Principles, Practices and Prospects for Burn Wound Management

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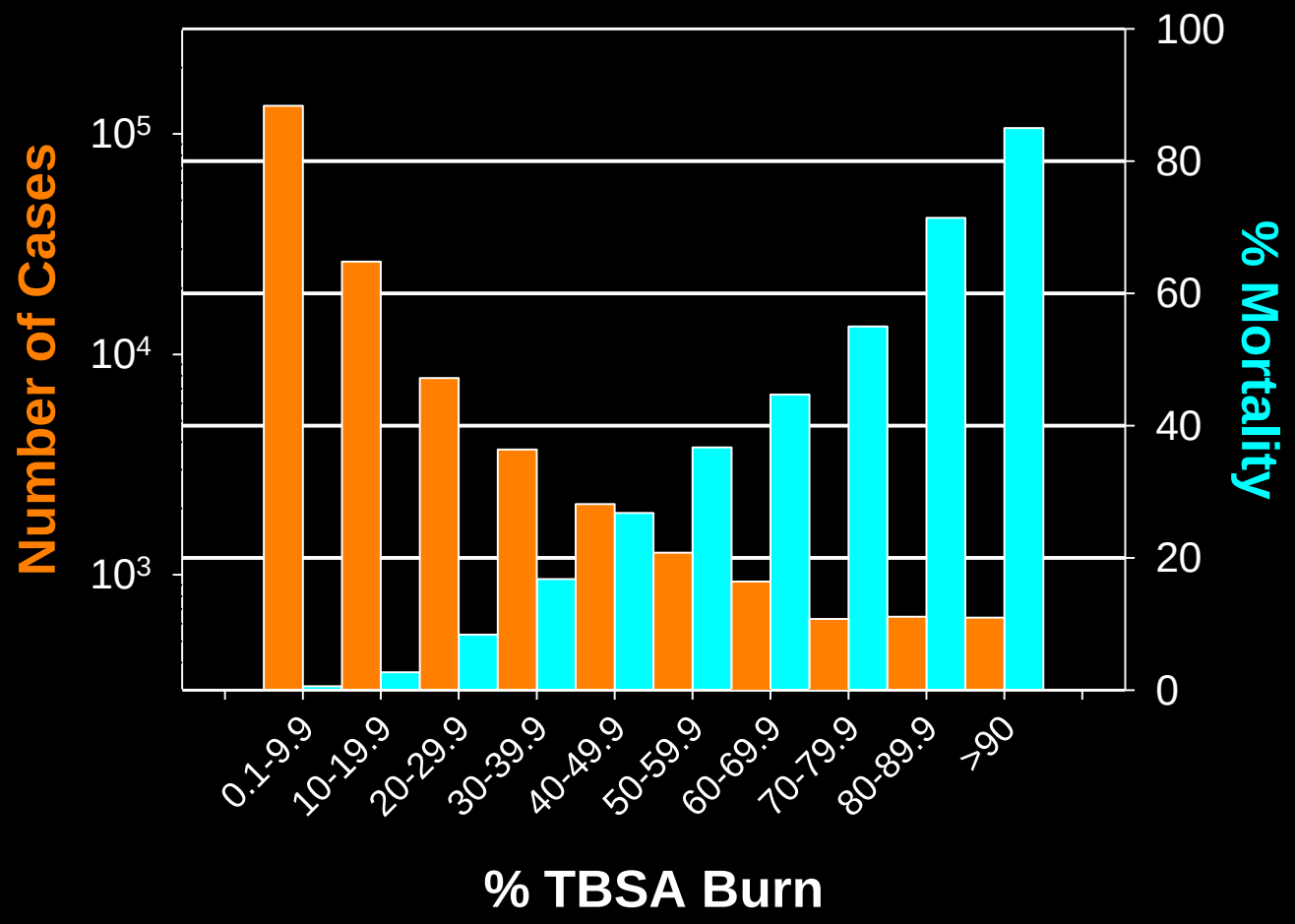
Disclosure

