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Application of Topical Negative Pressure (Vacuum Assisted Closure) managing the horse's wounds: Literature review and three years' experience.

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«…Fama di loro il mondo esser non lassa;
misericordia e giustizia li sdegna:
non ragioniam di lor, ma guarda e passa...»

Inferno Canto III, "Divina Commedia" - Dante Alighieri

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CHAPTER 1

Foreword

1. Foreword

Traumatic injuries causing major soft-tissue damage to the extremities are commonly encountered in animals (Ben-Amotz et al., 2007).

Horses often suffer wounds in relation to their habitat, their use, and their natural instinct. Veterinarians in equine practice are frequently called on to manage traumatic wounds, which can be labor-intensive and expensive. Healing of these wounds is often delayed and complicated compared with that occurring in other species, which leads to significant wastage, because a considerable number of animals are not able to continue their athletic career as a result of persisting lameness, swollen limbs, and extensive scars (Wilmink & van Weeren, 2005).

In a large, multicenter retrospective study of 122,642 cases presented to nineteen American Veterinary Colleges in a four-year period, 2% of all cases required wound treatment and 61% of these wounds were treated with debridement only and healed by second intention (Lindsay, 1990). Although primary or delayed closure is the preferred way of treatment, this can be followed by partial or total dehiscence of the wound, necessitating second-intention healing. Also, in cases of unmanageable contamination, excessive tissue loss, or severe compromise of the tissue, wound healing by second intention is often the only option (Caron, 1999).

To compound the problem, repair of full-thickness wounds by second intention is subject to numerous complications which compromise aesthetic and functional outcome in the horse, including:

- Chronic inflammation;
- Poor contraction;
- Development of exuberant granulation tissue (proud flesh);
- Slow epithelialization (Theoret, 2006).

To minimize cost and time spent on treatment, practitioners must be meticulous in their selection of techniques, dressing, and medications, which should be based on a clear understanding of the mechanisms involved in wound repair (Theoret, 2005). Therefore a lot of alternative strategies for traumatic wounds have been presented in the last years to avoid keloids formation and reduce healing time (Alford et al., 2012). The important of reliable and efficacious treatment is crucial not only to improve patient quality of life, but also to reduce cost of management. However, studies that clarify the correct utilization of medications are not available in literature; in fact, wound management is often labors and frustrate for veterinarians in equine practice.

1.1 Wound Repair in Horses

Wounds repair is a complex temporally and spatially coordinated series of cellular, molecular, physiologic, and biochemical events regulated by a delicately orchestrated cascade of mediators (Theoret, 2001). Damage tissue is removed, and structural integrity is restored.

Healing occurs by two processes: Repair and Regeneration. Aberration in the normal repair process lead to even less desirable outcomes including chronic wounds, fibrosis or contracture (Theoret, 2006).

Repair begins the moment a cellular barrier is broken and follows a pattern of synchronized phases including acute inflammation, cellular proliferation, and finally matrix synthesis and remodeling with scar formation (Fig. 1). The molecular mechanisms regulating this process are not fully understood, but is clear that inflammatory phase is the most important phase for horses (Theoret, 2006).
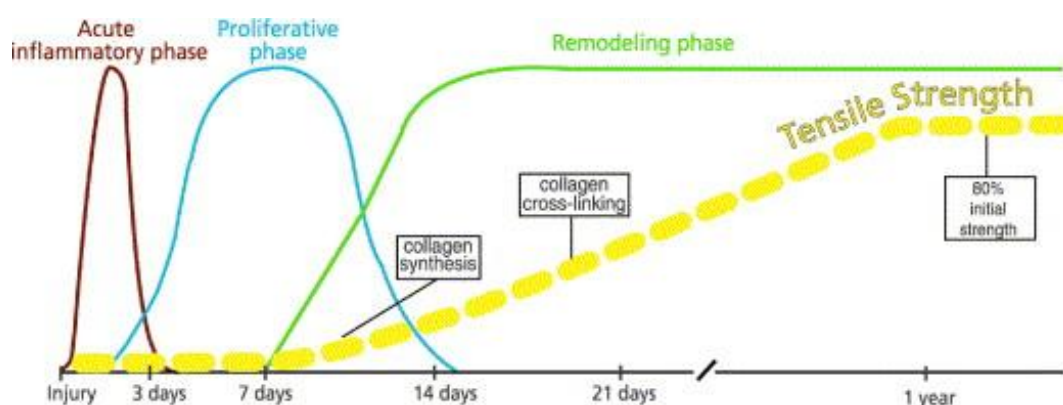

Figure 1 – Temporal profile of various processes and gain in tensile strength occurring during normal cutaneous wound repair. *Clinical Techniques in Equine Practice, 3,* Theoret CL, Update on wound repair, pp. 110–122, Copyright (2004).

The ***inflammatory phase*** is essential to protect against infection as well as to initiate the repair process. During this phase polymorphonuclear (PMN) cells and macrophages migrate to the wound site to clear it of contaminating bacteria and nonviable tissue. This process is referred to as cellular debridement. Macrophages additionally release a plethora of biologically active substances that are essential for the recruitment of more inflammatory and mesenchymal cells and initiate the healing process (Cotran et al., 1994).

Paradoxically, excessive or prolonged inflammation may contribute to the pathogenesis of a number of diseases. It has been shown experimentally that inflammation in horses is weak but protracted (Wilmink et al., 1999a; Wilmink et

al., 1999b) and the horse leucocytes produce less reactive oxygen species essential to bacteria killing. They also produce lower level of other mediators required to reinforce the inflammatory response and to induce tissue formation and wound contraction (Wilmink et al., 2003).

The **proliferative phase** involves epithelialization, fibroplasia and angiogenesis. Macrophages, fibroblasts, and endothelial cells move into the wound space as a unit and depend on one another (Clark, 1985). Macrophages provide a continuing source of cytokines and growth factors necessary for the stimulation of fibroplasia and angiogenesis. Fibroblasts construct new extracellular matrix (ECM) needed to support cell in-growth while blood vessels transport oxygen and nutrients necessary to cell metabolism (Clark, 1993). Fibroblasts use the fibrin clot as a provisional matrix for migration and rapidly replace it with a new loose ECM consisting of glycoproteins (fibronectin and laminin), proteoglycans (hyaluronic acid), and collagens (initially type III, later type I) (Moulin, 1995). The entity of cells and ECM is granulation tissue which fills the gap and is the basis for subsequent wound contraction and epithelial migration (Wilmink & van Weeren, 2005).

The proliferative phase of repair comes about as inflammation subsides, and is characterized by the appearance of red, fleshy granulation tissue, which ultimately fills the defect. The horse activates wound collagen formation to a greater extent and earlier during repair than do other species (Chvapil et al., 1979), which predispose it to the formation of exuberant granulation tissue (proud flesh), with subsequent retardation of contraction and epithelial migration.

The final phase of wound repair involves **wound contraction** and reorganization of the extracellular matrix. This usually occurs during the second week following injury. Wound contraction results from the action of differentiated fibroblasts (myofibroblasts) in the granulation tissue, which contain filaments of smooth muscle actin. Contraction of these filaments in the fibroblasts and connection among fibroblasts and with ECM pull the wound margins centripetally (toward the center) (Clark, 1993; Moulin, 1995) rapidly reducing the wound surface area by means of intact full-thickness skin. Consequently, wound contraction is a critical determinant of the speed of second intention wound healing as well as the final cosmetic appearance of the scar. Wound contraction not only accelerates closure, but also enhances the cosmetic appearance and strength of the scar because proportionally less wound area must be covered by newly formed inferior quality epithelium, which is fragile and lacks normal nervous, glandular, follicular, and vascular components.

For this reason, a high degree of contraction is a desired feature of wound repair, at least in the horse.

Epithelialization is the slowest phase of the wound healing process and concludes wound closure (**0,9 mm/10 days at the most in limb wounds of horses**) (Stashak, 1991). Although epithelialization starts a few hours after trauma with the migration of epithelial cells, it becomes macroscopically apparent only about 2 weeks after wounding. Epithelial proliferation occurs after 2 days, triggered by the secretion of cytokines and growth factors by fibroblasts, inflammatory cells, and the keratinocytes themselves (Clark, 1993; Moulin, 1995). Epithelialization is impaired by fibrin remnants of the clot and by chronic inflammation (Stadelmann et al., 1998). Newly formed epithelium lacks skin adnexa and is thin and fragile because it is poorly attached to the underlying basement membrane (Jacobs et al., 1984). The part of the wound that has healed by epithelialization remains visible as a superficial scar (Wilmink & van Weeren, 2005). Wound tensile strength continues to increase for months but never achieves that of unwounded skin (Theoret, 2006).

