Experimental reconstruction of teat cutaneous wound by ear skin graft in dairy cattle

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BACKGROUND: Teat lacerations are serious injuries that endanger the survival of a cow within the herd. In excessive lacerations, the wound should be left to heal by second intention healing process. However, tissue contraction during healing can change the alignment of the teat, creating problems for machine milking. OBJECTIVES: The purpose of the present study was to evaluate the transplantation of the full-thickness free skin graft in excessive teat lacerations. METHODS: Four healthy, non-pregnant, mixed Holstein-Friesian dairy cows with normal teats were used in the study. Under local anesthesia, a rectangular full thickness excision (2 Cm×2 Cm) was made on the lateral surface of the proximal part of the right and left fore teats of each cow, and simultaneously a rectangular full-thickness free skin graft (1.5 Cm×1.5 Cm) was harvested from the inner surface of the left pinna. The graft was then meshed and transplanted to the left teat defect. During a three-week study, the wound healing was evaluated clinically and at the end of the study, histopathologic samples (H &E staining) were taken from both wounds. RESULTS: The results of this study demonstrated that in non-grafted wounds, severe wound contraction caused a deformity in normal anatomical structure of the teats. However, in grafted wounds successful graft take was recorded in all wounds, and the teats restored their functional and anatomical structure with satisfactory cosmetic appearance. CONCLUSIONS: According to the result of present study, it can be concluded that using full-thickness free mesh skin graft can be considered a suitable method for treatment of excessive teat lacerations.

Introduction

Because of physiological and anatomical peculiarity of a cow's teat, it is exposed to more rigorous manipulation and a greater risk of injury than any other structure of the animal's body (Ghamsari et al., 1996). Teat wounds are common around parturition as a result ofudder size and clumsy movement of a cow, and there is a higher incidence in animals aged five or six years, i.e. the start of maximum yield (Ghamsari et al., 1995; Ghamsari et al., 1996). Most teat injuries occur within one month of calving, when cows are in high production (Bristol, 1992). Injuries to the teat can be caused by various factors, including trauma, infection, husbandry practices and environmental factors (Molaei et al., 2002). Treatment of teat laceration is economically significant to dairy producers. After teat injury, the incidence of mastitis in the affected quarter increases by 50% (Bristol, 1992, Modransky and Welker, 1993). Most teat injuries occur when cattle are housed (78%) than when they are kept in pasture (21%). Although some
studies have reported that the hind teats are more frequently affected than the fore teats, the opposite observation has also been reported. The incidence of injury to the right and left sides is approximately equal (Bristol, 1992). Teat lacerations are rarely clean, straight and incised wound. Many are flap wounds with irregular shapes and depth of penetration. Cleaner wounds may be sutured successfully, and favorable recoveries usually occur in those cases in which there is minimal delay between injury and surgical treatment (Honey, 1984).

Degloving injuries result when most of the skin of the teat is removed. Such injuries usually occur when the teat is stepped on while the cow attempts to rise. In these wounds, after debridment of necrotic and infected tissues, it may be possible to treat the remaining defect with good functional and cosmetic results (Steiner, 2004; Bristol, 1992). Beware that large amounts of scar tissue or flaps of skin may interfere with milking or have undesirable cosmetic effects (Johnson, 1988). If reconstruction of a teat is not possible, the teat may be amputated as a salvage procedure (Steiner, 2004). If full-thickness lacerations are left to granulate rather than sutured, 60% to 75% of them will heal by second intention, with fistula formation (Hull, 1995).

Skin graft is the transfer of a segment of free dermis and epidermis to a distant recipient site. It is used for defects that cannot be reconstructed by direct apposition or skin flap (Ghamsari et al., 1996).

Since it is not always possible for large teat skin wounds to be repaired by first or second intention healing process, the present study was designed for the first time to investigate the reconstruction of teat cutaneous wound by ear free skin autograft in cows.