- ANZBA travel support provided by Skin Tissue Engineering

SKIN
Tissue Engineering

Burn Size Group (% TBSA) vs Number of Cases and % Mortality

National Burn Repository: 2015 Report Amer Burn Assn; n=172,550

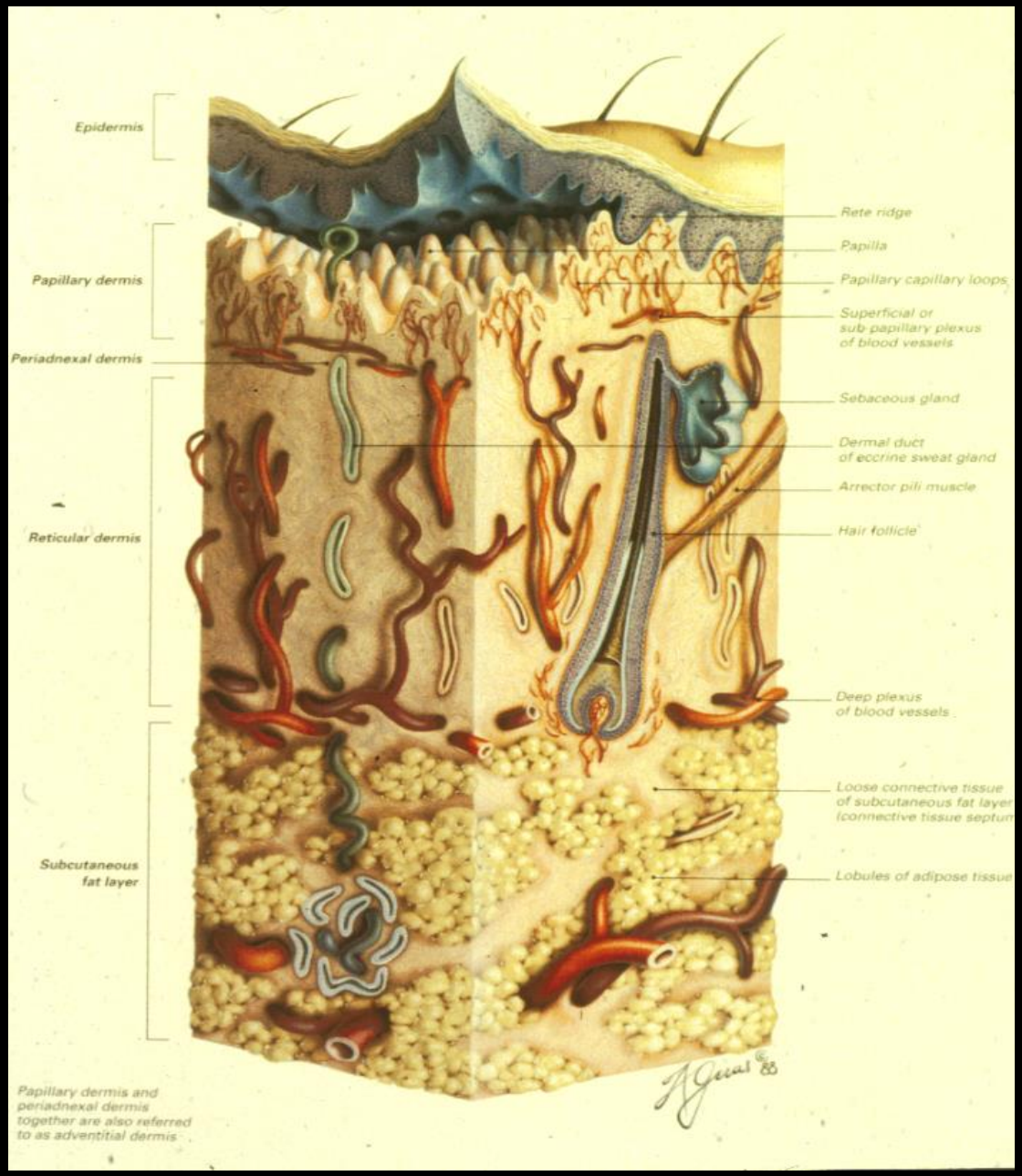


Medical Objectives

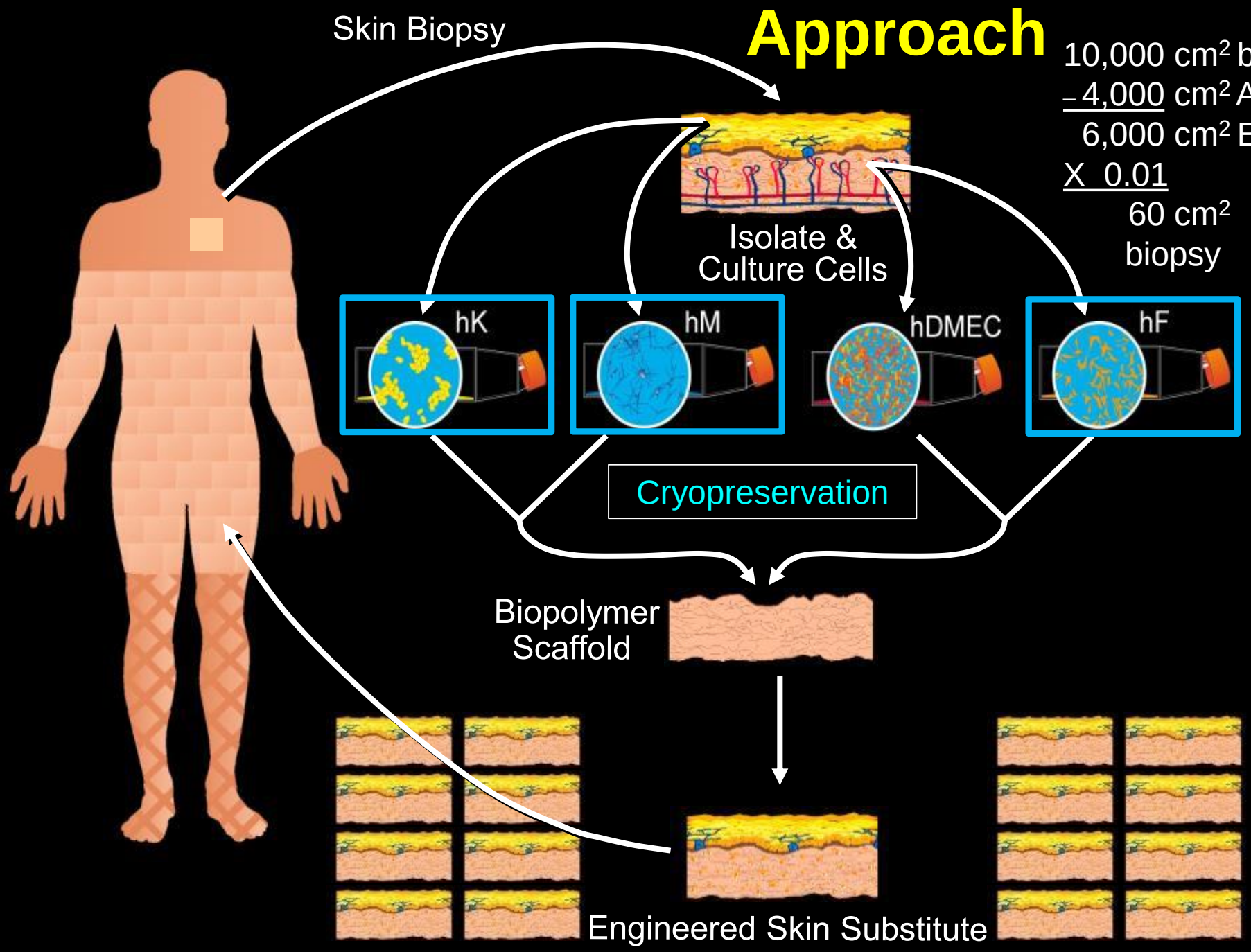
- Survival
- Earliest closure
- Least donor skin
- Minimum scar
  - Acute burns
  - Burn scars
  - Congenital lesions
  - Chronic wounds

Human Skin:
Structures & Functions

- **Three B's:**
  - **Barrier**
  - **Basement membrane**
  - **Blood supply**



Approach


Skin Biopsy
10,000 cm2 burn
– 4,000 cm2 AG
6,000 cm2 ESS
X 0.01
60 cm2
biopsy
Isolate &
Culture Cells
hK
hM
hDMEC
hF
Cryopreservation
Biopolymer
Scaffold
Engineered Skin Substitute

Ideal Properties of Biopolymers

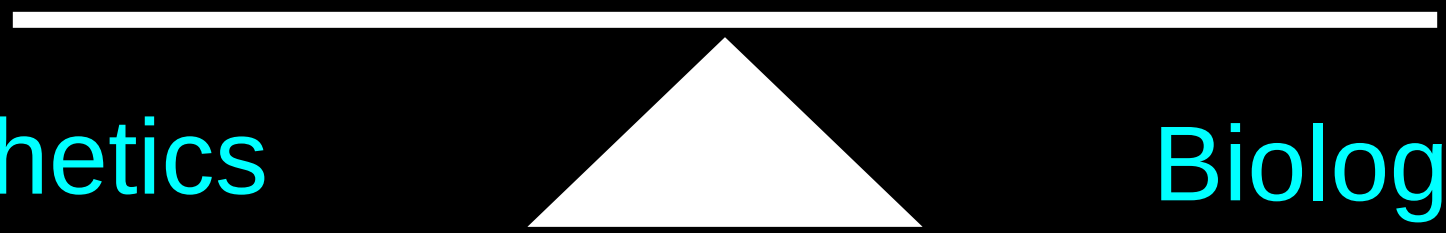
- Biocompatibility (low immunogenicity)
- Proper degradation rate for indication
- Self-assembly into higher order structures
- Appropriate mechanical properties
- Cell adhesion, migration, and proliferation
- Dry storage (banking)

Acellular Polymers & Matrices: Chemical Engineering

- Non-degradable
  - metals, alloys
    - stainless steel
    - titanium alloys
  - plastics
    - PTFE
    - HDPE
    - bone/dental cements
  - ceramic/glass composites
- Degradable
  - synthetic
    - PGA/PLA (Vicryl, Dexon)
    - PCL
    - PU
  - tissue-derived
    - collagens / gelatin
    - fibrin
    - hyaluronan
    - elastin

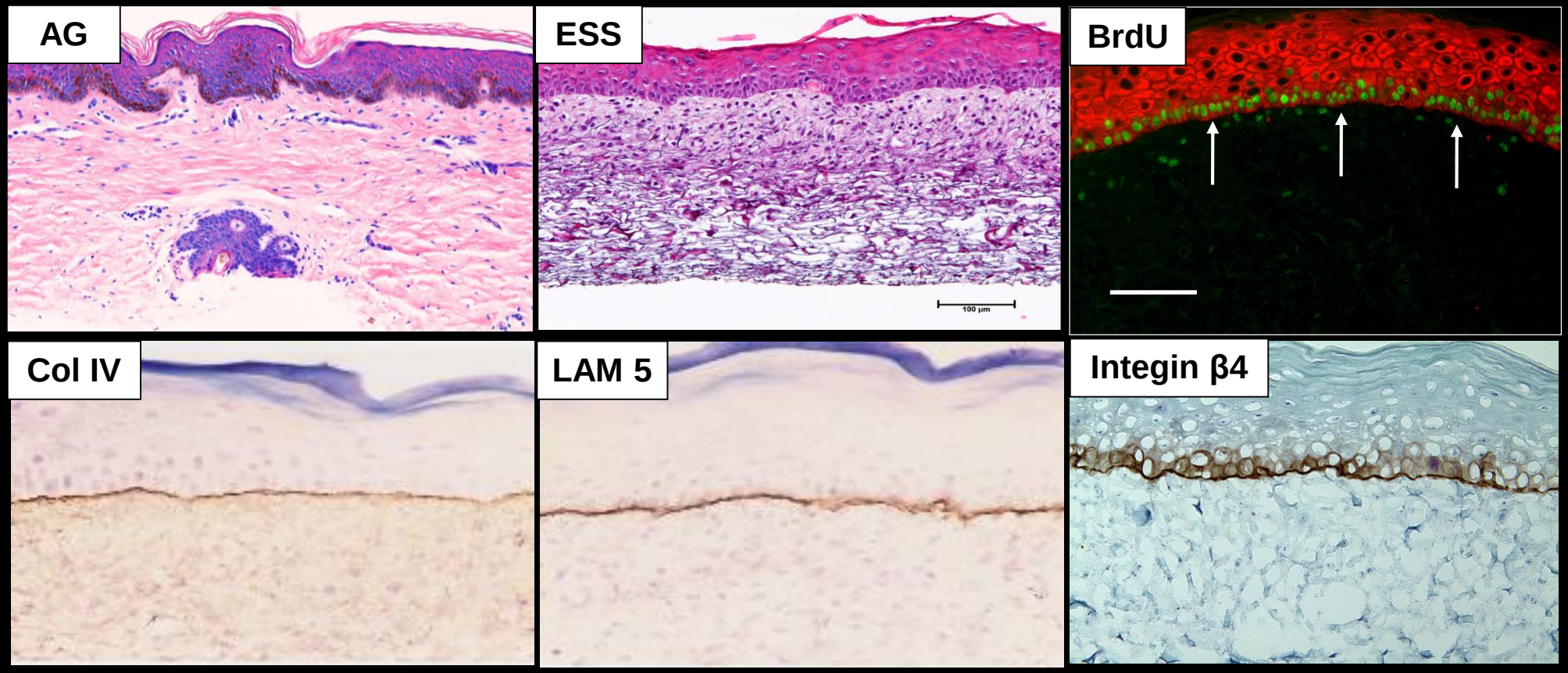
Challenge of Degradable Polymers

Synthetics	Biologics
Simple chemically	Complex biochemically
Pure pharmaceutically	Not pure pharmaceutically
Very reproducible	More variable
Biologically deficient	Biologically efficient
May be immunoreactive	Much less immunoreactive
Less biocompatible	More biocompatible



