Graduate School of Biomedical Engineering

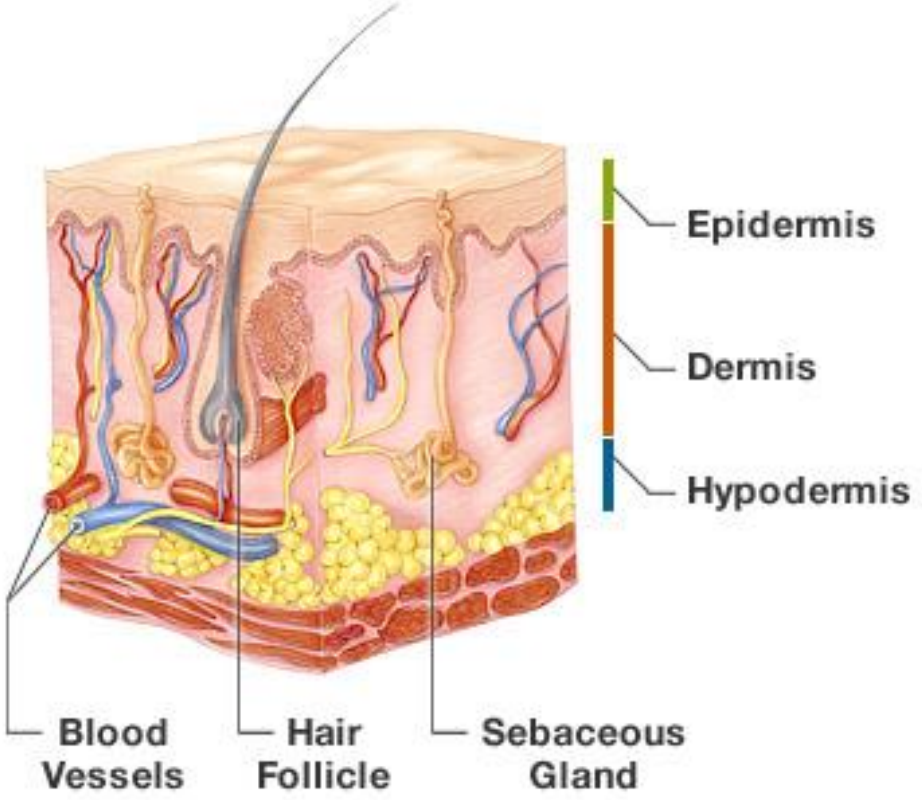
BIOM1010 – Week 2

Wound Healing

Dr Brooke Farrugia

Skin

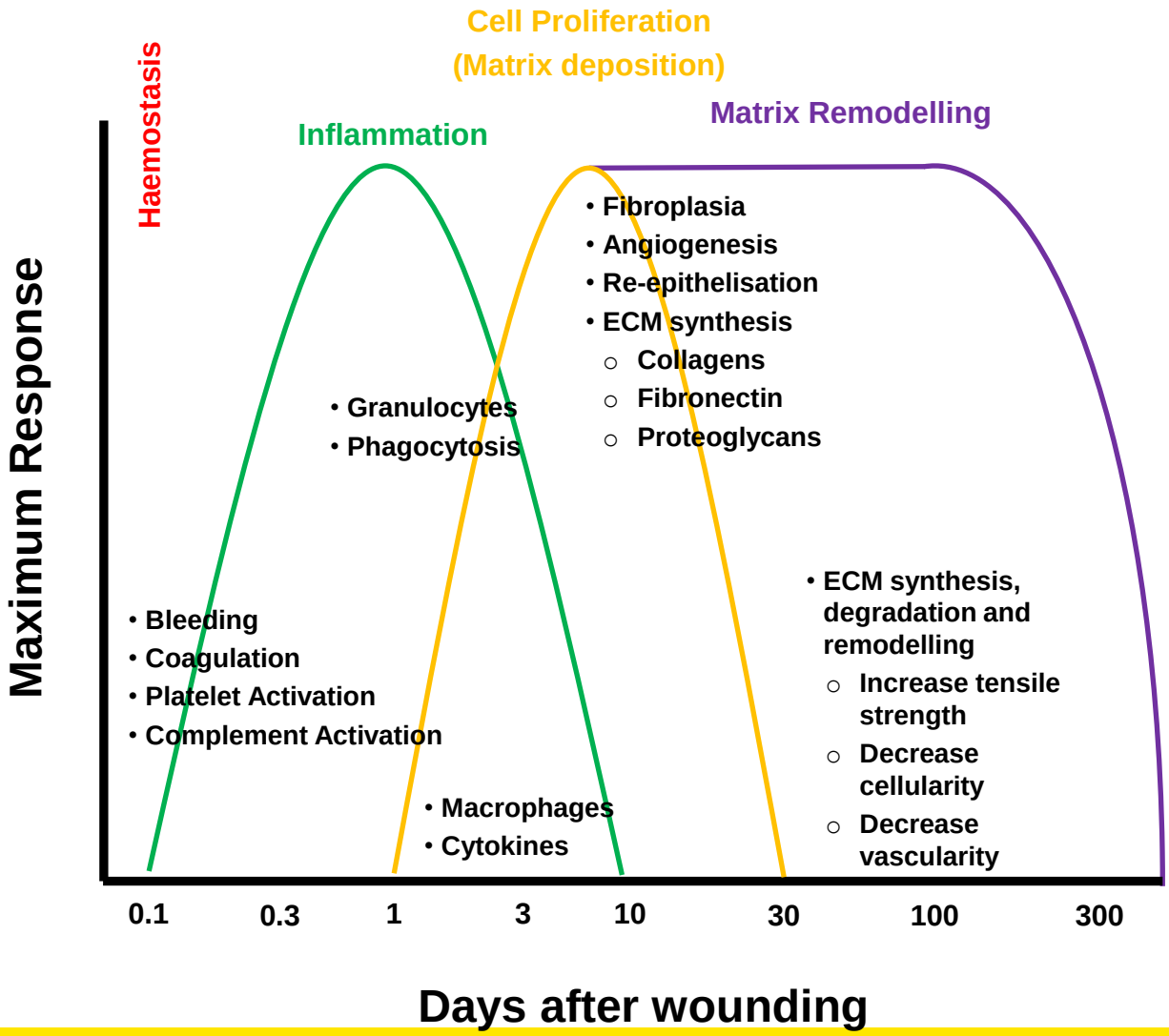
- Barrier between the internal and external environment
- How does it form this barrier?
- What happens when this barrier is compromised?