Materials and Methods

Four healthy, non-pregnant, lactating mixed Holstein-Friesian dairy cows, 3-6 years old, with normal teat were used in the study. The animals were fasted 24 hours preoperatively in order to reduce ruminal distention during the surgical procedures. The cows were sedated, with 2% xylazine hydrochloride (Alfasan, Woerden-Holland) at a dose of 0.08 mg/kg IV, and restrained in dorsal recumbency. The mammary gland, teats and inner surface of the left pinna were aseptically prepared and draped. The teat was anaesthetized by subcutaneous infiltration of 5-10 mL 1% lidocaine hy drochloride (Pasture, Tehran-Iran), at the base of the teat in the form of an inverted V block, and the inner skin of pinna was anaesthetized by subcutaneous infiltration of 5 mL 1% lidocaine hydrochloride, at the base of the pinna in the form of a line block. Rectangular full thickness skin defect (2 Cm×2 Cm) was created on the lateral surface of the proximal part of the right and left fore teats (Figure 1). After control of hemorrhage, the right teat wounds were left open, without graft. To harvest the free skin graft, a rectangular full thickness incision (1.5 Cm×1.5 Cm) was created on the inner surface of the left pinna; the skin was released, and then meshed. The meshed skin was then expanded and grafted to the left teat defect. The mesh graft was fixed on the wound bed, using naylon 3.0 (Supa, Tehran-Iran) and interrupted suture pattern (Figure 2). Both wounds were bandaged with a protective, non-pressured and non-adherent dressing. The dressing was left in place for three days; then, removed gently, followed by irrigation with saline-soaked gauze sponges and rebandaged. The process was repeated three times a week. The defect of pinna was left to heal by second intention wound healing. Penicilin-Streptomycine (20000 IU/kg - 20 mg/kg, i.m. bid) was administered preoperatively and continued for 5 days after the surgery.

During the study, immediately after changing the bandages, the teat wounds were evaluated clinically. For microscopic studies of wound healing process, under local anesthesia with lidocaine hydrochloride 1%, tissue specimens (0.5 Cm×0.5 Cm), were taken from right and left fore teats wounds, 3 weeks after the surgery. The obtained samples from the skin graft (left wound) or wound beds and underlying tissue (right wound), surrounded by margin of normal skin, were fixed in 10% buffered formalin. The samples were embedded in paraffin, sectioned at 4 ?m thickness, and stained with hematoxylin and eosin (H&E) for light microscopy.

Results

Clinical evaluation: During the 3-week study, we did not observe any complications such as infections in the wound sites of both groups.

The right teat wounds (without graft) showed an
Increase in size in the first days after the operation. After the initial enlargement, wound areas decreased rapidly in size during the study. This decrease was due largely to rapid wound contraction. Wound contraction caused a deformity in the normal anatomical structure of the teats after 3 weeks (Figure 3).
In the left wound, good graft contact with wound bed occurred and graft necrosis did not happen in none of the cases of the present study. 3 days after the surgery, swelling, edema and excessive exudation were observed, which gradually diminished in the wound during week 1. In all left wounds, the teats restored their normal structure with minimal wound contraction at the end of the study (Figure 4).

**Histopathological evaluation:** Three weeks after surgery, histologic examination of biopsy specimens revealed that the wounds that were grafted developed less granulation tissue, edema, and neovascularization (Figure 5). The regeneration of the epidermis was performed completely. The number of fibroblasts and endothelial cells in the subepidermal tissues had decreased. The collagen fibers were organized similar to normal surrounded tissues. The wound that did not receive any graft, showed lower organization of collagen fibers, more neovascularization with dilated lumens. Thicker granulation tissue was the prominent feature of these wounds. The skin was shrunken in the wounded area due to contraction of the wound (Figure 6).

**Discussion**

Teat injuries are common in dairy cattle (Turner and Mcilwraith, 1997), and compared with other frequently occurring diseases, these injuries often result in premature culling of affected cows (Beaudeau et al. 1995). Teat injuries can be divided into two categories (external or internal injuries). The external injuries are classified as being partial thickness (skin to submucosa) or full thickness (skin to submucosa with milk leaking out of the incision) (Nichols, 2008). The present study focused only on the reconstruction of partial thickness of teat laceration with degloving injuries.

Non-penetrating teat lacerations should be cleaned, debrided and, if possible, sutured to be closed (Modransky and Welker, 1993). At the time of suturing, the cosmetic features of the teats and milking comfort must be considered (Johnson, 1988; Nichols, 2008). In excessive teat laceration with a large skin defect, because very little “extra” tissue is available in the teats, skin flap techniques might not be possible for reconstruction of the wound (Waldorn and Zimmerman-pope, 2003). These wounds can be left to heal by second intention wound healing (Modransky and Welker, 1993).