Balance

Dermal-Epidermal Junction in ESS

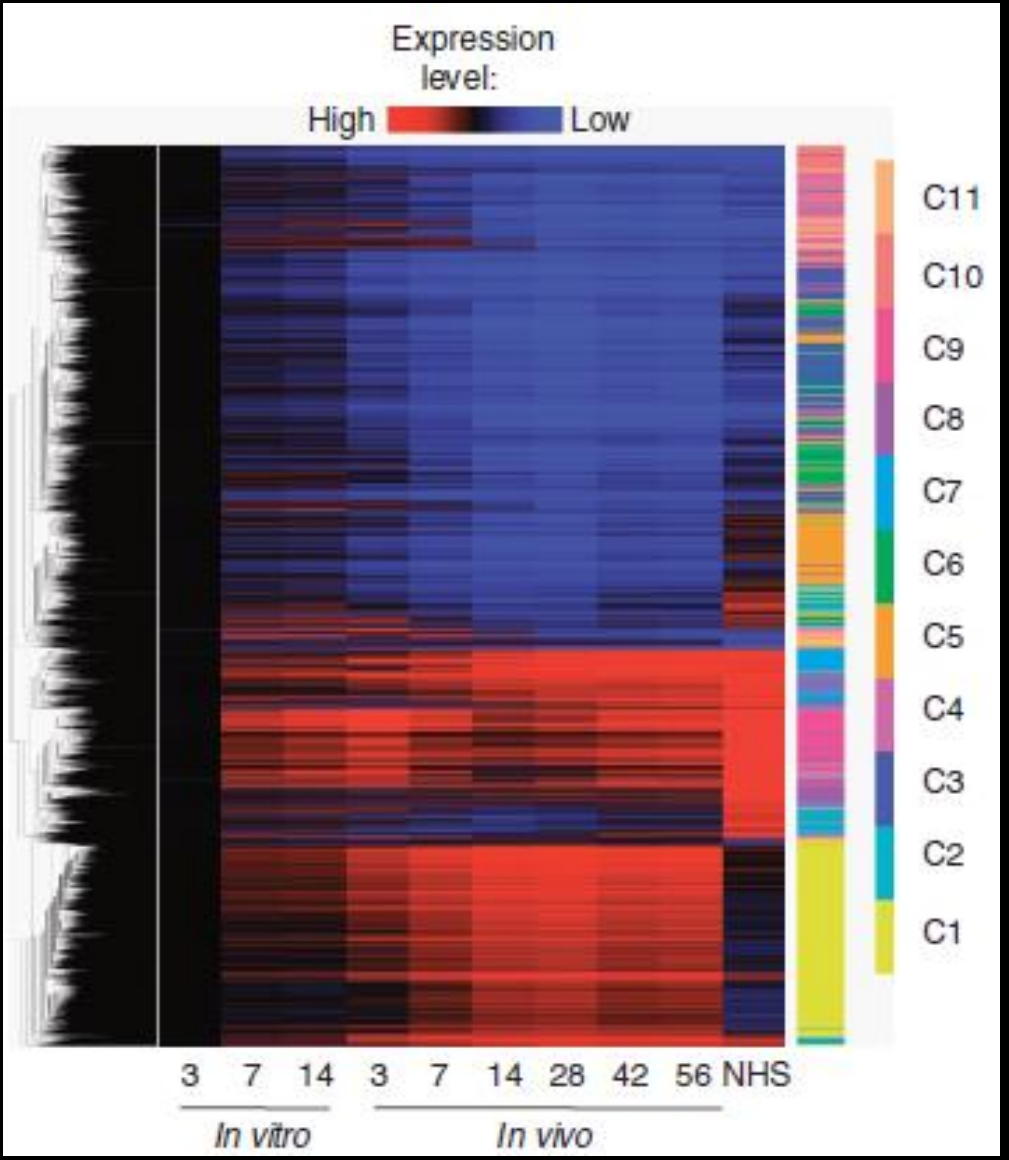


Boyce et al, J Invest Dermatol 118:565-572

Major Shifts in Gene Expression of ESS ± Grafting

Klingenberg et al, JID, 2010

- 10,359 probe sets
- 11 expression clusters
- normalized to d3 in vitro
- through 8 wks in vivo



Study Objectives

Compare an autologous engineered skin substitute (ESS) to meshed and expanded, split-thickness skin autograft (AG) for burns greater than 50% TBSA.

- Randomized, paired-site comparison to determine the:
  - percentage engraftment (POD 14)
  - ratio of closed wound to biopsy area (POD 28)
  - percentage TBSA covered
  - correlation of % TBSA closed with %TBSA FT burn
  - frequencies of regrafting
  - antibody production to the biopolymer implant

Demographic Data of the Patient Population

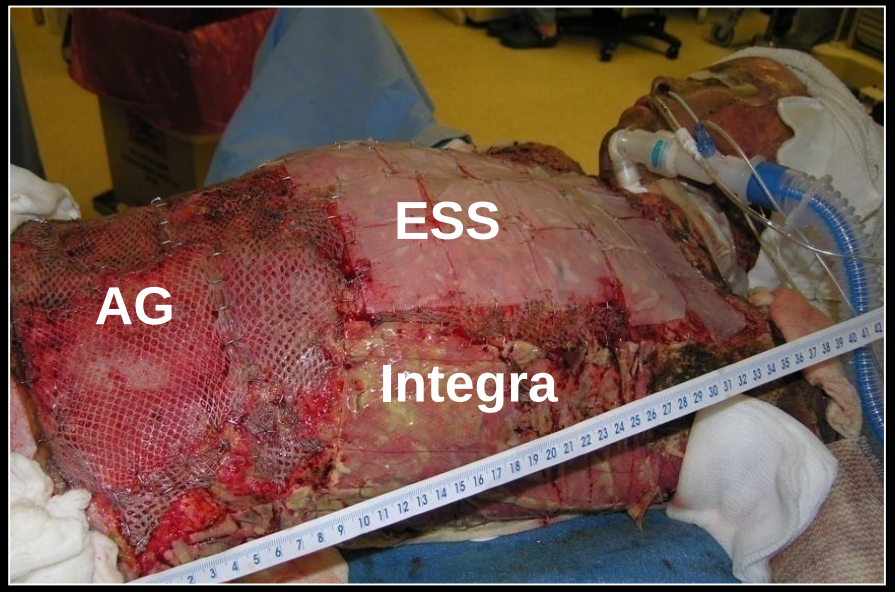
n = 16 enrolled

15 treated, 1 expired pretreatment

Parameter	Mean ± SEM	Range
Age (years)	6.3 ± 1.1	1.4 - 17.5
Gender (M/F)	(14 / 2)	n.a.
TBSA burn (%)	79.1 ± 2.2	59.5 – 95.0%
TBSA FT burn (%)	77.9 ± 2.4	58.8 – 95.0%
TBSA ESS/subject	33.4 ± 3.5	9.7 – 71.6%
Days to first ESS	32.1 ± 1.1	24 – 42

Study was performed under an
Investigative Device Exemption regulated by the US FDA.

Boyce et al, JBCR 38(2):61-70, 2017.



