Aseptically Processed Dehydrated Human Amnion/Chorion Allograft* Facilitates Closure of Chronic Wounds in a Diabetic Swine Model

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INTRODUCTION

Amniotic membrane has been employed in the treatment of wounds for over 100 years[1,2]. Human amnion/chorion membranes derived from the placenta are rich in collagens and various growth factors that support the healing process to both improve wound closure and reduce scar formation[3].

In addition, this tissue has unique properties including antimicrobial activity and the lack of immunologic markers[3,4]. Terminal sterilization has been well-documented over time to negatively impact allogeneic tissue properties[5,6].

Improved processing techniques and lack of terminal sterilization aid in preservation of the natural factors contained in the membrane, while ap-propriate dehydration techniques can provide a stable off-the-shelf solution for the treatment of wound care.

Objectives: The objectives of this study were to 1) determine the presence of matrix components and growth factors in a dehydrated placental tissue allograft containing both amnion and chorion and 2) test the efficacy of the allograft in facilitating wound closure in a diabetic swine model.

MATERIALS AND METHODS

Dehydrated placental tissue allograft containing both the amnion and the chorion layer was processed aseptically without terminal sterilization at the Musculoskeletal Transplant Foundation (MTF, Edison NJ).

Representative samples of the allograft were characterized via immuno-histochemical and dye staining at a 3rd party histology laboratory for matrix proteins, growth factors and cytokines that support wound healing and act as anti-inflammatory and anti-microbial mediators.

In vivo efficacy of the dehydrated placental tissue allograft was evaluated in a diabetic swine wound model (Sinclair Research, LLC Auxvasse MO). Full thickness square skin wounds measuring 3cm x 3cm were created on the dorsal-lateral area of diabetic pigs. Allograft tissue was implanted in full thickness wounds with non-adhesive dressing and explanted at 14 and 28 days for histological analysis using H&E and Masson's trichrome stains. Wound closure was also assessed over time by photographs of the wounds at every dressing change.

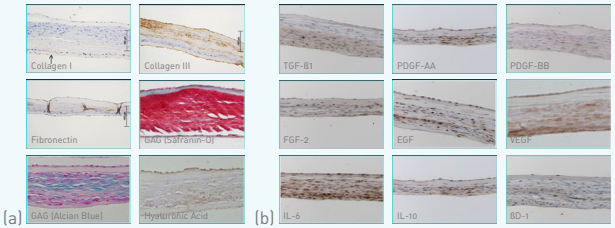

Figure 1:(a) Immunohistochemical and dye staining of dehydrated amnion/chorion allograft for matrix components show presence of collagen I, collagen III, fibronectin, GAGs, and hyaluronic acid; (b) Growth factors and cytokines TGF-ß1, PDGF-AA, PDGF-BB, FGF-2, EGF, VEGF, IL-6, IL-10, and ß-defensin-1 (ßD-1) are present in the allograft

When implanted in a diabetic swine wound model with full thickness wounds, the amnion/chorion allograft supported progressive wound closure over the 28 days of the study, with granulation tissue and re-epithelialization at 14 days (Figure 2), followed by complete re-epithelialization of the wound bed at 28 days.

H&E and Masson's trichrome staining (Figure 3) confirms granulation tissue filling the wound bed and re-epithelialization and revascularization of the wound by 28 days. Re-epithelialized areas resembled tissue structure of non-wounded epidermis. No necrosis was noted, indicating a healthy wound healing process via granulation tissue.

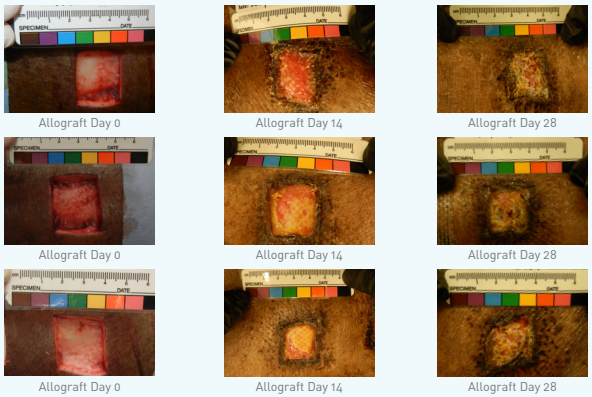

Figure 2. Initial application of allograft to full-thickness wounds in diabetic pig and wound healing progress at 14 and 28 days. Pink granulation tissue can be seen within the wound bed at 14 days and re-epithelization at the wound edges. The wounds are fully closed and re-epithelialized by 28 days.

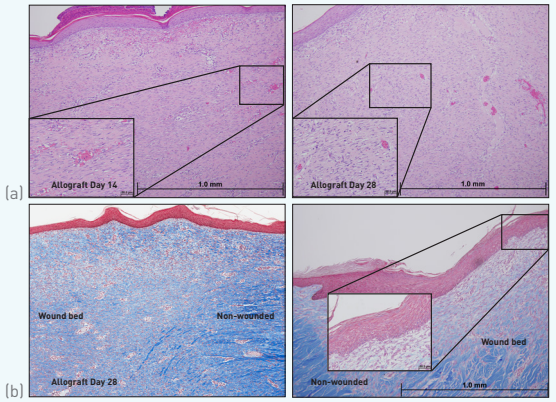

Figure 3. (a) H&E staining shows re-epithelialization and some revascularization by 14 days with further progression by 28 days. (b) Masson's trichrome showing granula-tion tissue filling the wound bed by 28 days. The wound has been completely re-epithelialized with very similar tissue structure as non-wounded epidermis.

CONCLUSION

Aseptically processed dehydrated amnion/chorion allograft without terminal sterilization retains growth factors and collagen-rich matrix that support the treatment of wounds. A diabetic pig model with full thickness wounds demonstrated that the graft facilitates wound closure, with granulation tissue forming by 14 days and re-epithelialization/revascularization by 28 days.

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*AmnioBand™ Membrane; MTF, Edison, NJ