1.2   Difference in Wound Healing

Wound healing is often divided into general phases of acute inflammation, proliferation, and remodeling. Because these phases overlap and occur simultaneously in all tissue components, it is *difficult to distinguish them from one another*. In practice it may be more realistic to divide healing into macroscopically apparent events: inflammation, formation of granulation tissue, wound contraction, and epithelialization. Although these events also partially overlap, they largely succeed one another and occur more or less chronologically. Moreover, they are clearly visible to the veterinarian. *These events occur as well during primary intention healing, but then they are shorter and not readily visible*. Remarkable differences in these phases between horses and ponies and between body and limb wounds determine the speed and efficiency of healing (Wilmink et al., 199a; Wilmink et al., 1999b)

1.2.1 Difference in second-intention healing between horse and ponies

Effective coordination of the healing process in horses, particularly of wounds involving the lower limb, is often problematic, resulting in chronic nonhealing wounds or exuberant granulation tissue that inhibits wound contraction and epithelialization (Dart et al., 2005). Compared with wounds of the trunk, distal limb wounds have a **longer preparatory phase** characterized by greater wound retraction, slower rates of epithelialization and earlier cessation of wound contraction (Jacobs et al., 1984; Wilmink et al., 1999a). Anatomic location,

specifically, body versus distal limb, contributes to physiologic differences in organization of the cellular inflammatory response have been found between the horse and pony to explain some of this phenomenon (Wilmink et al., 1999a; Wilmink et al., 1999b).

Recent investigations focus on the roles of epidermal growth factor, fibroblastic growth factor-B1 and the matrix metalloproteinases regulation of fibroplasia (Cochrane, 1997; Knottenbelt, 1997). These studies suggest that **the persistence of inflammation contributes to longer healing times in the horse compared to the pony**. These findings support earlier research, which demonstrated that horses have inherent dermal and subcutaneous precursors predisposing them to exuberant granulation tissue formation (Chvapil et al., 1979). Exuberant granulation tissue delays healing by inhibiting wound contraction and subsequent epithelialization and eventually resulting in a larger scar (Jacobs et al. 1984, Lees et al. 1989, Fretz et al. 1983). Where infection contributes to the prolonged lag phase of wound healing and, because wound healing is a dynamic process, a primary goal should be management of local factors to provide an environment that promotes acceptable wound healing. Requirements for this include microcirculation for oxygenation and nutrition, moisture, increased temperature, neutral to slightly acidic pH, and a bacterial population less than 10^5 organisms per gram of tissue (Wilmink et al. 1999a).

Modulation of the Inflammatory Response

The inflammatory response orchestrates the entire healing process. The acute inflammatory response is a prerequisite for launching healing, whereas chronic inflammation hinders the later phases of wound contraction and epithelialization. In horses the inflammatory response seems too weak initially and fails to resolve in a timely fashion. This implies that inflammation should initially be stimulated and later on be inhibited. Roughly speaking, this means that *inflammation should be encouraged until the wound is filled with granulation tissue, and that it should be inhibited from that point onward to reduce the formation of exuberant granulation tissue and facilitate contraction and epithelialization*. By modulating the inflammatory response, the successive phases of the healing process will simultaneously be influenced. The initial enhancement of inflammation will also boost the **formation of granulation tissue** and the initial impetus to wound contraction, whereas the preclusion of chronic inflammation will benefit both wound contraction and epithelialization. Therefore, treatment of wounds healing by second intention can be simplified by focusing on the inflammatory phase and can be subdivided in either stimulation or inhibition of the inflammatory response (Wilmink, 2008).

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Stimulation of the Inflammatory Response - The initial inflammatory response in horses should be encouraged and in no manner be limited. Simultaneously, the formation of granulation tissue will be enhanced, which is desirable until the wound cavity has filled in. First, the need for inflammation and the risk of infection can be reduced by surgical debridement, regardless of whether a wound is to be sutured. **Surgical debridement** is required when necrosis, exposed cortical bone, or frayed tendons are present in the wound of a horse because cellular debridement, by the inflammatory response, is slow in horses. Furthermore, it is advisable to bandage the wound. Bandages themselves favor the inflammatory response by *increasing local temperature*—which speeds up biological processes in general and angiogenesis in particular—thus *supplying leukocytes* and *lowering pH*, which causes a shift of the hemoglobin dissociation curve, increasing availability of oxygen for the leukocytes. Simultaneously, the formation of granulation tissue is enhanced, although the most important stimulus for this is the increase in oxygen-gradient between the granulation tissue and the wound surface and the decrease in tissue pH found under bandages or casts (Fretz et al., 1983).

Resolution of the Inflammatory Response - As soon as the wound cavity has filled in with granulation tissue, the inflammatory response should no longer be encouraged but inhibited. Chronic inflammation, inherent to limb wounds of horses, enhances the formation of exuberant granulation tissue and delays wound contraction through persistently elevated levels of several mediators. Possible stimuli causing a chronic inflammatory response should be diagnosed and eliminated.

The wound must be **protected** from environmental microorganisms and foreign material. Further, the number of bacteria on the wound surface must be controlled because they are strongly chemoattractive to leukocytes and perpetuate inflammation. **Reduction of surface contamination** can be achieved by wound excision. Superficial layers of tissue are removed and the number of clefts reduced. This also helps eliminate many leukocytes and mediators that accumulate in the superficial layers of the granulation tissue.

Treatments that focus on reducing surface contamination and chronic inflammation concomitantly favor wound contraction and epithelialization. First, they limit the number of leukocytes and thus their toxic effects on epithelialization and reduce levels of contraction-inhibiting inflammatory mediators. Second, such treatments reduce the risk of developing exuberant granulation tissue along with its detrimental influence on both wound contraction and epithelialization.

That there is a need to specifically target wound contraction, particularly in horses, is clear since the rate of contraction determines the speed of second intention healing and the final cosmetic outcome (Wilmink et al., 1999a)

Conditions for optimum epithelialization include a **smooth, healthy bed of granulation tissue** that does not protrude above the level of the adjacent skin, along with a moist wound environment.

Because epithelialization is the predominant mechanism of closure of limb wounds in horses, measures to optimize the conditions for epithelialization are more essential in horses than in ponies (Wilinmk, 2008).

As with all fields of medicine and surgery, wound management has seen advances in the science and art of wound care over the last several years. These are in medication, materials, and methods that are used in treating and reconstructing injured tissues. When considered from the standpoint of comparative medicine, some of the advances apply to management of wounds in both animals and humans.

Therefore, in Veterinary Medicine, we are looking for an innovative technique to treat the chronic and delayed wound healing. After a careful review of human literature, we identified an innovative device able to accelerate the wound healing process, and improve the granulation tissue formation and healing avoiding complications.

1.3 Vacuum-Assisted Closure technique in Human Medicine

Negative pressure therapy has been used in clinical applications for over five decades (Argenta & Morykwas, 1997; Morykwas et al., 1997).

The concept of applying topical negative pressure in the management of wounds emerged in the late 1980s and is increasingly used for a wide variety of wounds. The technique is also known as vacuum assisted closure (VAC), negative pressure wound therapy (NPWT), vacuum sealing technique (VST), sealed surface wound suction (SSS), sub-atmospheric pressure therapy or dressing, foam suction dressing, and vacuum pack technique (VPT).

This technique consists of an noninvasive system that creates a localized controlled sub-atmospheric (negative) pressure.

The practice of exposing a wound to sub-atmospheric pressure for an extended period to promote debridement and healing was first described by Fleischmann et al in 1993 (Fleischmann et al., 1993), following the successful use of this technique in 15 patients with open fractures. They reported that the treatment resulted in "*efficient cleaning and conditioning of the wound, with marked proliferation of granulation tissue*". In two further papers, Fleischmann and colleagues described the treatment of 25 patients with compartment syndromes of the lower limb (Fleischmann et al., 1996) and 313 patients with acute and chronic infections of various types (Fleischmann et al., 1997). The average duration of the vacuum therapy treatment for the patients with compartment syndrome was 12.7 (4-31) days with 2.1 (1-8) dressing changes per patient. These wounds were subsequently either closed by secondary suturing (n=20) or by skin grafts following partial closure by suturing (n=5). The average duration of vacuum therapy in the treatment of the 313 patients with infected wounds was 16.7 days with an average of 3.1 dressing changes (Fleischmann et al., 1997).

Further success with topical negative pressure treatment in Germany was reported by Muller (1997) following the treatment of 300 patients with infected wounds, and in 1998 Kovacs et al described how 'vacuum sealing' could be used for the treatment of chronic radiation ulcers. The results of a prospective trial involving 45 patients with soft tissue injuries including sacral pressure ulcers, acute traumatic soft tissue defects and infected soft tissue defects following rigid stabilization of lower extremity fractures were described by Mullner et al. (1997). They reported that in 38/45 patients (84%), the use of the vacuum sealing technique following irrigation and debridement decreased the dimensions of the initial wound, thus facilitating healing time and the eradication of any pre-existing infection.