Surgical Methods

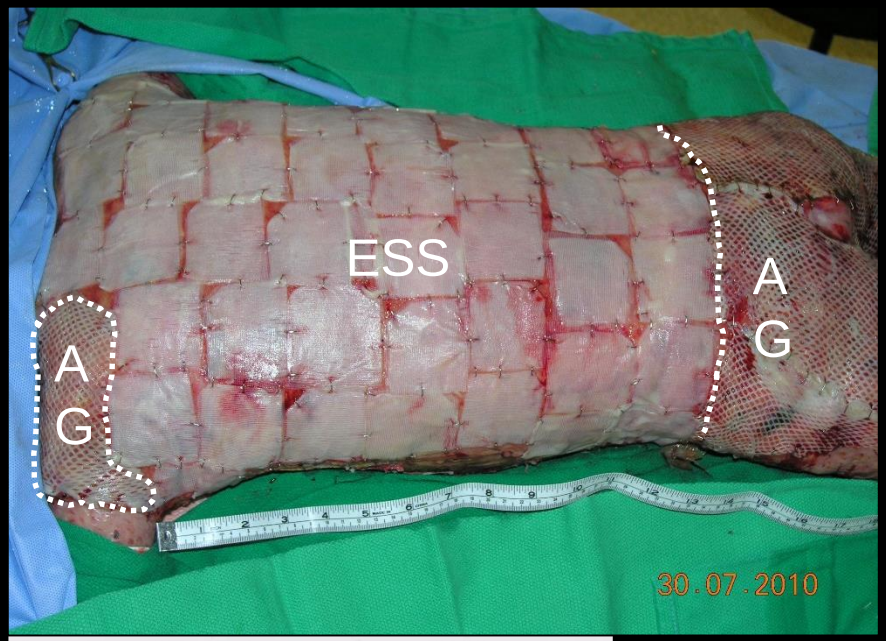
Lower left, ESS application

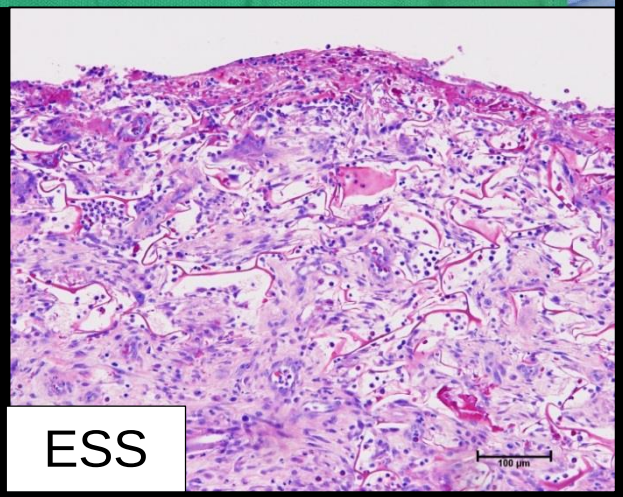
Upper left, Comparative grafting

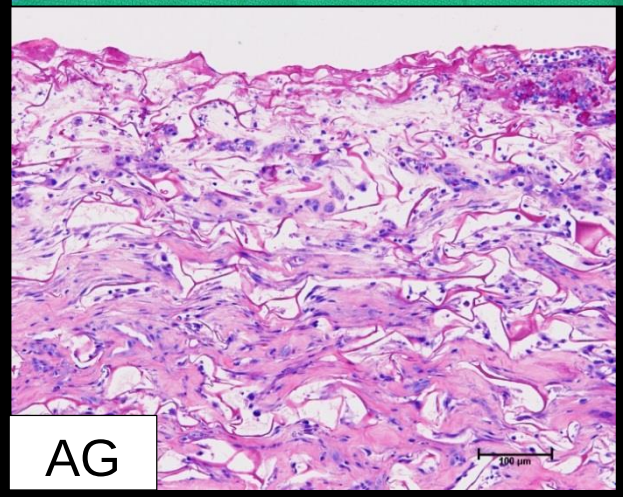
Upper right, Wet dressings
Non-cytotoxic antimicrobials

Surgical Methods: ESS and AG

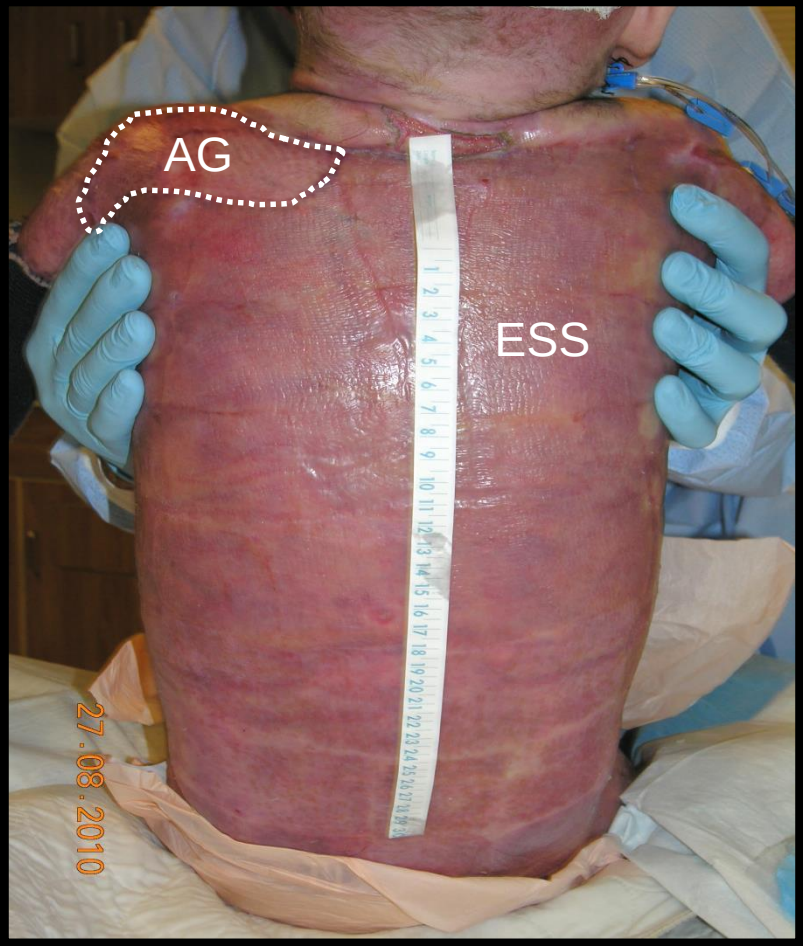
2056 ESS grafts, 4.89 m^2, 59 operative procedures



