Dehydrated Amnion/Chorion Membrane Contains Key Extracellular Matrix Proteins and Maintains *In Vitro* Properties Following Degradation

Katrina A. Harmon[1], Kelly A. Kimmerling[1], and Katie C. Mowry[1]

[1]Organogenesis Discovery Center, Birmingham, AL

INTRODUCTION

With long-documented use in various wound applications, placental-derived allografts have been shown to retain extracellular matrix (ECM) components for use as a protective barrier. Here, a minimally manipulated dehydrated amnion/chorion membrane (dACM*) was evaluated to assess the retention of its biophysical and barrier properties in addition to the impact of *in vitro* degradation of the ECM scaffold.

*NuShield®, Organogenesis, Canton, MA

METHODS

- Biophysical structure and barrier properties were evaluated using immunohistochemistry and ECM arrays.
- Degradation characteristics, including mass loss and collagen content, were assessed using an *in vitro* simulated wound fluid (SWF) degradation model for up to 10 days.
- To evaluate retention of barrier properties, an *in vitro* primary human dermal fibroblast model was used to evaluate cell attachment on intact (non-degraded) and degraded dACM.

BIOPHYSICAL STRUCTURE AND BARRIER CHARACTERIZATION

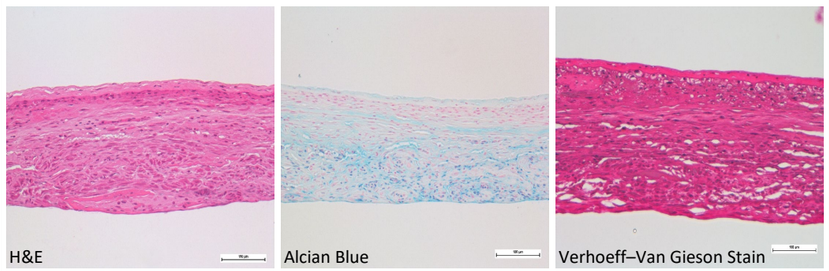

Figure 1. Biophysical structure and barrier characterization of dACM. Histological evaluation of dACM including H&E, Alcian Blue, and Verhoeff-Van Gieson staining. Scale bars = 100 µm.

BIOPHYSICAL STRUCTURE AND BARRIER CHARACTERIZATION

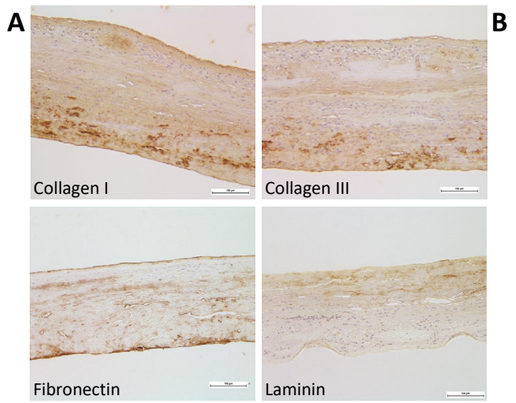

Figure 2. Biophysical structure and barrier characterization of dACM. (A) Immunohistochemical characterization of dACM including Collagen I and III, Fibronectin, and Laminin. Scale bars = 100 µm. (B) dACM retains extracellular matrix components of a protective barrier.

IN VITRO BARRIER PROPERTIES OF dACM

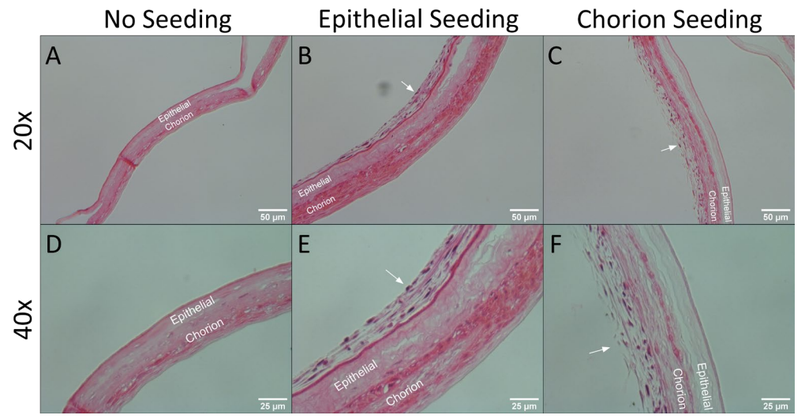

Figure 3. Histological evaluation of dACM highlighting barrier properties following seeding of fibroblasts on the epithelial (B,E) or chorion side (C,F). White arrows denote cell attachment onto dACM. Images were taken at 20x (A,B,C; 50 µm scale bar) and 40x (D, E, F; 25 µm scale bar).

IN VITRO BARRIER PROPERTIES FOLLOWING DEGRADATION

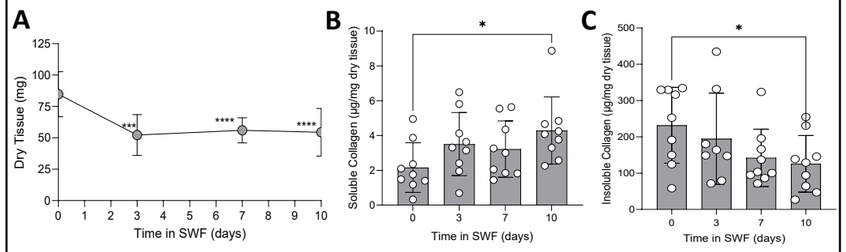

Figure 4. Physical barrier properties after exposure to SWF in an *in vitro* model. (A) Dry tissue weight and (B) soluble and (C) insoluble collagen content were evaluated over 10 days. Average ± stdev reported. *P≤0.05, *** P≤0.001, ****P≤0.0001 compared to day 0.

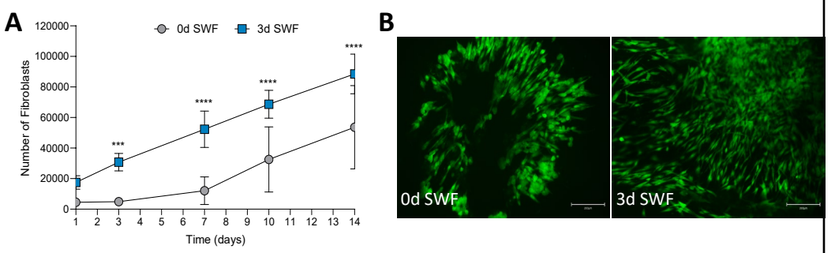

Figure 5. *In vitro* barrier properties after exposure to SWF. (A) Fibroblast attachment and growth over 14 days on intact (0d) and 3-day (3d) SWF-degraded dACM, (B) representative Calcein AM-stained images of fibroblasts after 3 days of attachment (20x; 200 µm scale bar).

CONCLUSIONS

- dACM is a minimally manipulated, protective barrier that retained the components of native tissue.
- dACM is resistant to rapid *in vitro* degradation and matrix properties highlighting its role as a protective barrier.
- Matrix properties were maintained throughout degradation as evidenced by fibroblast attachment and growth.
- Overall, results demonstrate the retention of native structure and barrier properties of dACM and highlighted its ability to maintain properties throughout *in vitro* degradation.

ORGANOGENESIS