in vivo *26*: 139-142 (2012)

A Successful Combined Treatment with Dermal Substitutes and Products of Regenerative Medicine in a Patient Affected by Extravasation Injury from Hypertonic Solution

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Abstract. *In neonatal intensive care units, extravasation is one of the most common injuries occurring in infants as a complication of infusion therapy. These very preterm infants have immature skin which is easily damaged. They often require a longer duration of intravenous therapy, and obtaining intravenous access can be difficult. An invasive treatment should be avoided, whenever possible, particularly for very immature infants. In our Special Operative Unit for ulcers and difficult-to-heal wounds, University of Rome, we successfully treated a premature neonate, who experienced extravasation of hypertonic fluid, using dermal substitutes and products of regenerative medicine.*

Extravasation of intravenous fluids is the non-intentional leakage of infused fluid into the surrounding tissue, causing damage. The incidence of extravasation of intravenous fluid after routine infusions in children and babies has been reported as 11-58% (1, 2). The prevalence of extravasation injuries that cause skin necrosis is 38‰ (3). Most injuries occur in infants of less than 26 weeks gestation (3). This is most likely due to the immature skin of these infants. Hypertonic solutions, like alimentation solutions, may cause osmotic imbalance, impaired cell function and lead to tissue injury. Tissue extravasation can occur with varying degrees of morbidity. The severity of damage depends on the volume of the substance infused, the stage of infusion and the nature of the fluid extravasated. The patient with an extravasation injury in moderate cases can present with local swelling, erythema, blistering and pain; in severe cases ulcers and damage of the underlying tissues, with subsequent functional and aesthetic impairments can occur. Preterm babies are especially at risk for severe damage from extravasation because of their prematurity, the fragility and small calibre of their peripheral veins (4). Furthermore, they cannot localize pain, thus allowing the infusion to continue unnoticed. If left untreated, these iatrogenic injuries may lead to extensive skin necrosis and damage to tendons, muscles, nerves and joints causing limb contractures (5, 6). Moreover, if extravasation is next to a major artery in the forearm or leg, vascular flow can be obstructed and amputation will be required (7). Injuries to the physis by extravasation is reported in literature and growth plate arrest have been described (8). The most common sites for extravasation injuries are the dorsum of the hand, the dorsum of the foot, the volar surface of the wrist and forearm, the leg, the scalp and the antecubital fossa. Most injuries are moderate and resolve spontaneously, and only a low rate (0.24%) of them are severe and result in loss of skin tissues, infections and involvement of underlying structures (6). Specific therapies are based upon assessment of the degree of the injury. The goal in managing tissue damage after extravasation is to improve tissue perfusion and prevent progression of tissue necrosis. Significant progress has been made over the years in the development of biomaterials to aid rapid wound closure. These therapeutic devices allow a conservative approach thus resulting in reduction of extravasation injuries occurring in neonatal intensive care units. Furthermore they are well-tolerated and atraumatic, and have the advantage of avoiding or delaying surgery.

We document the successful combined treatment using dermal substitutes and products of regenerative medicine of a patient affected by a severe injury from extravasation caused by a hypertonic solution.

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Key Words: Extravasation, premature neonates, dermal substitutes, regenerative medicine.

Case Report

Our male patient was born at 35 weeks gestation and weighted 2192 g. On admission to the neonatal intensive care unit, he had a diagnosis of respiratory distress and

equinovarus deformity of the right ankle. On the second day of life, he was affected with a full-thickness intravenous extravasation injury from parenteral nutrition administered *via* a peripheral cannula into the medial side of the right foot. Four days after extravasation, a plastic surgery consultation was undertaken. On objective examination the medial side of the leg and the dorsum of the foot appeared swollen and covered by wet scabs with blistering of the surrounding surface. The plant of the foot was characterized by oedema. The lateral side of the leg was noted to be erythematous (Figure 1). Initially the right limb was maintained in an elevated position in order to reduce swelling and allow vein drainage, and demarcation of the devitalized tissue was anticipated. Doppler ultrasound of the vessels did not show any vascular damage.

Three days after the first observation, eschars appeared demarcated and the blisters reddish. The subsequent treatment including cleaning and applications of topical silver sulfadiazine until the swelling had decreased and the erythema of the leg disappeared. After 8 days of dressing, on removal of the necrotic tissue, the wound bed looked vital and was covered by healthy granulation tissue thus allowing the subsequent application of an acellular dermal substitute, Hyalomatrix PA® (Fidia Advanced Biopolymers, ABANO TERME, Italy).