In the early studies, negative pressure within the wound was achieved by the use of conventional methods such as wall suction apparatus or surgical vacuum bottles. Both these systems are associated with practical problems in terms of the

delivery, control and maintenance of the required levels of negative pressure, as discussed by Banwell et al. (1998).

In 1995, a commercial system for promoting Vacuum-Assisted Closure (VAC®) was introduced into the United States market. This equipment, called the VAC®, was designed to overcome some of the problems described by Banwell and coll.. The heart of the system is a microprocessor-controlled vacuum unit that is capable of providing controlled levels of continuous or intermittent sub-atmospheric pressure ranging from 25 to 200 mmHg.

Two types of unit are available, a mains operated system with a canister volume of 300 ml for patients with limited mobility or very heavily exuding wounds, and a lightweight, battery-powered unit with a canister volume of 500 ml. This system has a battery life of about 17 hours. The large system is fitted with various audible and visual alarms to indicate if the unit is tipped greater than 45 degrees, the canister is full, or the dressing has an air leak.

1.3.1 Vacuum-Assisted Closure: mode of action

In early studies no attempts were made to investigate the physiological basis for the observed clinical effects, or to determine the optimum levels of pressure required. In a paper Morykwas et al. (1997), addressed both of these issues following a series of animal studies. Deep circular defects, 2.5 cm in diameter, produced on the backs of pigs were dressed with open-cell polyurethane foam with a pore size ranging from 400-600 μm. In the first series of experiments, a laser Doppler technique was used to measure blood flow in the subcutaneous tissue and muscles surrounding the wounds were exposed to increasing levels of negative pressure, applied both continuously and intermittently. Their results indicated that whilst an increase in blood flow equivalent to four times the baseline value occurred with negative pressure values of 125 mmHg, blood flow was inhibited by the application of negative pressures of 400 mmHg and above. A negative pressure value of 125 mmHg was therefore selected for use in subsequent studies.

The rate of granulation tissue production under negative pressure was determined using the same model by measuring the reduction in wound volume over time. Compared with control wounds dressed with saline soaked gauze, significantly **increased rates of granulation tissue formation** occurred with both continuous (63.3 +/- 26.1%) and intermittent (103% +/- 35.3%) application of negative pressure.

The observation that intermittent or cycled treatment appears more effective than continuous therapy is interesting although the reasons for this are not fully understood. Two possible explanations were advanced by Philbeck et al. (1999). They suggested that intermittent cycling results in rhythmic perfusion of the

tissue which is maintained because the process of capillary autoregulation is not activated. They also suggested that as cells which are undergoing mitosis must go through a cycle of rest, cellular component production and division, constant stimulation may cause the cells to 'ignore' the stimulus and thus become ineffective. Intermittent stimulation allows the cells time to rest and prepare for the next cycle. For this reason it is suggested that cyclical negative pressure should be used clinically, although some authors (Collier, 1997; Tang et al., 2000) suggest that this may follow a 48-hour period of continuous vacuum, which can be applied to exert a rapid initial cleansing effect.

Microbiological studies were also undertaken which involved inoculation of punch biopsy wounds with large numbers of microorganisms. These indicated that, compared with control values, ***tissue bacterial counts of vacuum-treated wounds decreased significantly*** after four days. In a final part of the same study using a standard technique, the effect of vacuum therapy was found to ***increase flap survival by 21%*** compared with control values (Morykwas et al., 1997).

Following these investigations, Morykwas and colleagues postulated that multiple mechanisms might be responsible for these observed effects. In particular, they suggested that removal of interstitial fluid decreases localized oedema and increases blood flow, which in turn decreases tissue bacterial levels. It has since been proposed that the application of sub-atmospheric pressure produces mechanical deformation or stress within the tissue resulting in ***protein and matrix molecule synthesis*** (Morykwas & Argenta, 1997) and enhanced ***angiogenesis*** (Fabian et al., 2000).

Fabian et al. (2000), using the rabbit ear model, provided further hard evidence for the stimulatory effects of sub-atmospheric pressure on the production of granulation tissue and also demonstrated a trend to enhanced epithelialization. In experimental partial-thickness burns in pigs, sub-atmospheric pressure was shown to prevent progressive tissue damage in the zone of stasis that surrounds the area of the initial injury. This effect was demonstrable within 12 hours following injury, with treatment times of as little as six hours being sufficient to exert a measurable effect (Morykwas et al., 1999). The authors proposed that removal of oedema fluid containing suspended cellular debris, osmotically active molecules and biochemical mediators, released following the initial injury, may prevent cessation of blood flow.

1.3.2 Clinical Experience with VAC®

Following these animal studies, the same research group described the clinical use of the commercial VAC® in 300 wounds of varying aetiology (Argenta & Morykwas, 1997). These were treated until completely closed or could be covered with a split-thickness skin graft, or were suitable for surgical reconstruction by rotating a flap on to the healthy granulating wound bed. Overall 296 wounds responded favorably to treatment and the authors concluded that *VAC® is an extremely efficacious modality for treating chronic and difficult to heal wounds.*

Numerous other papers have described the use of VAC® in the treatment of a variety of wound types including extensive degloving injuries (Meara et al., 1999; DeFranzo et al., 1999), infected sternotomy wounds (Tang, 2000; Obdeijn et al., 1999; tang et al., 2000), and various soft tissue injuries prior to surgical closure (Bauer et al., 1998), grafting or reconstructive surgery (Avery et al., 2000).

Smith et al. (1997), in a retrospective review, described the use of VAC® over a four-year period in 93 patients who required open abdomen management for a variety of conditions. A total of 171 dressings were applied to the wounds of 38 surgical patients and 55 patients with traumatic injuries. The authors concluded that with careful subsequent management good patient outcomes could be achieved and recommended Vacuum-Assisted Closure as the treatment method of choice for open abdomen management and temporary abdominal closure.

Vacuum therapy has also been used in the treatment of donor sites, particularly in areas that are difficult to manage using conventional techniques (Blackburn et al., 1998) such as those on the radial forearm (Avery et al., 2000). It has been reported that as many as one third of all patients undergoing radial forearm free flaps develop exposed tendon complications and it has been suggested that these individuals may derive particular benefit from the use of VAC® therapy (Greer et al., 1999).

Vacuum assisted closure has also been used in conjunction with split thickness skin grafts in the treatment of burns and is claimed to be particularly useful for body sites with irregular or deep contours such as the perineum, hand or axilla (Schneider et al., 1998; Pfau et al., 2000). In all these situations the vacuum helps to hold the graft securely onto the wound bed thus preventing pooling of tissue fluid which would otherwise make the graft unstable.

Molnar et al. (2000) described how they used VAC® in conjunction with skin grafts to treat four patients with full thickness loss of the scalp following a burn injury or excision of an extensive carcinoma. Normally, if such wounds cannot be closed with a flap, the outer surface of the skull is removed to obtain punctate bleeding and a skin graft is applied a week or two later once granulation tissue has started to form. Without this delay the graft take is usually very poor, but

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with the use of VAC® it was possible to apply a successful skin graft immediately after the initial operation.

Numerous case histories describing the successful use of VAC® in a variety of non-healing or chronic wounds have also been published. These include a recalcitrant below knee amputation wound and a suspected Brown Recluse Spider bite (Mendez-Eastman, 2000), pressure sores (Collier, 1997;Deva et al., 1997; Hartnett, 1998; Baynham et al., 1999; Greer et al., 1999; Mendez-Eastman, 1999), leg ulcers (Mendez-Eastman, 1999), and a group of 30 patients with longstanding wounds that were deemed unsuitable for reconstructive surgery, 26 of whom responded favorably to the treatment (Deva et al., 200).

To function correctly, the adhesive membrane applied over the foam wound insert must form an airtight seal with the skin. Obtaining such a seal can be particularly difficult near the anus or vagina or where the surrounding skin is moist.

Some of the practical problems associated with the application of the VAC® system have been discussed previously by Greer et al. (1999), who developed techniques to allow it to be used successfully on sacral pressure ulcers close to the anus and to multiple large ulcers on the lower extremities.

Fabian et al (2000) in a controlled animal study, investigated the possibility that sub-atmospheric pressure might act synergistically with hyperbaric oxygen (HBO2). They found, however, that although negative pressure increased the rate of healing compared with control values, HBO2 therapy did not offer any significant benefit.

1.3.3 Cost of treatment

The cost of VAC® therapy in Human Medicine is not insignificant. In addition to the purchase cost or hire charges of the machine itself, it is necessary to purchase disposable foam dressings and drainage tubes, canisters and adhesive drapes, which together could easily cost in excess of $25 per day (KCI USA Inc., San Antonio, TX). Nevertheless, Philbeck et al (1999) claimed that the technique was very cost effective in use. In a retrospective study, they compared the treatment costs of VAC® with those of a more conventional therapy by comparing the results of an analysis of the healing rates achieved with the vacuum technique with those recorded for similar wounds in a previously published study. Treatment records of 1032 Medicare patients with 1,170 VAC®-treated wounds of all types that had failed to respond to previous interventions were reviewed. From these data, the healing rates of patients nursed on a low air loss surface (LAL) with 43 pressure ulcers (stages III and IV) located on the trochanter and trunk were abstracted and compared with previously published

values for a comparable group of patients also nursed on a LAL surface and whose wounds were dressed with saline-gauze packs.