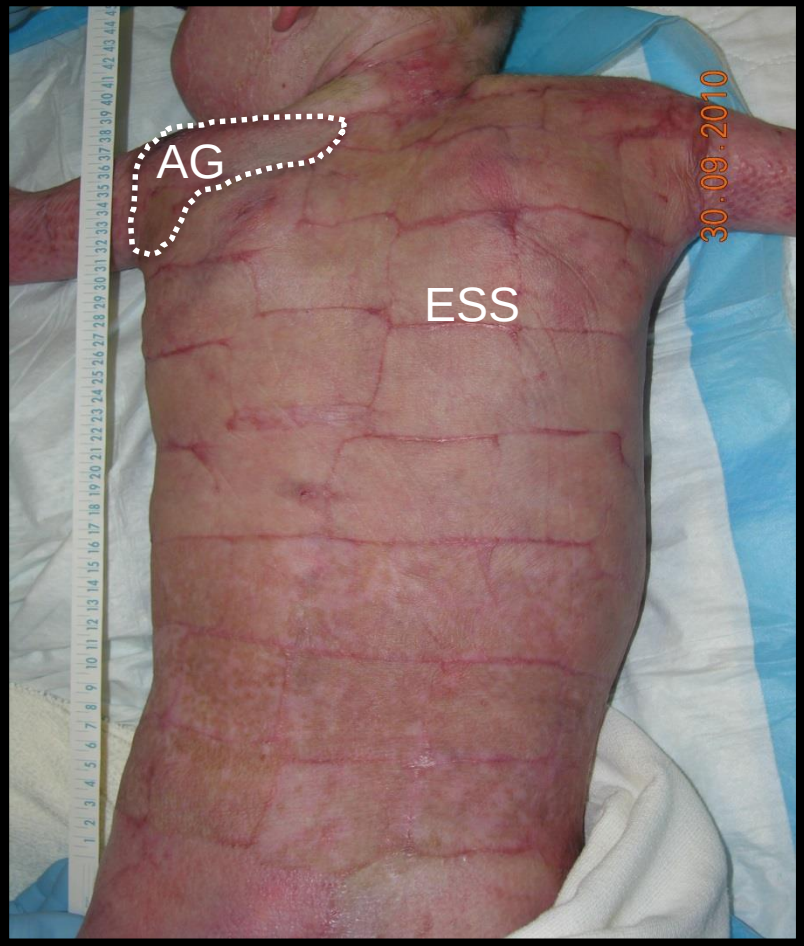




Results: Clinical Examination POD 28, 62

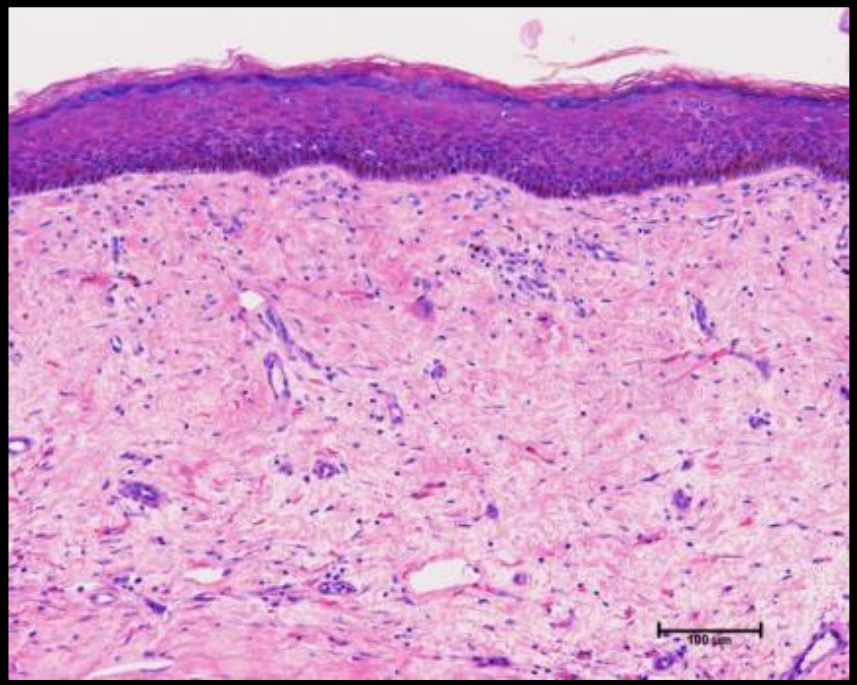


POD 28

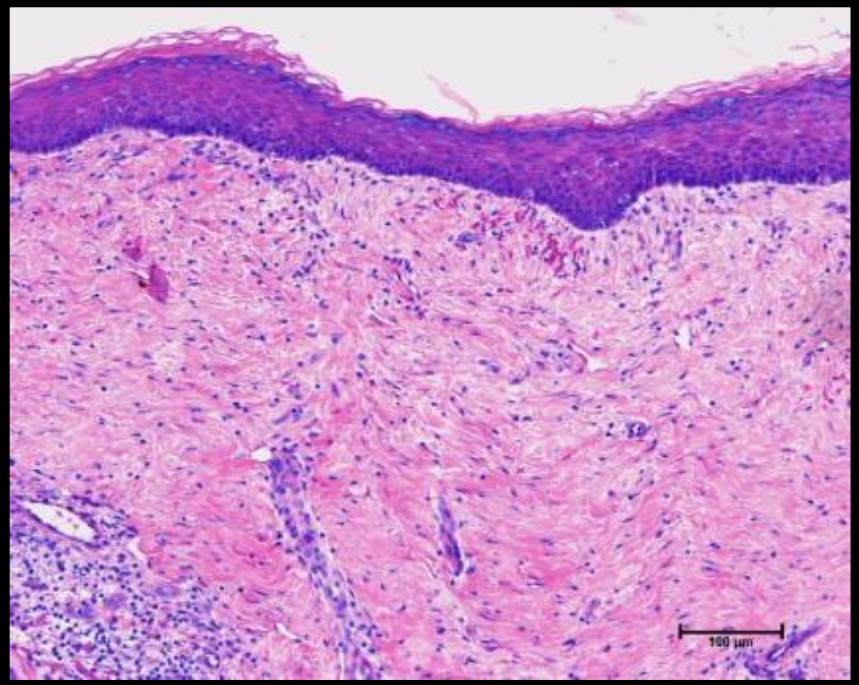


POD 62

Results: Histology, POD 105



ESS



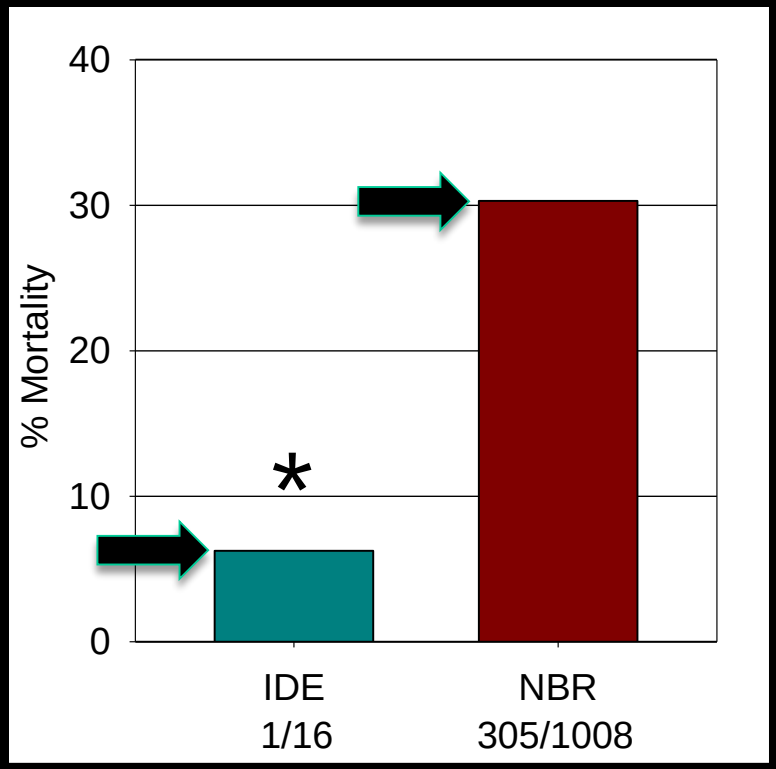
AG

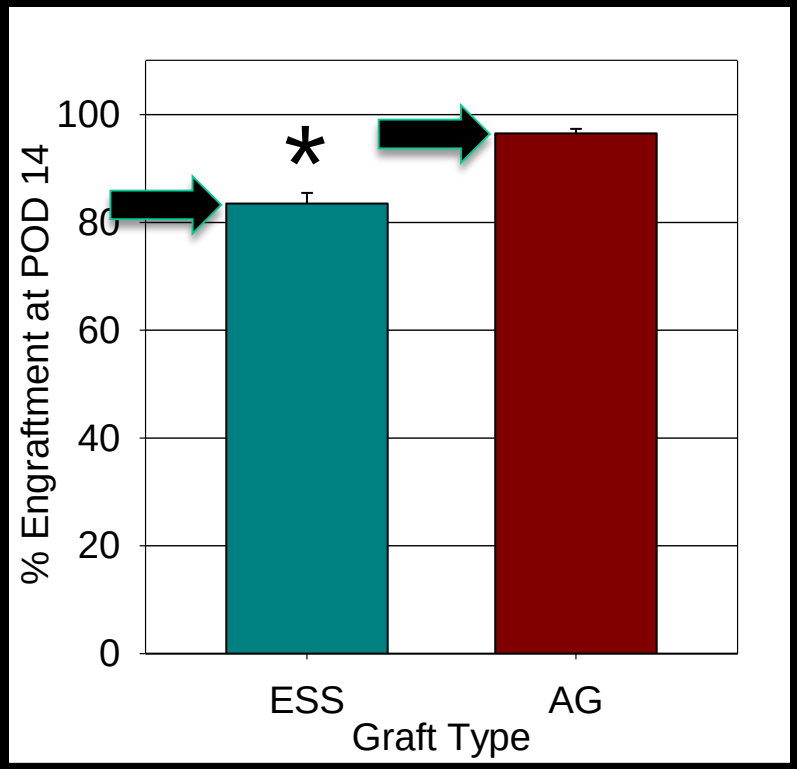
74% TBSA, FT Burn: POD 360

Upper body

Comparative sites

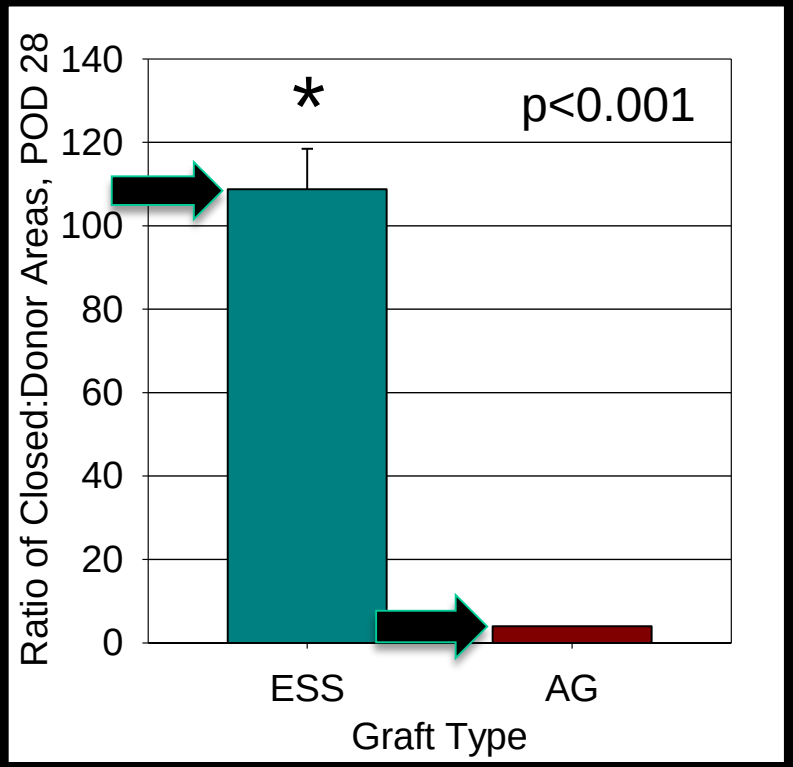
Results: % Mortality and % Engraftment (POD 14)

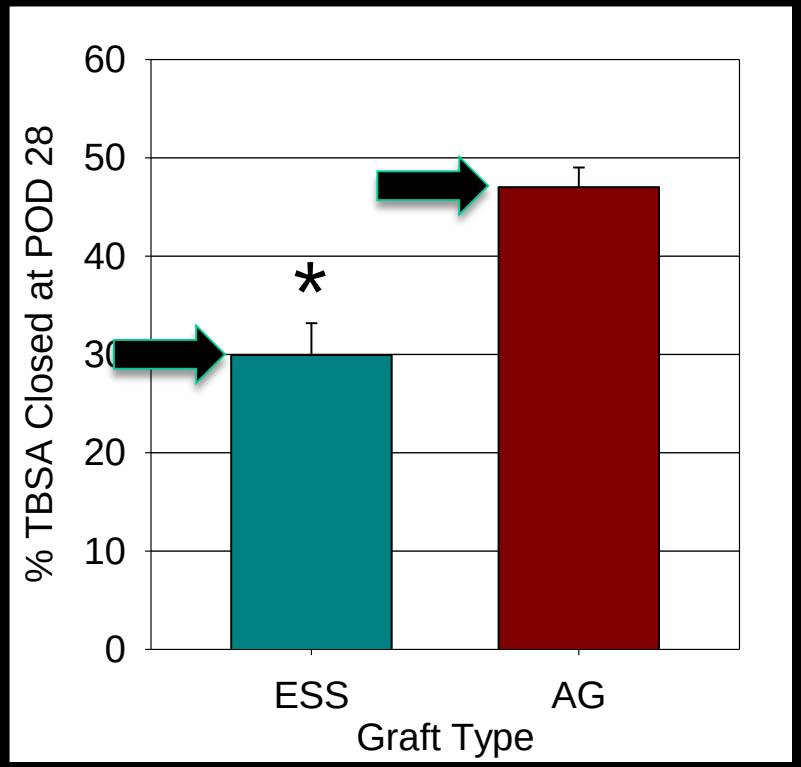




Boyce et al, JBCR 38(2):61-70, 2017.