Contemporarily, a small skin biopsy was collected to initiate a culture of autologous keratinocytes for the coverage of the wound. After three weeks Hyalomatrix PA was removed and autologous keratinocytes were applied. The same procedure was repeated a month later. At the follow-up (two months), a good bioengineered graft take rate was observed. The inevitably resulting scars were apparent at nine months and the equinovarus deformity of the right foot had worsened. The patient underwent surgery for correction of the deformity and reconstruction of the scars. At the same time, a cellular dermal substitute Integra Dermal Regeneration Template (SIAD Healthcare S.p.a, Assiago(MI), Italy) was applied to the open area and a skin biopsy for autologous cultures was collected in order to cover the wound later with autologous keratinocytes and fibroblasts (Figure 2). Three weeks later, the neo-dermis was covered with the cultured autografts (Figure 3), with a 80% take observed at 5 days of follow-up. At three months after surgery, and at 14 months after the extravasation episode, the wound had healed completely without any sequelae (Figure 4).

Discussion

Extravasation is a serious complication of the peripheral intravenous therapy commonly used in neonatal care. An invasive treatment should be avoided, whenever possible, particularly on very immature infants. Advances in tissue bioengineering have led to the development of biomaterials that are innovative and suitable for managing wounds. Dermal substitute biomaterials provide a neodermis ready for the subsequent application of a skin grafting or cellular cultures as products of regenerative medicine. Such materials can be divided into cellular and acellular, the latter based either on allogeneic, xenogeneic or synthetic materials.

Hyalomatrix PA® (9-12) is an acellular dermal substitute based on synthetic materials. It is composed of two layers; the internal layer is a three-dimensional dermis-like matrix consisting of fibres of a hyaluronic acid ester called Hyaff, while the external layer is a flexible and transparent elastomer film that operates as a semi-permeable barrier to external agents. The biological matrix acts by modifying the chemical and physical characteristics of hyaluronic acid, a naturally occurring extracellular matrix playing an important role in the processes of tissue regeneration (13, 14). The transparent silicone membrane prevents liquid loss (15) and allows monitoring of the wound without changing medication. It is used to cover the wound directly or to prepare the wound bed for subsequent skin graf or cellular cultures. Integra Dermal Regeneration Template (16) is an acellular dermal substitute based on allogeneic materials. It is made of bilaminate material, a collagen chondroitin sponge overlayed with silicone. When Integra is placed on a healthy wound, histioblasts migrate into the material from the base of the wound, activating angioblasts, and then creating a living tissue. The change in colour of the matrix is a predictor of its vascularization. When the colour has progressed from pink through pale yellow and finally to peach, vascularization and neo dermis formation are complete. This usually occurs within 15-20 days, after which the silicone layer is peeled off and thin epidermal autografts can be placed on the neodermis. Post -operative care is minimal because histogenesis is observable through the silicone. This biomaterial was successfully clinically tested in managing burn wounds in 1981 (17) and since then has become widely adopted for treatment of full-thickness burn-treatment (18) clinically becoming a 'gold standard' dermal substitute biomaterial. Advantages of the product include its long shelf life, simple handling, low risks of immunogenic response and disease transmission, good cosmetic outcomes with reduced rates of contraction and scarring (19). Disadvantages consist of meticulous surgical preparation of the wound bed to guarantee a good take of Integra, the inability to use it on infected wounds, the relatively long time of 10-14 days required for vascularization and, the necessity for a second operative procedure to achieve permanent wound closure with a skin graft. In attempts to achieve a single-stage surgical procedure, the product has been seeded with disaggregated cultured (20) or non-cultured (21) autologous keratinocytes, using *in vivo* experimental models. Results were promising, but further clinical follow-up is required.