Prior to treatment, the VAC® dressed wounds averaged 22.2cm² compared with 4.3cm² for the saline-soaked gauze wounds. Wounds dressed with VAC® closed at an average of 0.23 cm² per day compared with 0.090 cm² for the historical controls. Using these healing rates they calculated that the time to heal a group of patients with wounds 22.2 cm² in area would be 97 days with VAC® and cost $14,546, compared with 247 days with traditional therapy at a cost of $23,465. Whilst acknowledging all the limitations of their study, the authors concluded that negative pressure therapy is an "effective treatment modality for a variety of chronic wounds" producing healing in certain types of pressure ulcers 61% faster than saline soaked gauze whilst reducing costs by 38% (www.kci1.com).

This technique consists of an noninvasive system that creates a localized controlled sub-atmospheric (negative) pressure.

Vacuum-Assisted Closure therapy consists of placing a dressing sponge made of open-cell polyurethane ether foam in the wound. Embedded in this sponge is a no-collapsible, side-ported evacuation tube connected to an adjustable vacuum pump. The vacuum pressure is adjustable up to 250 mmHg (50 to 250 mmHg) and may be used intermittently or continuously (Fig. 2).

Figure 2 – The wound VAC® system includes the suction canister, suction tubing, GranuFoam® pad, negative pressure therapy machine and TRAC® Pad.

The foam dressing sponge was of an open-cell communicating design. With this design, all cell in the foam communicate so that the sub-atmospheric pressure applied to the sponge is applied equally and completely to all wound surfaces in contact with the sponge. The pore size (400 to 600 μm) of the foam sponge promotes maximum ingrowth of granulation tissue. The foam dressing should be cut geometrically to fit the wound, with care taken place the foam materials into the deepest portion of the wound. A plastic adhesive drape is then placed over the sponge, overlapping the wound margins by 5 cm or more to obtain an airtight seal. The tubing should be elevated off the skin surface and "flagged" by the adhesive drape to avoid undue tubing pressure to skin. After vacuum pressure is applied and an airtight seal is achieved, the sponge will collapse and apply equal sub-atmospheric suction pressure to the sides and base of the wound. An open wound is clinically converted to a controlled, closed wound.

In a porcine model, the VAC® has been shown to improve wound healing by increasing:

- Blood flow
- Bacterial clearance;
- Granulation tissue formation.

Other purported mechanisms include:

- Attract the wound edges centripetally in increasing tissue oxygenation by decreasing the pressure on small vessels;
- Removes inflammatory mediators;
- Promote angiogenesis;
- Reduce the need for frequent bandage changes;
- Reduce the need for maintaining intact skin (Jacobs et al., 2009).

1.4 Vacuum-Assisted Closure technique in Veterinary Medicine

There are only 6 case reports and 1 retrospective clinical case series on use of NPWT in the Veterinary literature (Gemeinhardt & Molnar, 2005; Adkesson et al., 2007; Ben-Amotz et al., 2007; Guille et al., 2007; Lafortune et al., 2007; Owen et al., 2009, Jordana et al. 2011).

VAC® therapy was first used at the University of Florida Veterinary Medical Center in 2001 for the management of severe, traumatic, degloving wounds in a **tiger cub** (Lafortune at al., 2007). Since that time, different case reports are presented. Ben-Amotz and coll. (2007) used this device for management of severe, traumatic distal extremity wounds in 15 **dogs**; subsequently this device was used also in two **cats** (Guille at al., 2007; Owen et al., 2007) and one **tortoise** (Adkesson et al., 2007).

In equine practice, to our knowledge, VAC® therapy has been reported to accelerate second intention wound healing in a large traumatic neck lesion in a **horse** (Gemeinhardt & Molnar, 2005) and afterwards this device was used to enhance skin graft acceptance in a horse (Jordana et al., 2011).

This equine practitioner sustains that this technique offers some new options for the care of severely infected wounds in horses and should be considered when the clinician is faced with the management of a large and infected wound in the equine patient.

CHAPTER 2

Objectives

2. Objectives

Vacuum-Assisted Closure (VAC[®]) is a method of wound management that has been used in human surgery for several years. The benefits of VAC[®] therapy have been demonstrated in several human studies (Wilson, 2008).

To our knowledge, VAC[®] Therapy for treatment of traumatic wound in horse has been reported only in two clinical cases (Gemeinhardt & Molnar., 2005; Jordana et al., 2011). Based on human literature, we hypothesized that VAC[®] therapy may be useful for the treatment of traumatic injuries in horses and may decrease wound healing and hospitalized time. Thus, the purpose of this study is to evaluate clinical outcome after Vacuum-Assisted Closure (VAC[®]) therapy in horses with traumatic wounds and report our early experience.

CHAPTER 3

Material and Methods

3. Material and Methods

The study was conducted at the Faculty of Veterinary Medicine at the University of Liége in association with the Faculty of Veterinary Medicine at the University of Milan.

3.1   Criteria for inclusion

Horses with traumatic wounds treated with VAC® therapy were included (January 2010- December 2012). For inclusion, horses had to have no other major abnormalities beyond their wound based on physical examination, hematologic and serum biochemical profiles.

3.2   Application of the VAC® Therapy system

VAC® therapy was initiated after emergency medical and supportive care. Horses, if necessary, were sedated with ACP (0,1 mg/kg intravenously [IV]), and the wounds was clipped and cleaned, explored, thoroughly debrided of any devitalized tissue, and thoroughly lavage with physiologic saline (0.9% NaCl) solution under aseptic conditions. All nonviable tissues were removed before placement of then vacuum-assisted closure sponge.

The skin around the wound edges were meticulously clipped with shaver or depilatory cream and meticulously degreased with alcohol. The wounds were covered with an open cell polyurethane foam (VAC®, GranuFoam®) precut to the wound shape. Over the granulation tissue, where present, was applied the white polyvinyl alcohol foam (VAC® White Foam Dressing®). The foams were fixed to the skin with adhesive drape (VAC® Drape®) after the application of adhesive spray upon the perilesional skin (Opsite Spray®- Smith & Nephew®). A hole of approximately 2 cm diameter was created in the middle of the adhesive drape over the foam, and the evacuation tube (SensaTRAC® Pad) was applied over it connect the foam to the VAC® system. The evacuation tube exited in a proximal direction to decrease the likelihood of the tube becoming kinked or dislodged. Given the short length of the tubing it was decided to cross-tie the horses in a narrow stable to restrict its movement. The VAC® suction system (VAC ATS® Therapy Unit- KCI USA Inc., San Antonio, TX) was hung over the horses head and was set at 125 mmHg of continuous negative pressure.

VAC® sponges were changed every 72 hours and the reapplication was decided as needed. The wound was inspected, the vacuum pump turned off, and the evacuation tube disconnected, which allowed re-expansion of the foam beneath the adhesive drape. The adhesive drape and the sponge were removed; wounds

were inspected and flushed with physiologic saline solution and the skin thoroughly dried. Sterile gloves were used to perform foam changes and apply new adhesive drape, then the evacuation tube was reconnected to the vacuum pump, and continuous suction at -125mmHg re-established. The VAC® Unit contained a 500 ml VAC® Canister for collection of the aspirated exudate from the wounds (Fig. 4).

For horse where full thickness grafts were applied, VAC® therapy was used as a method of immobilizing the graftwound bed interface immediately after reconstructive surgery. In these horses the device was removed after 5 days and graft viability assessed.