Expansion Ratio and % TBSA closed (POD28)

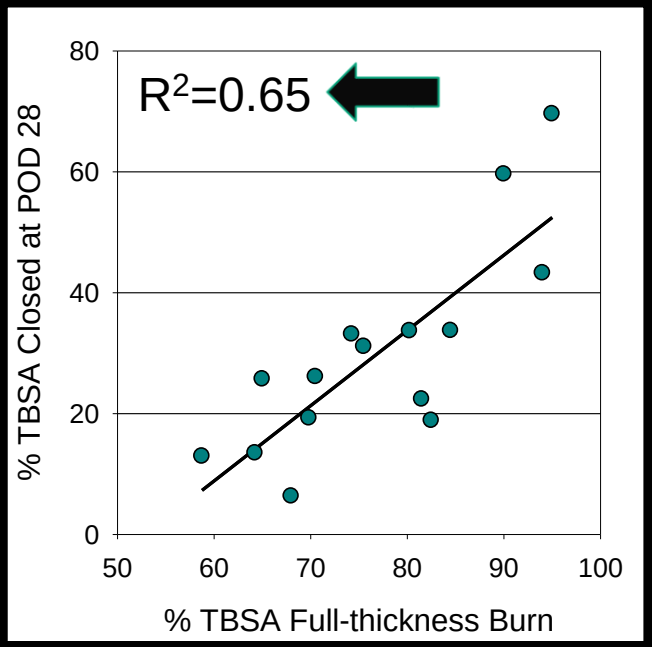


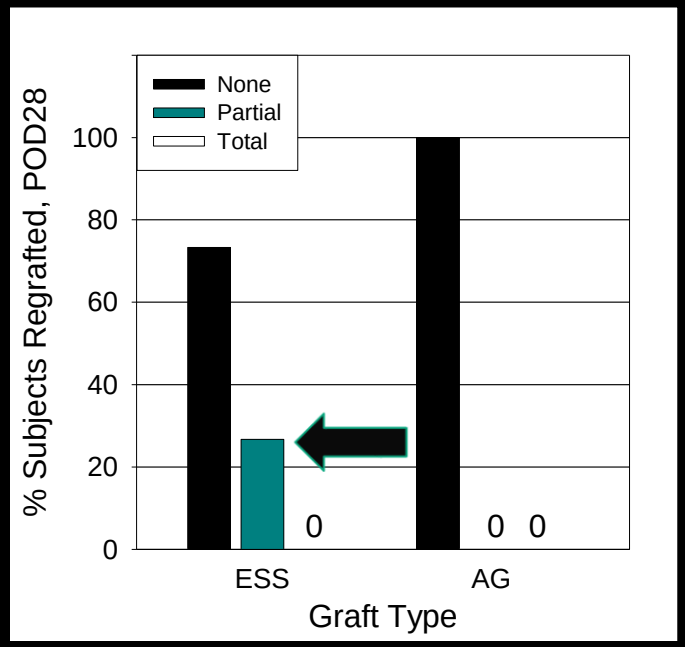


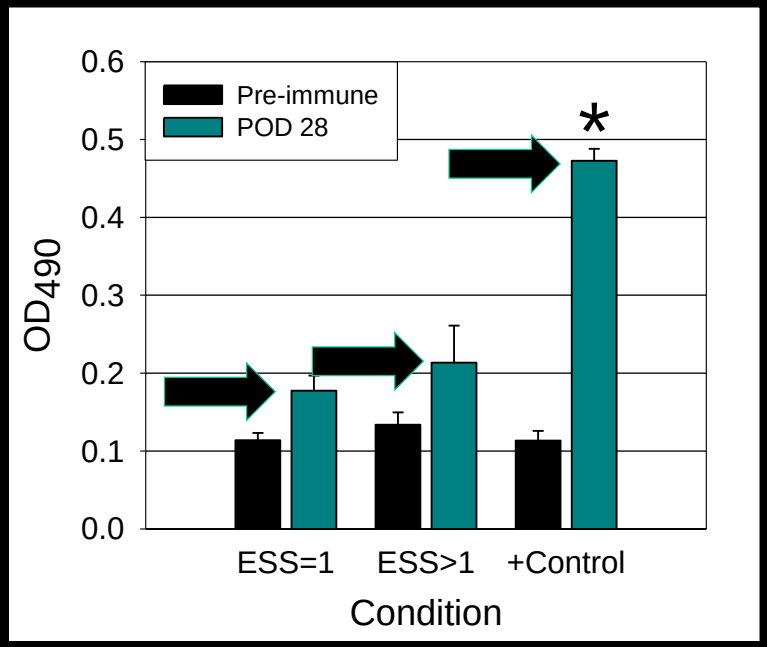
Boyce et al, JBCR 38(2):61-70, 2017.

Results: POD28

% TBSA burn vs % TBSA closed
Frequencies of regrafting
ELISA for bovine collagen







Boyce et al, JBCR 38(2):61-70, 2017.

94% TBSA FT Burn; Clinical Exam PBD 225

Recovery of Normal Activities after 94% Burns

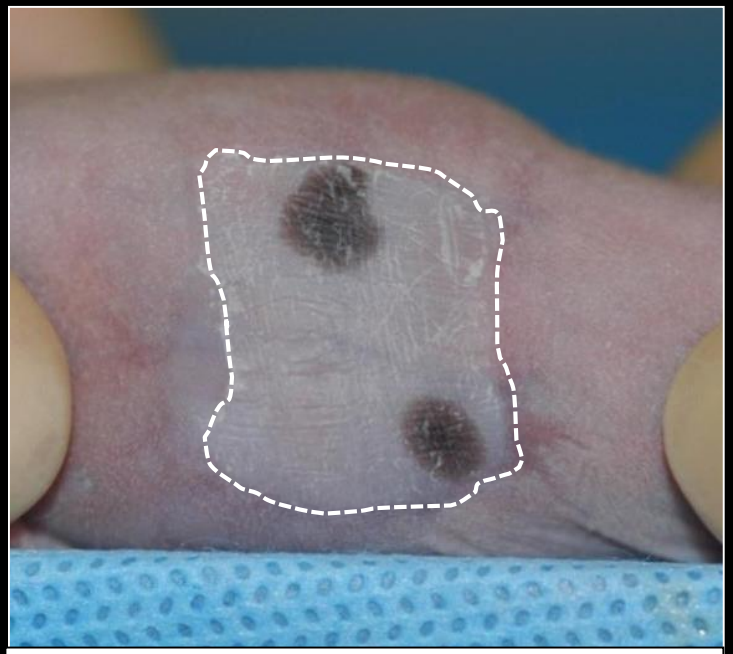
POD 405

Conclusions & Interpretations

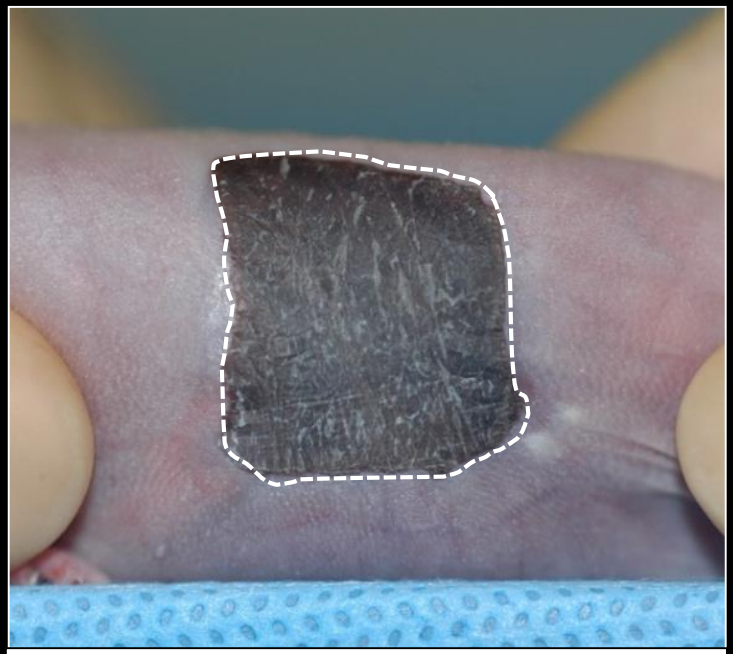
- Autologous ESS can reduce mortality and donor site harvesting in pediatric burns greater than 50% TBSA.
- Stable wound closure is interpreted to result from:
  - preservation of epidermal progenitor cells
  - formation *in vitro* of basement membrane between fibroblasts and keratinocytes, and
  - generation of fibro-vascular tissue by the biopolymer scaffold.