What is a Wound?

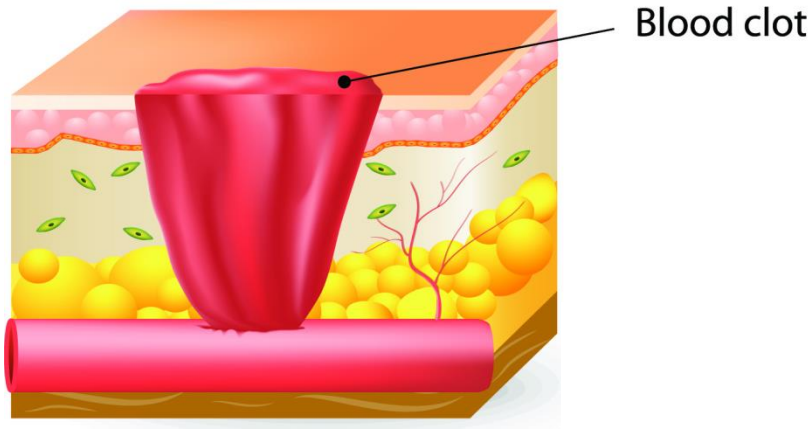
How does a wound heal?

- 4 main phases
  - Haemostasis
  - Inflammatory
  - Proliferative
  - Regeneration



Haemostasis

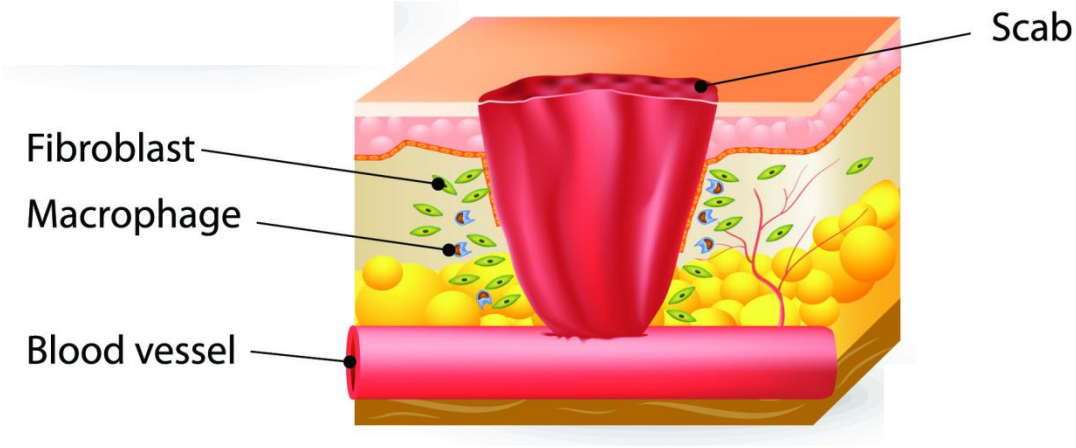
- Injury to microvasculature
- Constriction of blood vessels, activation of the coagulation cascade
- Formation of blood clot
  - Platelets
  - Proteins – fibrin, fibronectin, vitronectin, von Willibrand factor
- Platelets release growth factors
  - Initiation of wound healing cascade
  - Attract and activate fibroblasts, endothelial cells, macrophages



