces in about 5 minutes for the full antiseptic effect to occur and can last for hours (Harbarth, 2006). Considering this feature of PHMB, in this clinical study, a protective bandage was applied 5 minutes after applying the antimicrobial spray containing 0.1% PHMB (Actolind® w Gel, ACTO, Germany).

Lee et al. (2004), to eliminate bacterial pathogens in the laboratory environment, determined that PHMB impregnated gauzes were more effective in inhibiting the growth of all gram-positive bacteria in agar plates than the control group. In the present study, although no antibiogram was performed, the clinical results obtained in a short time were interpreted as the antimicrobial gel containing 0.1% PHMB and spray prevented the colonization of the pathogens in the wound area.

In another clinical human study conducted by Mulder et al. (2007), PHMB was applied in 26 wound treatments in total. They detected a decrease in the wound size of eight patients from an average of 6.79 $cm^2$ to 4.57 $cm^2$ on average of day 25. In the first week of our study, although the wounds were quite exudative, necrotic, fibrinous, and suppurative, these findings were seen to disappear in all wounds on day 15 of PHMB treatment. Also, the size of the wound shrank from 592.05 $cm^2$ on day 0 to 3.5 $cm^2$ on day 30 is an indication that PHMB provides effective recovery in chronic wounds due to various reasons. In 1 of 20 cases in total, the wound healing process of the wound due to plate osteosynthesis was delayed.

In wounds with tissue loss, the closure of wound areas begins with contraction. This happens as the wound edges surrounding the wound gradually cover the wound as a result of the movement towards the center. The myofibroblasts which are originating from fibroblastic cells in granulation tissue drag the collagen fibers along the wound tension line to allow the wound edges to move towards the wound center and the wound area to shrink. In wounds with tissue loss, it may be seen that the contraction begins in the wound approximately 5–9 days later (Swaimve et al., 2001; Hendrickson, 2006). The red color in the wound is an indication that the new granulation tissue has developed and epithelialization has begun on it (Gray et al., 2006).

As a result of using PHMB impregnated gauze in ulcer treatments, the ulcer was completely covered with healthy granulation tissue on day 15 (Fumarola et al., 2010). In another study, the completion of the granulation stage of the ulcer on day 18 as a result of topically use of the PHMB (Glover and Wicks, 2009) has led many researchers to think that low concentrations of PHMB can have a positive effect on the proliferation of human keratinocytes.

However, there are no proven studies related to this feature of PHMB. According to our observations in this study, incompatible with other studies; becoming red in color of the lesions on day 15 of the 0.1% PHMB application, shrinking of the wound from the periphery to the center, and decreasing its depth, were interpreted as occurring of the granulation tissue. On day 15 of our study, wound epithelization was found to be advanced in 15 cases. On day 30, it was interpreted that wound healing was completed in 19 of 20 cases, where all wounds were covered with epithelialization tissue. During this period, the epithelization process was prolonged in only one case. In all cases, the pink color of the wound was accepted as an indication that the wound has reached the final stage of wound healing when it began to be covered with new epithelium (Boateng et al., 2008).

The fact that PHMB is effective against *Staphylococcus aureus*, which is the biggest cause of non-healing wound contamination in hospitals, will provide faster results on wound therapy.

Clinical studies of PHMB are promising. In this study also, positive response feedback was obtained in a short time. However, more controlled experimental clinical and laboratory studies are needed to improve the 0.1% PHMB applications in chronic wound treatments and to determine their superiority over other antimicrobial dressings.

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