Restoration of Skin Color

Clinical
hypopigmentation



Engineered skin
no melanocytes added

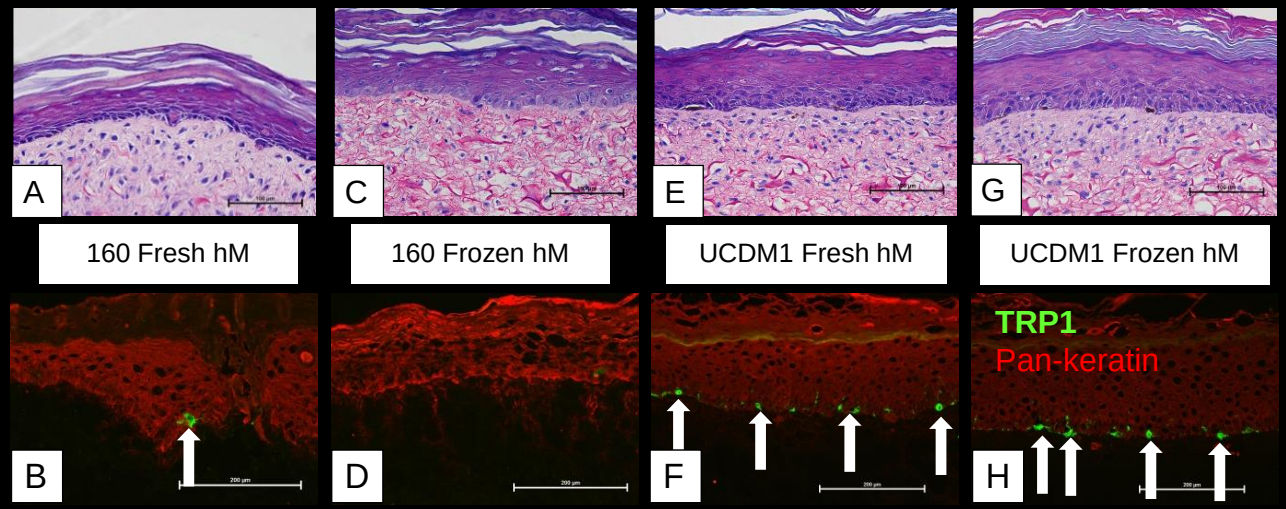


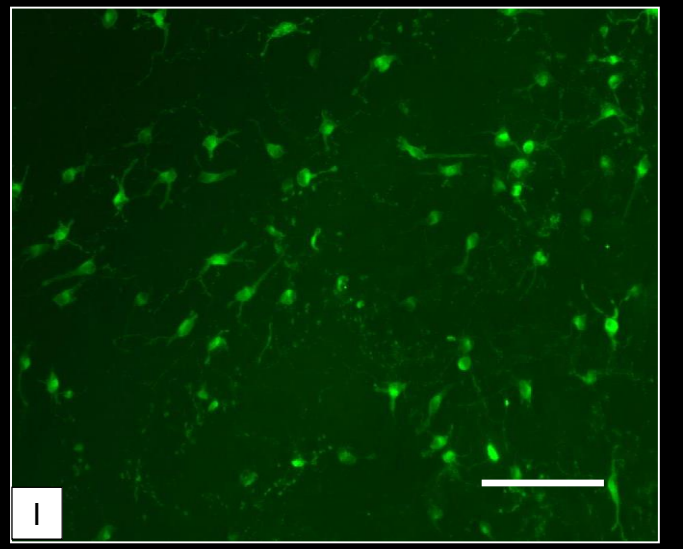
Engineered skin
with melanocytes added

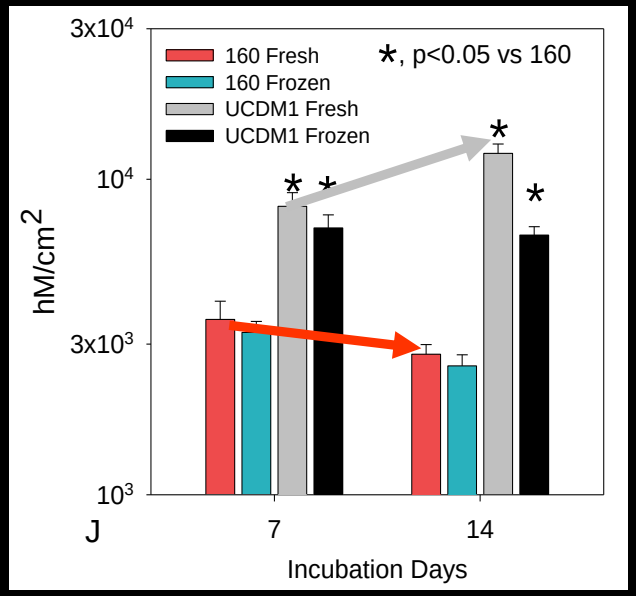
Swope et al, Wound Rep Regen, 2006

Melanocyte Density *In Vitro* (7-14 days)

TRP-1 staining *en face* image analysis



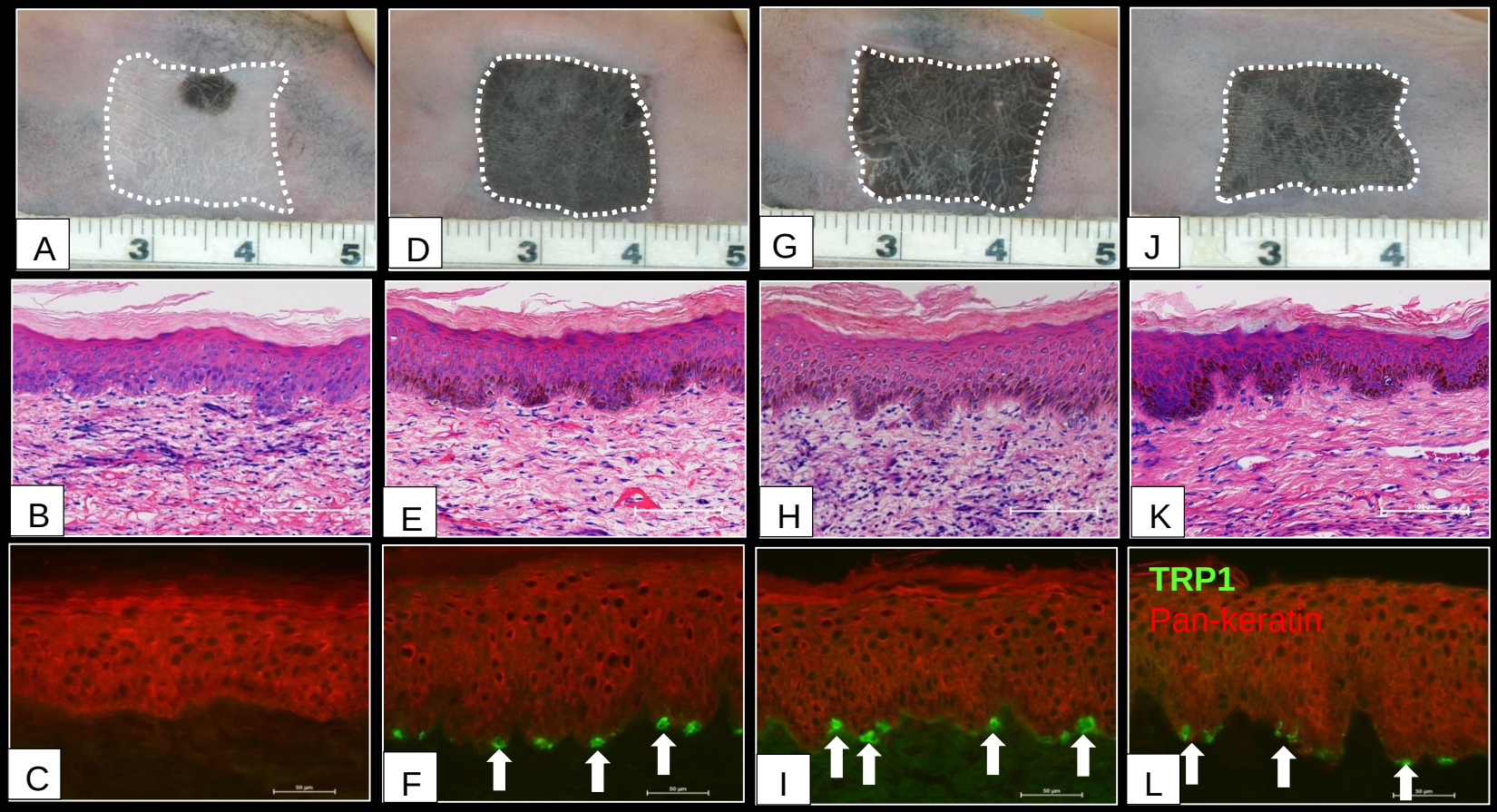




Boyce et al, Pigment Cell & Melanoma Res; epub, 22 Jun 2017

hM Retention in ESS-P *In Vivo* (12 wks)