In second intention wound healing, closure is achieved by contraction and epithelialization. Wound contraction is defined as the centripetal movement of the original wound margins. Contraction occurs as the contractive forces of the myofibroblasts exceed the retractive force of the surrounding skin on the wound edges. A greater contribution of wound contraction accelerates healing because contraction occurs faster than epithelialization, especially when lacerations occur in loose skin. (Wilmink et al., 1999). If there is a laxity in the surrounding skin, wound contraction can be unimpeded (Waldorn and Zimmerman-pope, 2003). In the present study, the teats have a loose skin; therefore, wound contraction plays a major role in second intention process in non-grafted wounds. It is explained that wound contraction often causes sever functional and cosmetic problems (Shah et al., 1994). Disadvantages of second intention healing include time and expense, possible contracture or stenosis, and thin or fragile epithelialized tissue (Waldorn and Zimmerman-pope, 2003). Nichols (2008) stated that in excessive teat wound, secondary healing by medical management of the wound might be sufficient. However, contraction of the tissue during healing can change the alignment of the teat, which in turn creates problems during machine milking. With show cows, reconstructive surgery of the teat is necessary to obtain a satisfactory cosmetic outcome (Nichols, 2008). We obtained the same results in that non-grafted wounds, which showed wound contraction, resulted in deformity in normal anatomical structure of the teats, associated with a poor cosmetic appearance.

The deleterious consequence of contraction and excessive scar tissue, including deformity and loss of function, are referred to as wound contracture. Contracture near body orifice may result in stenosis of normal structure (Waldorn and Zimmerman-pope, 2003).

Skin grafts can be used effectively to inhibit wound contraction (Brown et al., 1992). Skin grafts are indicated when there has been a major loss of skin from trauma or other factors, such as tumor removal and major thermal injuries (Waldorn and Zimmerman-pope, 2003). Skin grafts may be full thickness.
(epidermis and entire dermis) or partial thickness (epidermis and a variable portion of the dermis) (Fossum, 2002). A various kind of full thickness skin graft such as mesh, seed, strip and stump grafts can be performed with basic surgical instruments (Waldorn and Zimmerman-pope, 2003). In the present study, full-thickness meshed autograft was used to repair the teat skin defect. Autografts are clinically the most successful type of grafts and are used as permanent grafts because the graft and host are antigenically identical (Pavletic, 1998). Mesh grafts have several advantages over other types of grafts. The slits in the graft provide flexibility for the graft to conform to wound surface. The graft is stable because it can be fixed to the wound surface by sutures through the slits. In addition, as granulation tissue grows into the mesh holes, it provides further immobilization and a source of additional vascularization. Exudate, serum and blood can drain from wound surface through the slits, allowing the graft to keep contact with the wound for revascularization. If the graft bed be treated properly, survival of mesh graft is 90% to 100% (Swaim, 1986; Waldorn and Zimmerman-pope, 2003). In our study, macroscopic and microscopic evaluations confirmed that a successful graft take was observed in all grafted wounds after a 3-week study. Regenerative processes in a skin graft must overtake degenerative processes by postoperative day 7 or day 8 for the graft to survive (Fossum, 2002).

In the current study, because of the hairless skin of the teats, the inner skin of the external ear that has a low density of hair follicles was considered as a donor site for skin graft. Donor site skin should have the same hair texture as the hair surrounding the recipient site (Fossum, 2002). Clinical evaluation of all treated wounds by mesh graft showed all teats restored their normal anatomical structure without any deformity or wound contraction. To the author’s knowledge, this is the first study that presents the beneficial effect of free skin, grafted to large external teat wound. In Molaei et al.’s (2002) study, the use of vestibular mucosal graft for reconstruction of teat mucosa following internal teat injury was recommended (Molaei et al., 2002).

The comparison of the wound healing process between non-grafted and grafted teat wound suggests that skin graft inhibits wound contraction and produces functionally and cosmetically acceptable healing. According to the result of the present study, it can be concluded that using full-thickness free mesh skin graft can be considered a suitable method for treatment of excessive teat laceration.

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**References**


با آزمایش تجربی زخم‌های پوستی سرپستان با استفاده از پیوند پوستی، گوش در گاو شیری

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چکیده
زمینه مطالعه: زخم‌های پوستی سرپستانی یکی از عوامل رایج در کودکان و نوجوانان است. این نوع زخم‌های پوستی به جای درمان نخوردن می‌گیرد. اکتشاف و شناسایی روش‌های کارکرده در درمان زخم‌های پوستی سرپستانی به چشم می‌خورد. 
روش‌کار: در این مطالعه از چهار گروه درمانی سالمند جراحی سلول‌پوش با پیوستن پوست به سطح مخاط کننده و چه سه روش‌کار ساخته شدند و در طول ۳ ماه به گروه‌های مختلف واگذاری گردیدند. 
نتایج: نتایج نشان داد که در سه روش کارکرده طی ۴ هفته انجام داده شد و در خانم‌های فیزیوتراپی، در زخم‌های پوستی سرپستانی با پیوستن پوست به سطح مخاط کننده بهترین نتایج به دست آمد.

واژه‌های کلیدی: گروه جراحی پیوستن پوست، سلول‌پوش، پوستی سرپستانی

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