The horses were checked every 4 h during the treatment.
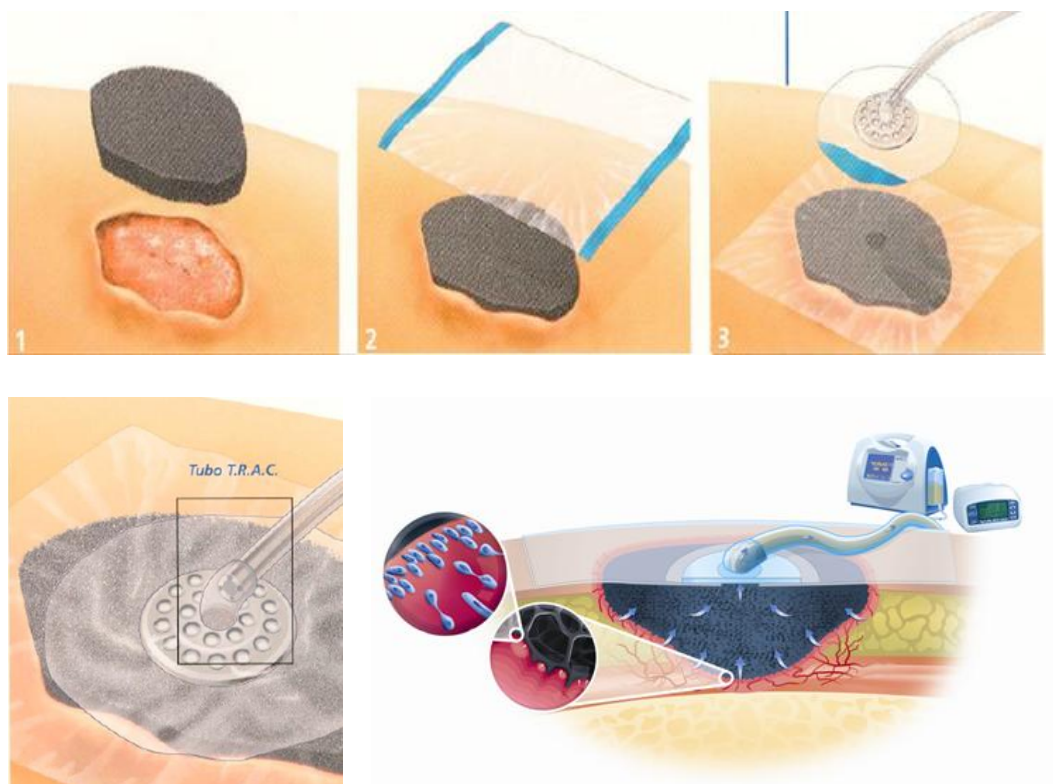
Figure 3 – How to applied VAC® Therapy
(reproduced with permission from KCI Medical Ltd).

3.3   Assessment of wound healing

Photographs were taken with digital camera at a 20 cm focal distance after wiping the wound clean of any exudate and clipping any hair interfering with identification of the wound margin. A horizontal metric scale was used to provide a calibration reference. A label was placed below the wound to identify wound location, date, and horse. The total wound area, wound perimeter, and granulation area was determined by use of planimetry software (Adobe Photoshop CS4®). Each digital image was displayed on a 40 cm colour monitor (FIMI®) and was scale fit the window before the program was calibrated to a 1 cm distance from the metric scale in the image. The wound area and perimeter were determined by tracking the hair margin of the wound periphery. Using Adobe Photoshop CS4® (Fig. 5), with the selection tool, we selected the wound's area, and get pixels count using "Histogram window". Then, with the same technique, we get the quantity of pixels for 1 cm^2 using the ruler present in the photo. Finally, dividing the pixels count of the wound by the pixels count for 1 cm^2, we got the surface dimension of the wound. Each wound was measured 3 times, and the mean value was recorded.

Wounds were considered to have exuberant granulation tissue when the periphery of the granulation bed was above the surrounding level of the skin or the advancing epithelium.

Figure 4 – The wound area and perimeter were determined by tracking the hair margin of the wound periphery using Adobe Photoshop CS4®.

3.4   Qualitative Measurements

At each dressing change, the wound bed appearance, peri-wound status, and discomfort upon dressing removal were evaluated subjectively and recorded (Tab. 1). Subjective evaluation of the wound bed included the designations: pre-granulation tissue or granulation tissue. When granulation tissue was present, it was defined as "smooth" (depth variations estimated to be <0.5 cm), or "irregular" (depth variations estimated to be >0.5 cm), and the colour assigned as red, purple-red, or pale.

Table 1 - Subjective evaluation wounds at dressing change.

Fluid colour	Clear	Pink/red	Brown	Yellow	Green
Wound fluid	Serous	Serosanguineous	Sanguineous	Purulent film	Purulent ooze
Periwound	Normal	Oedema	Erythema	Ulceration	Induration
Hydration status	Normal	Maceration+	Maceration++	Maceration+++	Desiccation
Granulation issue	None	Healty red and granular	Dark red and granular	Purple and friable	Pale and fibrous
Quality	Smooth			Irregular	
Discomfort	None	Mild irritation		Painful	Marked painful

CHAPTER 4

Results

4. Results

4.1 Signalment

Seven horses were included in this study (Tab. 2). Breed distribution was unknown breed (1), Trotter (1), Thoroughbred (1), KWPN (2) and Warmblood (2). Mean age at admission was 13.4 years (range, 2 years- 25 years). All wound were caused by trauma and when were treated with VAC® therapy all wound were classified like chronic. Only one horse was admitted within 24 hours of injury but was treated with VAC® after first alternative treatment.

Three wounds were located in the body; cranial antebrachium (1), neck (1), thorax (1) and four were located in distal limbs; carpus (1) and three horses had hook's wound (3). All the patient were horses and no ponies were present in the study.

Table 2 - Summary data for 7 horses with traumatic wounds treated with Vacuum-Assisted Closure Therapy.

G= Gelding; S= Stallion; F=Female.

Horse	Age (years)	Sex	Breed	Weight (cm)	Height (Kg)	Wound location
1	15	Gelding	?	1,65	487	Upper cranial antebrachium
2	18	Gelding	Warmblood	1,76	500	Dorsal aspect right hook
3	2	Stallion	Trotter	1,63	420	Neck
4	5	Gelding	KWPN	1,72	523	Thorax
5	7	Stallion	Thoroughbred	1,75	560	Dorsal aspect right hook
6	22	Female	Warmblood	1,74	554	Caudal aspect hook
7	25	Female	KWPN	1,80	575	Dorsal aspect left carpus

4.2   VAC® Application

For every horse, the bandage was easy to apply. In one case (case n°1), we encountered an inability to obtain vacuum, probably due to an incorrect preparation of the skin and a wrong application of the surgical spray (Opsite Spray®- Smith & Nephew®). We noticed a slight skin irritation after the application of the depilatory cream used to remove the hair from the horse's skin.

Five horses tolerated the bandage and VAC® therapy well, two horses (case n°3 and 5) may be for the boisterous nature, had not tolerated the therapy and so the treatment was prematurely interrupted.

Minor line separation caused loss of vacuum in 1 horse early (case n°2), which was correct immediately by simple equipment replacement. Because of the scheduled checks, the maximum time negative pressure could have been lost in our horses were 4 hours. Horses did not display any abnormal vocalization, tachycardia, tachypnea, inappetence during the treatment. At first bandage change (Day 3), discomfort was noted in two horses (case n°1 and 3) but in no one horses we encountered pain in association with bandage change.

At every bandage change granulation tissue appeared to be proliferating into the interstices of the foam dressing (Fig. 6) and in all cases the sponge was filly of exudate.

Figure 5 - Granulation tissue can be seen growing into the interstices of foam dressing, causing mild disruption of the vascular tissue and some minor hemorrage.

Periwound skin irritation was evident in 2/7 (case n°1 and 4) wounds in after days 3. In one case (case n°1) skin irritation was done to the use of a depilatory cream that result in serous discharge from the skin for several hours after application. In one case (case n°4) the therapy was prematurely interrupted for owner request, because the horse was contracting an massive ventral oedema.

All wound fluid evaluations were serous or serosanguinous, and no maceration and desiccation was noted in any wound. The median amount of fluid aspirated by the canister was 567 ml (range, 300 ml-800 ml) with a mean of 159 ml (range, 100 ml-200 ml) of fluid aspirated in the first 24 hours.

Granulation tissue appeared in all horses after the first treatment (day 3); "smooth" granulation tissue was recorded more frequently than "irregular" and the colour was more frequently red (Tab. 4).

The therapy has had no negative impact on the long-term function to any prolongation of horse immobilization.

The total wound area has been determined, for each case, every time that the bandage has been changed. Visually, that was possible to see differences in wound contraction and granulation tissue formation. All wounds of horses showed a massive contraction during the therapy (Tab 3).

Table 3 – Total wound area and % of contraction for each dressing change.

N°	Day	Size (cm^2)	% of contraction
1	0	72,6	
	3	68,5	5,64[*]
	6	55,2	19,4[*]
	9	44,6	19,2[*]
2	0	13,5	
	3	12,3	8,9[*]
	6	11,2	8,9[*]
4	1	53,36	
	4	49,2	7,8[*]
6	0	131	
	3	120	8,3[*]
	6	108	10[*]
7	0a	92,7	
	3a	89,3	3,66[*]
	0b	88,5	
	3b	85,3	3,61[*]

(*): The contraction percentage has been calculated respecting the previous step of the therapy.

Table 4 - Subjective evaluation wounds at dressing change for each horse.