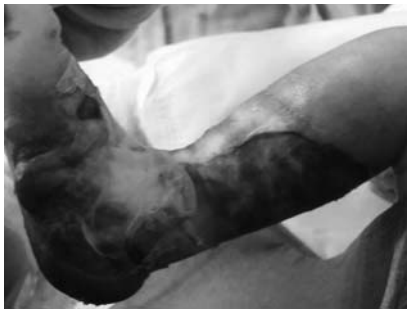

Figure 1. *Appearance of the wound after extravasation.*

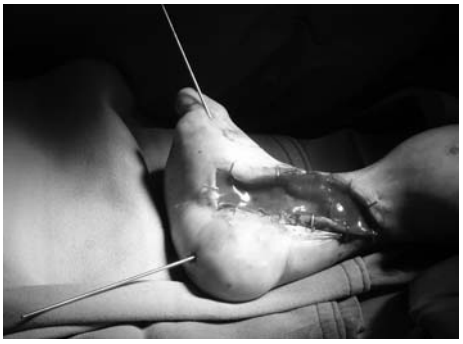

Figure 2. *Application of Integra.*

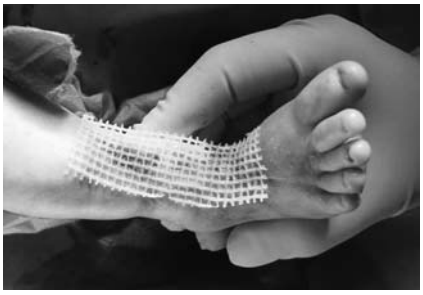

Figure 3. *Application of the cultured autografts.*

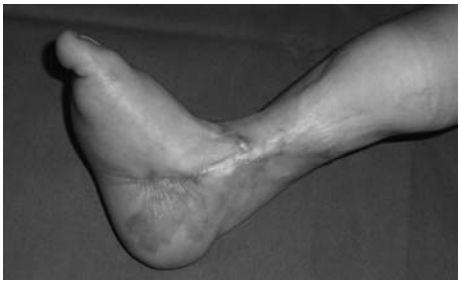

Figure 4. *Follow-up at 14 months.*

Significant progress has been made recently in the development and clinical use of cultured autologous fibroblasts and keratinocytes (22, 23). These products of regenerative medicine consist of autologous keratinocytes cultured on a scaffold made of hyaluronic acid, which promotes fibroblast and keratinocyte migration and proliferation. Hyaluronic acid is also reported to participate in scarless foetal wound healing (24). Preliminary studies using such products have shown promising potential, giving a good graft take rate and biocompatibility, as well as low infection rates.

Patients with full-thickness wounds suffer from a substantial loss of dermal tissue. In general, these wounds are grafted with autologous split-skin grafts or cultured keratinocytes. Nevertheless, the lack of a dermal component often negatively influences the outcome because split-skin grafts alone often fail to correct contour defects, are prone to contractures, and can lead to a poorly developed dermo-epidermal junction (25, 26). In addition, cultured epithelial grafts are also known to regenerate the dermo-epidermal junction slowly and to blister and ulcerate for several months after grafting (25, 27). It is now generally accepted that a dermal component is needed to improve the final outcome of cultured epithelial grafts (28, 29). Most artificial skin substitutes include autologous fibroblasts, providing the healing wound with growth factors and cytokines. It also lays down extracellular matrix components 'conditioning' the wound for split skin grafting. Fibroblast autologous bioconstructs are primarily used with keratinocyte autologous epidermal bioconstructs (30). They represent cellular dermal substitutes and it has been reported that they enhance keratinocyte take, and reduce hypertrophy and wound contracture rates when compared with exclusive application of keratinocyte cultures (31).

Conclusion

On the basis of our experience, the combined treatment using dermal substitutes and products of regenerative medicine allowed us to achieve a complete healing of a full-thickness wound caused by extravasation of a hypertonic solution. We used an acellular dermal substitute Hyalomatrix PA®, for immediate wound coverage after the removal of necrotic tissues, to protect the residual dermal layer and stimulate the re -epithelialization from the wound edge and skin appendages. This allowed the wound bed to be protected and maintained ready for treatment with cultured autologous keratinocytes.

Moreover, since our patient was unable to undergo surgery because of his severe clinical condition, Hyalomatrix PA® allowed application in the incubator. The results obtained were a complete healing of the wound, with an inevitable debilitating scar. The surgical reconstruction consisted of the removal of the scar and the application of an acellular dermal substitute as Integra. The results achieved suggest that the acellular artificial dermis can effectively promote dermal

regeneration and epithelial migration, and the newly secreted human extracellular matrix appeared to serve as a stable matrix for the attachment and proliferation of cultured cells. Furthermore, it provided a suitable thickness of neodermis for a full-thickness wound such as the one in our patient. The use of cultured autologous keratinocytes and fibroblasts allowed us to accelerate the wound healing. We strongly feel that the management of neonatal patients affected with extravasation injuries should involve a multidisciplinary team of consultants in pediatric medicine, vascular surgery and plastic surgery, and a specialist nurse, thus avoiding severe complications deriving from such injuries in preterm neonates.

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Received June 5, 2011
Revised October 17, 2011
Accepted October 19, 2011