Media: UCMC160 vs UCDM1



Closed Wound

Micro Anatomy

TRP1 for hM

No added hM

Fresh hM UCMC160

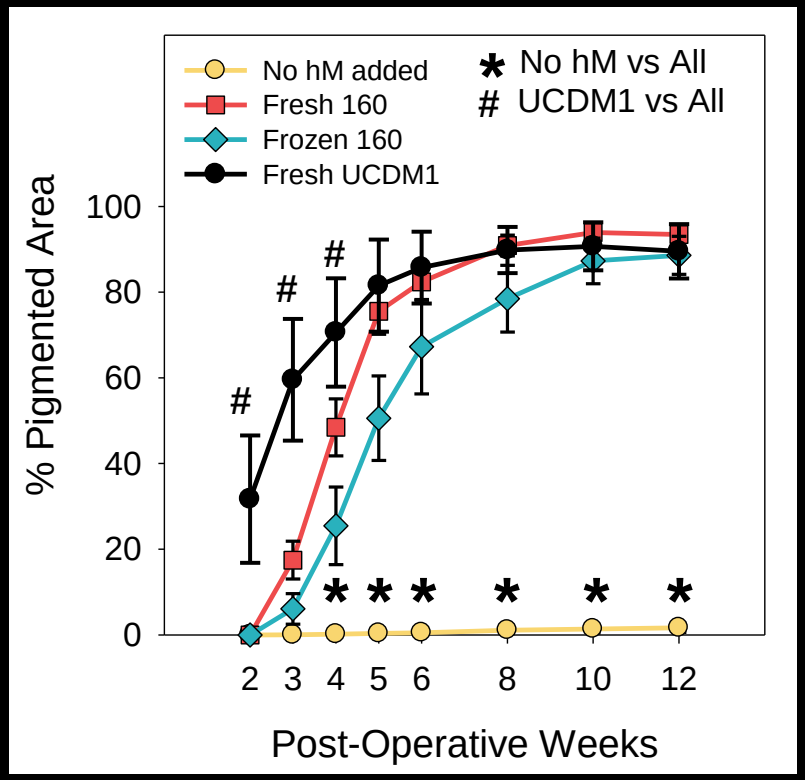
Frozen hM UCMC160

Fresh hM UCDM1

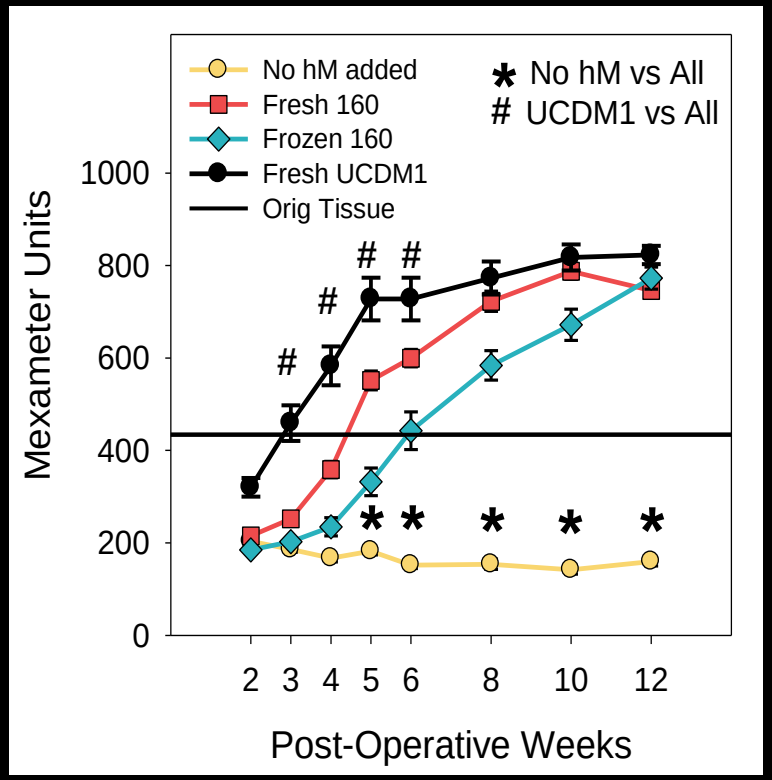
Grafting of ESS-P to Immunodeficient Mice

% Pigmented Area vs time; Mexameter vs time

Pigmented Area



Pigment Density

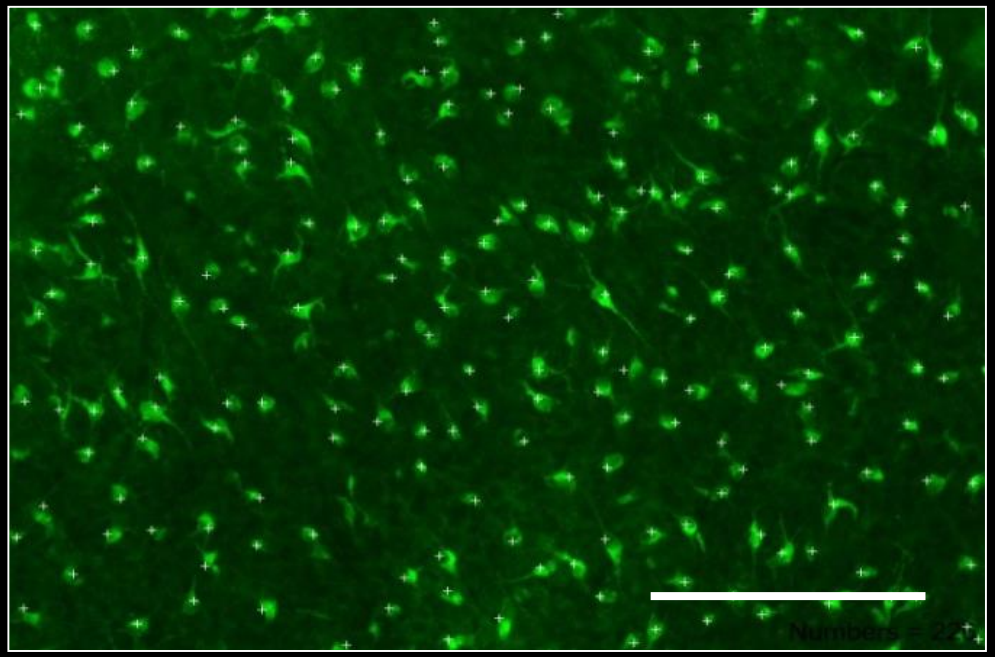


Boyce et al, Pigment Cell & Melanoma Res; epub, 22 Jun 2017

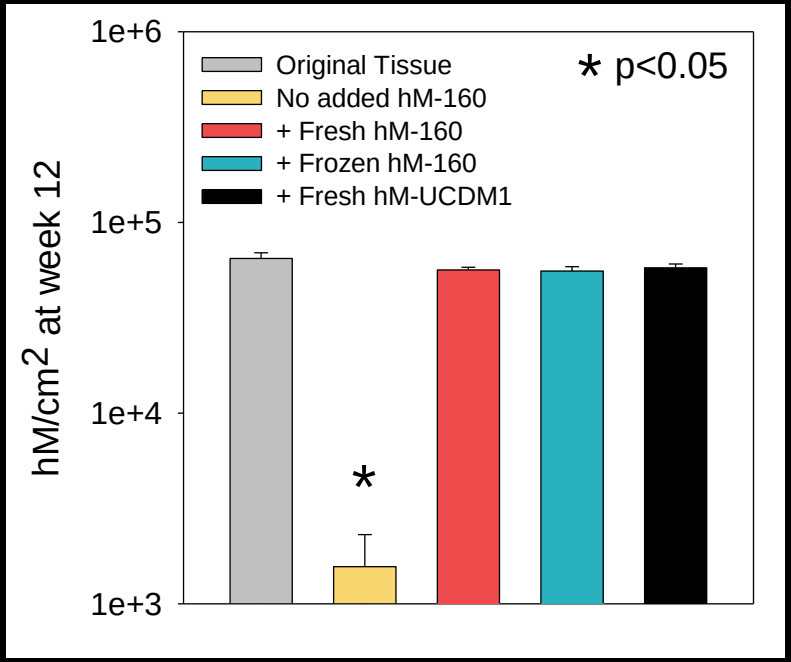
Melanocyte Density *In Vivo* (12 wks)

Mel-5 staining, *en face* image analysis

hM in Epidermis



hM Density



Boyce et al, Pigment Cell & Melanoma Res; epub, 22 Jun 2017

Summary

- Autologous ESS may offer an alternative therapy for FT burns >50% TBSA
- Natural pigment may be restored in ESS by transplantation of autologous melanocytes
- Optimized media promote increased efficiency of melanocyte transplantation

Engineered Skin Collaborators

Clinical

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Glenn Warden, MD
Richard Kagan, MD
Peggy Simpson, RN
Residents, post-docs and staff

Shriners Hospitals for Children™

UNIVERSITY OF Cincinnati

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- Shriners Hospitals for Children
- National Institute of General Medical Sciences
- US Army / Dept of Defense
- State of Ohio Department of Development
- FDA Office of Orphan Product Development