N°	Day of change bandage	Fluid colour	Wound fluid	Periwound	Idratation status	Granulation tissue	Quality of tissue granulation	Discomfort
1	Day 3	Pink/red	Serosaunguineous	Erythema	Normal	Healty red and granular	Smooth	Mild irritation
	Day 6	Pink/red	Serosaunguineous	Normal	Normal	Healty red and granular	Smooth	None
	Day 9	Pink/red	Serosaunguineous	Normal	Normal	Healty red and granular	Smooth	None
2	Day 3	Pink/red	Sanguineous	Normal	Normal	Healty red and granular	Smooth	None
	Day 6	Pink/red	Sanguineous	Normal	Normal	Healty red and granular	Smooth	None
4	Day 1	Pink/red	Sanguineous	Normal	Normal	Dark red and granular	Smooth	Mild discomfort
	Day 4	Pink/red	Sanguineous	Erythema	Normal	Dark red and granular	Smooth	Moderately painful
6	Day 3	Yellow	Serosaunguineous	Normal	Normal	Dark red and granular	Smooth	None
	Day 6	Pink	Serosaunguineous	Normal	Normal	Dark red and granular	Smooth	None
7	Day 3a	Yellow	Purulent film	Ulceration	Maceration +++	Pale and fibrous	Irregular	Moderately painful
	Day 3b	Pink/red	Sanguineous	Erythema	Normal	Dark red and granular	Smooth	None
	Day 6c	Pink/red	Sanguineous	Oedema	Normal	Dark red and granular	Smooth	None

Case n°1

A 15 year-old gelding was referred to the University with a 2 weeks-old, very large wound at the level of the upper craniolateral aspect of the left antebrachium. The injury had been sustained on fence wire and had been treated by the referring veterinary surgeon since the first day but without success.

On admission the animal was in a relatively poor body condition. A grade lameness of the left forelimb was evident. The wound was very large and extended both cranially and laterally. A significant amount of exuberant granulation tissue was present. The tissue was pale pink with irregular surface and there was a massive loss of tissue.

Surgical debridement of the wound was performed under standing sedation and with a loco-regional anesthesia. All exuberant tissue was excised, the wound edges were refreshed and the wound was covered with an antiseptic dressing. The bandage was changed every one or two days during the first two weeks of hospitalization and after this period the team decided to apply VAC$^®$ system.

VAC$^®$ therapy was initiated after this treatment (Day 0) and was continued for 9 days with three times bandage changes (Day 3-6-9). Nine days after starting therapy 800 ml of hemorrhagic wound exudates had been collected and the VAC$^®$ therapy was discontinued. Approximately 200 ml of wound exudate was collected during the first 24 hours. The following days, the amount of fluid aspirated in the canister progressively decreased with only 20 ml begin collected by the last day. The bottles were exchanged 2 times during the treatment.

At **first** bandage removal (Day 3) was observed a granulation tissue of good quality. Hypergranulation did not occur and the surface of the granulation tissue was red, slightly bleeding and "smooth" with irregularity most likely due to suction of the tissue onto the foam. The skin all around the wound was irritated and the horse show mild discomfort at the bandage change. This was probably due to the use of the depilatory cream used 4 days before for shaved the skin.

At **second** bandage removal (Day 6) the granulation tissue had increased its extension. Hypergranulation did not occur and the surface of the granulation tissue was pink with some area pale and "smooth" with irregularity.

At **third** bandage removal (Day 9) was observed a granulation tissue of good quality. Hypergranulation did not occur and the surface of the granulation tissue was red, "smooth" and had covered part of the surface.

Case n°2

A 18 year-old Warmblood gelding was referred to the University with a 3 weeks-old, large wound at the level of the dorsal aspect of the right hook. The injury had been treated by the referring veterinary surgeon since the first day but without success.

On admission the animal was in good body condition. A low grade lameness of the right hind limb was evident. The wound was large (dimension ….) and extended both dorsally and medially. A significant amount of exuberant granulation tissue was present in all surfaces. The tissue was pale pink, yellow with irregular surface and was malodorous. The affected limb also showed a significant degree of cellulitis of the soft tissue from the pastern to the tarsus.

Surgical debridement of the wound was performed under loco-regional anesthesia and the same day was applied for the first time the VAC® therapy (Day 0 - Fig.8A).

During the first day of treatment there was loss of vacuum for the uncorrected creation of the hole upon the sponge. This was early corrected by simple equipment replacement (Fig. 8D)

Six days after starting therapy 500 ml of hemorrhagic wound exudates had been collected and the VAC® therapy was discontinued. Approximately 100 ml of wound exudate was collected during the first 24 hours.

At **first** bandage removal (Day 3) was observed a granulation tissue of good quality. Hypergranulation did not occur and the surface of the granulation tissue was red, bleeding and "smooth" with irregularity most likely due to suction of the tissue onto the foam (Fig. 8B). The skin all around the wound was not irritated and the horse didn't show discomfort at the bandage change. Furthermore, was observed a considerable reduction of perilesional oedema with the resorption of cellulitis.

At **second** bandage removal (Day 6) the granulation tissue had increased its extension. Hypergranulation did not occur and the surface of the granulation tissue was pink with some area pale and "smooth" with irregularity. The wound was bleeding and there was a margin of epithelialization on the proximal aspect of the wound (Fig. 8C).

__Case n°3__

A 2 year-old Trotter stallion was referred to the University with a 4 week-old, large wound at the level of the left aspect of the neck. The injury had been treated by the referring veterinary surgeon since the first day with a slight improvement.

On admission the animal was in good body condition. The wound was large and a significant amount of exuberant granulation tissue was present in all surface. The tissue was pale pink with irregular surface and there were scars in all the wound edges.

Surgical debridement of the wound was performed under standing sedation, with a twitch applied to the nose. All exuberant tissue was excised, the wound was cleaned with sterile saline solution and the wound edges were refreshed. VAC® therapy was initiated after this treatment (Day 0) and was continued for only 20 hours for problem related to the nature of the horse. When the sponge is first connected to suction and subatmospheric pressure is transmitted to the wound, pain started for the horse. The amount of fluid aspirated in the canister during this period was 20 ml.

Case n°4

A 5 year-old gelding was referred to the University with a large wound at the left thorax. The injury had been sustained on fence wire and happens 2 hours before the hospitalization.

On admission the animal was in a good body condition and at this moment was performed a surgical debridement and suture. The suture procedure was performed under standing sedation using constant rate infusion of a 0.5 mg/ml detomidine 1% solution at 6 μg/Kg/hr. The skin around the wound edges was shaved and aseptically prepared. The wound was rinsed with sterile physiologic isotonic solution. A suture was performed and the wound was covered with an antiseptic bandage. The wound was kept covered with a bandage with a different choice of primary wound dressing according to the stage of wound healing.

The bandage was changed every one or two days during the first two weeks of hospitalization and after this period the team decided to apply VAC® system.

VAC® therapy was initiated (Day 0) and was continued for 4 days with two times bandage changes (Day 1-4). Four days after starting therapy 300 ml of hemorrhagic wound exudates had been collected and the VAC® therapy was discontinued. Approximately 100 ml of wound exudate was collected during the first 24 hours. The following days, the amount of fluid aspirated in the canister progressively decreased with only 20 ml begin collected by the last day.

At **first** bandage removal (Day 1) was observed a granulation tissue of good quality. Hypergranulation did not occur and the surface of the granulation tissue was red, slightly bleeding and "smooth" with irregularity most likely due to suction of the tissue onto the foam. The skin all around the wound was irritated and the horse show mild discomfort at the bandage change.

At **second** bandage removal the granulation tissue had increased its extension. Hypergranulation did not occur and the surface of the granulation tissue was red, "smooth" and had covered part of the surface. The skin all around the wound was markedly irritated (erythema) and the horse show moderate pain at the bandage change.

Due to the ventral oedema and skin erythema the therapy was interrupted and the horse has been give back to the owner.

Case n°5

A 7 year-old Thoroughbred stallion was referred to the University with a 4 week-old, large wound at the level of right hook. The injury had been treated by the referring veterinary surgeon but hyperganulation and exuberant granulation tissue compared.

On admission the animal was in good body condition and a grade lameness of the right hind limb was evident. The wound was very large and a significant amount of exuberant granulation tissue was present. The tissue was pink with irregular surface and bleeding.

Surgical debridement of the wound was performed under standing sedation and locoregional anesthesia. The wound was cleaned with sterile saline solution and the wound edges were refreshed. VAC® therapy was initiated after this treatment (Day 0) and was continued for only 2 hours because the horse show an important discomfort when the therapy start. When the sponge is first connected to suction and subatmospheric pressure is transmitted to the wound, pain started for the horse. He started to kick and show stringhalt movement. The therapy was then disconnect.

Case n°6

A 22 year-old female was referred to the University with a 1 months-old, very large wound at the level of the caudal aspect of the left hook. The owners didn't want to treat, during the first period, the wounds for economic reason. After 1 month on admission the animal present a large wound extended both medially and laterally (dimension). A significant amount of exuberant granulation tissue was present. The tissue was pale pink and yellow with irregular surface and there was a massive amount of scars and exudation. The affected limb also showed a significant degree of cellulitis of the soft tissue from the pastern to the tarsus (Fig. 15A).

Surgical debridement of the wound was performed under standing sedation. All exuberant tissue was excised, the wound edges were refreshed and the Vacuum therapy was initiated (Day 0).

VAC® therapy was continued for 6 days with two times bandage changes (day 3-6). Six days after starting therapy 700 ml of similpurulent wound exudates had been collected and the VAC® therapy was discontinued. Approximately 200 ml of wound exudate was collected during the first 24 hours. The following days, the amount of fluid aspirated in the canister progressively decreased with only 50 ml begin collected by the last day. The bottles were exchanged 3 times during the treatment (Fig. 15D).

At **first** bandage removal (Day 3) was observed a granulation tissue of good quality. Hypergranulation did not occur and the surface of the granulation tissue was red and "smooth" with irregularity most likely due to suction of the tissue onto the foam (Fig. 15B). The skin all around the wound was not irritated and was observed a considerable reduction of perilesional oedema with the resorption of cellulitis.

At **second** bandage removal (Day 6) the granulation tissue had increased its extension. Hypergranulation did not occur and the surface of the granulation tissue was red, slight bleeding and "smooth" with irregularity (Fig. 15C).

Unfortunately, after the bandage change, the owners decided to not go on therapy. The treatment has been interrupted and the horse has been give back to the owner.

Figure 6 – Case n°6. A: Day 0; B: Wound at first bandage change (Day 3); C: Wound at second bandage change (Day 6); D: Canister.

Case n°7

A 25 year-old female was referred to the University with a 3 months-old, very large wound at the level of the dorsal aspect of the left carpus. The injury had been treated by the referring veterinary surgeon since the first day but without success. At the admission the wound was very large, interesting al the dorsal and medial aspect of the left carpus and with a pale pink tissue with irregular surface. The affected limb also showed a significant degree of cellulitis of the soft tissue from the pastern to the upper antebrachium (Fig. 16).

Surgical debridement of the wound was performed under standing sedation. All exuberant tissue was excised, the wound edges were refreshed and the Vacuum therapy was initiated for the first time (Day 0a – Fig. 16 A).

VAC® therapy was continued for 3 days (Day 3a). Three days after starting therapy 200 ml of similpurulent wound exudates had been collected and the VAC® therapy was discontinued.

At bandage removal (Day 3a) was observed a yellow tissue all over the wound bed. The surface of this tissue was "irregular" and with a similpurulent film (Fig. 16B). The skin all around the wound was ulcerated and was observed a considerable pain all around the wound.

A second surgical debridment of the wound was done and all tissue was excised, the wound edges were refreshed and the wound was covered with an antiseptic dressing. The bandage was changed every one or two days during the first two weeks of hospitalization and after this period the team decided to apply VAC® system for the second time (Day 0b- Fig. 17A).

VAC® therapy was continued for 3 days (Day 3b) and after these three days 300 ml of serosaunguineous wound exudates had been collected.

At bandage removal (Day 3b) was observed a granulation tissue of good quality. Hypergranulation did not occur and the surface of the granulation tissue was red, bleeding and "smooth" with irregularity most likely due to suction of the tissue onto the foam (Fig. 17B). The skin all around the wound was irritated and the horse didn't show discomfort at the bandage change. Furthermore, was observed a considerable reduction of perilesional oedema with the resorption of cellulitis.

Considering the size and the location the wound, it was decided to accelerate further healing with a skin graft. Over the next, 7 days, the wound was prepared for grafting according to the recommendations of Wilminks et al. (2006). The skin grafting procedure was then performed and VAC® therapy was chosen for enhance the good results of this procedure (Day 0c- Fig. 17C).

VAC® therapy was continued for 6 days without bandage changes (Day 6c). Six days after starting therapy 700 ml of hemorrhagic wound exudates had been collected and the VAC® therapy was discontinued. Approximately 200 ml of wound exudate was collected during the first 24 hours. The following days, the

amount of fluid aspirated in the canister progressively decreased with only 500 ml begin collected by the last day. The bottles were exchanged 3 times during the treatment.

At bandage removal (Day 6c) the granulation tissue had increased its extension. Hypergranulation did not occur and the surface of the granulation tissue was red, bleding and "smooth" without irregularity. A graft acceptance of 99% was observed and the wound did not excessively exudate anymore (Fig. 17D and 18A).

After the VAC® therapy, the granulation tissue remained of good quality and the grafts gradually epithelized to coalesce with each other on wound edges. Complete epithelialization was observed 36 days after skin grafting (Fig. 18B).

A   B

Figure 7 – Case n°7. A: Day 0a; B: Wound at first bandage change (Day 3a).

A

B

C

D

Figure 8 - Case n°7. A: Wound before second VAC therapy (Day 0b); B: Day 3b; C: Wound at day of skin grafting (Day 0c); D: Wound at (Day 6c).

Figure 9 – Wound epithelialization after the removal of VAC® therapy.
A: after 3 days; B: after 36 days.

CHAPTER 5

Discussion

5. Discussion

In clinical practice, different methods for wound healing in horses are used. Studies that clarify the correct utilization of medications are not available in literature; in fact nowadays, wound management in equine practice is often based on their own knowledge and preferences.

Clinicians usually have a preference for a particular treatment method because they have experience and feel comfortable in using their preferred treatment method. Also, cost aspects may play a role. Changing the way of clinical practice is very difficult and usually not based on available evidence.

In recent years, the VAC technique is a novel approach in wound healing management in human. VAC® therapy was first described by Morykwas and Argenta in 1997; until now the effectiveness and the use is documented in over 400 peer-reviewed journal articles. Currently in Veterinary Medicine there are only anecdotal reports of the use of this device in horses.

The reason for the significant beneficial effect on wound healing observed in human with the use of vacuum-assisted closure requires further explanation. Three factors responsible for accelerated wound healing with this therapy have been postulated:

1. Removal of excessive oedema fluid with increase in tissue blood flow;
2. Associated decrease in the amount of bacteria;
3. Movement and growth of tissue surrounding the wound in response to the mechanical force of suction pressure.

Oedema with the collection of third-space fluid compromises the microcirculation and lymphatics. Such oedema also interferes with the normal inflow of oxygen and nutrients and outflow of toxins and inhibitory factors. Vacuum-assisted therapy removes third-space fluid effectively, drawing 200 ml or more of fluid from large wound in 24 hours in human medicine. Inhibitors of wound healing have been identified in oedema fluid surrounding chronic wounds (Argenta & Morykwas, 1997; Morykwas et al., 1997). Removal of oedema fluid surrounding the wound, therefore, benefits wound healing in two ways: it increases circulation and decreases the presence of wound inhibitors. Thus, healthy granulation tissue appears much more quickly the expected.

The exact mechanism by which the negative pressure of the VAC® procedure increased the blood flow in soft tissue needs to be analyzed in more detail. However, recent findings of various authors show that VAC® increases the capillary diameter and thereby the blood flow velocity and it could be demonstrated that VAC® stimulated angiogenesis and endothelial cell proliferation (Chen et al., 2005).

More recently, computer stimulations and in vitro studies have demonstrated that the presence of negative pressure can cause cellular strain and micro-deformations on the wound. Furthermore, 5-20% strain on cells could promote cellular proliferation and molecular changes (Greene et al., 2006).

Inserting this system into the therapy of complex wounds in horses provides several advantages. VAC® therapy is **easy to handle**, may **reduce the frequency of dressing changes**, provide reliably **sterile conditions**, **decrease contamination** and effectively **drain the wound**.

The VAC® had been designed to promote the formation of granulation tissue for faster healing in the beds of acute and chronic wounds and is therefore useful in both devitalized and infected tissue.

In our experience, wound coverage using VAC® therapy allows inspection of the wound, checking adequate surgical wound cleaning and obviates superficial infection of granulations or the relics of necrotic tissue hidden in wound pockets and immature, poorly perfused scar tissue.

The VAC® system provides sterile wound coverage with a variable drainage effect which may reduce tissue oedema. In our patient VAC® therapy removes interstitial fluid effectively, drawing a mean of 200 ml of fluid from this large wound in the first 24 hours of treatment. The total quantity of exudates aspirated was higher than larger was the wound. In 3 horses with severe limb wounds were observed a considerable reduction of perilesional oedema with the resorption of cellulitis. This pronounced reduction of cellulitis was observed in all cases after 3 day of continuous therapy.

The VAC® system accelerates wound closure by enhancing the rate of granulation tissue formation. The smoothness of the granulation tissue was quite notably when VAC® therapy was applied then during the day before. Unfortunately we could not accurately assess alignment of collagen fibers histologically to the support the macroscopic findings. The reason of this may have been because our failure to obtain the sample; we were afraid to stimulate the hypergranulation tissue formation with sample collection.

Granulation tissue was not exuberant in any horse, this may be a direct mechanical effect of the negative pressure pressing on the wound. The ability of the negative pressure modality to produce a smooth, vascular bed of granulation tissue within a few days of wounding could be of great value when a reconstructive effort is being contemplate. The reason for the superior quality of granulation tissue production with the VAC® therapy is not fully understood (Orgill et al., 2009). It is postulated that the deformation of cells occurs also in tissues distant stretching may additionally enhance the formation of the new periwound tissue.

Plassman (1995) states that variations in photographic perspective of 20° have a significant effect on repeated 2D area measurements, causing a 10%

measurement error. Finally, a slight change in a patient's position while maintaining the same photographic prespective could affect the technique's precision by altering wound morphology. In our study, all pictures have been taken respecting the same distance between the picture camera and the horse. But the position of the animal was not all the time easy to reproduce. For this reason, it was not always possible to get precisely wound dimensions. And so, we estimated some wound dimensions that probably are not the really dimensions.

The VAC® system functioned properly during all the treatment period without major complications. However, other authors report difficulties in getting the system to function properly despite the reports of good results with the use of VAC® therapy in horses (Gemeinhardt & Molnar, 2005). The most frequent problems are difficulties in maintaining an airtight seal over the wound and in keeping the VAC® dressing in place due to the irregular geometry and motion at the distal limb of the horse. Intolerance of the VAC® therapy due to sensitivity or pain caused by negative pressure has also been reported in some patients. Excessive coiling and kinking of the evacuation or extension tubing may occur in horses that are allowed to move freely around the stable. Some authors also reported problems positioning the vacuum pump to keep it out of the way yet readily accessible. In our study, the VAC® system was tolerated very well and only two horses showed signs of pain due to the negative pressure. A complete airtight seal was obtained by shaving the skin for 20 cm around the wound, degreasing it with alcohol, drying the skin carefully and applying adhesive spray under the adhesive drape (Opsite Spray®- Smith & Nephew®). The application of a bandage over the VAC® dressing in some cases had provided additional help to maintain the airtight seal. In our horses maintenance of negative pressure was greatly facilitated by location of the VAC® suction system hung over the horses head. This location allowed freedom of movement for each horse in its stable whilst constantly connected to the vacuum pump, and minimized the chances of accidental line disconnection, tangling or chewing.
Nowadays, portable battery-driven VAC® systems exist (VAC Freedom™, KCI). These smaller, lighter units (1.45 kg versus 5.6 kg) may be fixed on the horse allowing the patient to move free in the stable. However, they may be more inconvenient as they have smaller canister (300 ml instead of 500 ml) and the batteries need recharging frequently.

It should be noted that loss of negative pressure for prolonged periods, would convert the wound to an occlusive environment and lead to wound maceration and bacterial proliferation. Regular monitoring of negative pressure settings, either by staff or owners, is required for this therapy. In our horses none show loss of negative pressure for long time; only in one case (case n°2) during the

first day of treatment there was loss of vacuum for the uncorrected creation of the hole upon the sponge. This was early corrected by simple equipment replacement and the therapy has continued without other problem.

In this study VAC® therapy was easily applied and was easily to handle. Periwound erythema developed in some patients with prolonged used of VAC® therapy, but was not associated with a real discomfort or pain. In some horses (case n°1 and 4), possibly those with sensitive skin, periwound skin erythema occurred in the first 3 days. This has been reported in people and in dogs, and has been attributed to the irritating effects of the adhesive used to ensure integrity of the negative pressure environment (Ben-Amotz et al., 2007). The depilatory cream we have been using causes an effusive inflammatory response in the skin, necessitating a delay of at least 24hours between depilation and application of the VAC® system. A less reactive means of depilating the surrounding skin would be advantageous. Simply clipping the hair and shaving the skin is inadequate, as hair regrowth compromise the seal between skin and drape before the next dressing change is due. This occurrence could be avoided by the use of a shaver for shaving the skin before the treatment and by minimizing draft during the bandage change.

There is definitely an initial learning curve when using this modality, and we had some issues with the boisterous nature of our stallion horses. That is important to kwon because also the nature of our horses can influence our results; we lost the vacuum pressure in 2 horses (case n° 3 and 5) early in the study.

One advantage of VAC® therapy is the longer time interval between dressing changes. Typically, once the dressing has been properly applied and vacuum established, minimal care is required and dressing changes need only be performed every 3 days. Although periodic checks are required to assure proper machine function and dressing integrity, this lower intensity of care reduces the need of specialized supervision, and could be managed by owners. In comparison, a wet-to-dry dressing requires bandage changes under sedation every 12-24 hours, with increased risk of strike-through.

Some authors have recommended bandage changes every two to three days, when using VAC® therapy to prevent the granulation tissue from growing into the foam (Fig. 6). Different types of foams are available as primary dressings that can be applied over the wound when using VAC® therapy. Polyurethane foams (V.A.C. GranuFoam®) are intended to stimulate granulation tissue formation while aiding wound contraction. Polyvinyl alcohol foams (V.A.C.® WhiteFoam Dressing) on the other hand are hydrophilic dressings that control the growth of granulation tissue and prevent adherence of the foam to the wound (clinical guidelines provided by the VAC® company KCI Medical). In the present study we changed the dressings every 3 days because in our experience after that there

were less complications. Removal of the foam dressing created a small amount of sanguineous oozing from the surface of the granulation bed; however, dressing changes did not appear difficult to done. Nevertheless, it may be prudent to change the dressing at intervals of no greater than 3 days, to minimize disruption of the granulation bed and migration of epithelial cells from the wound edges. In each cases were applied polyurethane foams because there was necessity of extra granulation tissue formation. Only in one case was used the polyvinyl alcohol foams (case n°2) because granulation tissue was already present and no extra granulation tissue was necessary.

The VAC® system can be used with either continuous or intermittent subatmospheric pressure. Experimental research in porcine models has shown that intermittent therapy (5 min on/ 2 min off) results in faster granulation tissue formation than continuous negative pressure alone (Morykwas et al., 1997; 2001). On the other hand, the application of continuous negative pressure stimulates granulation tissue formation significantly faster than the application of simple, non-adherent dressings (Morykwas et al., 1997). Continuous therapy is indicated during the first 48 hours of wound management and for patients which experience significant discomfort during intermittent therapy, i.e. when difficulties occur while maintaining the airtight seal, when there are high levels of drainage, when treating skin flaps or grafts to prevent shear or when a splinting effect is required (clinical guidelines provided by the VAC® company KCI Medical). In the present study, continuous negative pressure were always used because we want stimulated the formation of granulation tissue and we were afraid of a possible failure of the therapy possible with the intermittent modality (i.e. leakage of air, pain or discomfort with intermittent therapy).

We selected 125 mmHg negative pressure based on observations that wounds treated with this negative pressure had a significant increase in granulation tissue formation compared with treatment at 25 or 500 mmHg negate pressure (Morykwas et al., 2001).

In different studies some author report that was contraindicated applying continuous suction to wounds in which blood vessels were exposed, as there was a potential risk for vessel rupture and subsequent exsanguination if the patient was not monitored closely. In our cases none of our horses presented blood vessels exposed or bleeding and we encountered no vascular problems by applying the VAC® system directly to these wounds.

The VAC® system further contributes to wound healing by limiting or preventing two important impediments to second intention healing in horses; infection and desiccation (Stashak, 1991). The foam dressing and barrier drape protect the wound from dust, flies and other contaminants, and they preserve a correct environment on the wound surface. In addition, VAC® system permitted

a continue evacuation of the wound exudate thus avoiding graft loss after skin graft. In one horse (case n°7) during our study, the use of VAC® therapy to greatly enhances acceptance of punch skin grafts in large distal wounds resulting in a survival of 99% of the grafts .The bandage was not changed during the five days of VAC® therapy thereby allowing a constant wound environment and an appropriate union between graft and recipient bed to form. This shows that vacuum-assisted closure therapy results in a nearly perfect punch grafts survival and significant enhancement of healing of a large distal wound.

In our opinion our result, together with those of previous reports, is very encouraging and supports the use of VAC® therapy in horses to treat large, chronic or infected wounds and to enhance acceptance of different types of skin grafting in different locations of the body.

In conclusion, with multiple applications in a variety of wounds including those that can prove difficult to heal, the VAC® is a promising new tool in the field of wound healing in horses.

A limitation of our study is the fact that sometimes it has been stopped prematurely. One could argue that no solid conclusion can be drawn from this study because it was not completed, but in our opinion, the results are clear. It is important look at both the scientific aspect and behaviour results. Clinical studies in wound healing are difficult to execute and do not resemble clinical practice in most cases. Every patient and every wound is different and blinding of treatment is most of the time not possible.

Research continues to explain the benefits of this therapy on wound healing. It is hoped that the modifications of wound healing with vacuum devices will provide much needed insight into the remarkable process of wound healing.

CHAPTER 6

References

6. References

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CHAPTER 7

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