Inflammation

Early stage

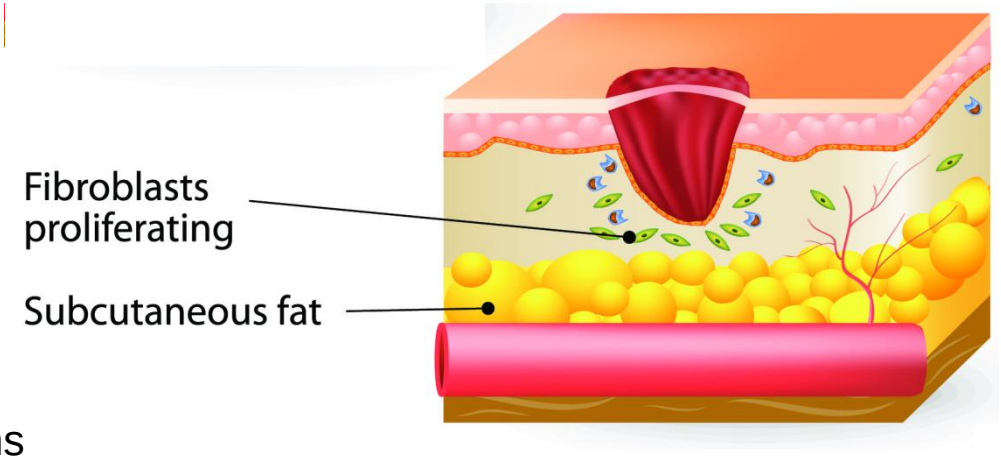
- Infiltration of inflammatory cells – granulocytes or polymorphonuclear leukocytes
- Remove bacteria and foreign materials – phagocytosis
- Prevent infection



Late stage

- Monocytes – macrophages
- Macrophages
  - Cytokine and growth factor release
  - Recruit fibroblasts, keratinocytes, endothelial cells
  - Proteases

Cell Proliferation

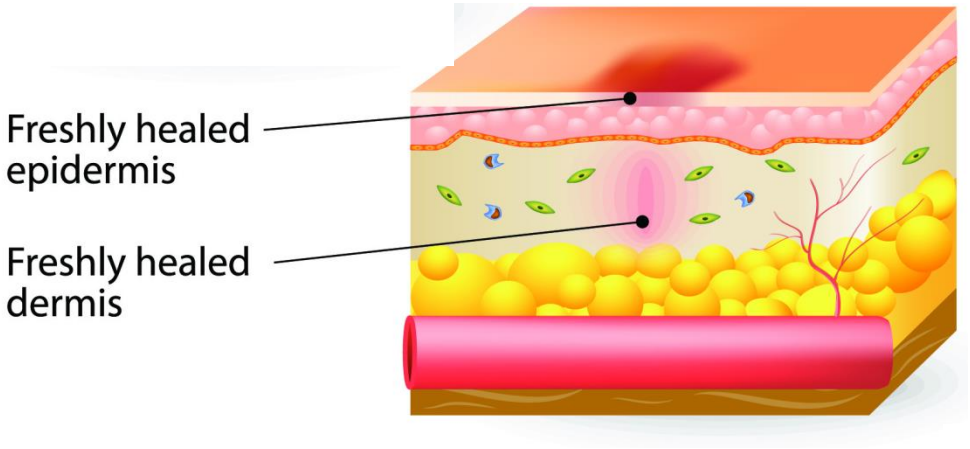


- Migration of fibroblasts
- Proliferation of fibroblasts
  - Production of ECM proteins

  – fibronectin, collagen, proteoglycans
- Collagen synthesis
  - Provide structure and strength
- Angiogenesis – formation of blood vessels from pre-existing ones
  - Macrophages release angiogenic factors
- Granulation tissue formation
  - Capillaries, proliferating fibroblasts, macrophages, collagen, glycoproteins
- Epithelisation
  - Migration of keratinocytes from the wound edges

Matrix Remodelling

- Occurs in conjunction with granulation tissue formation
- Occurs over prolonged time periods
- Breakdown and remodelling of ECM proteins, including collagen
- As time passes this process slows down



Abnormal Wound healing

Hypertrophic Scar

- Hypertrophic – increased volume
- Confined to border of original wound
- Regress over time
- Collagen

Keloid Scar

- Extend beyond border of original wound
- Will not regress overtime, can continue to grow
- Thick collagen

Chronic Wound

‘Normal’ process of wound healing has been impaired

Most commonly ‘stuck’ in the inflammatory or proliferative phase

Majority can be classed into three different categories

- Venous ulcers
- Diabetic ulcer
- Pressure ulcers

Venus Ulcers

Result from non-functioning venous valves – usually of the legs

Chronic wounds occur in 70-90% of venous ulcers

Venous valves no longer function properly

- Build up of inflammatory cells
- Insufficient removal of ‘waste’

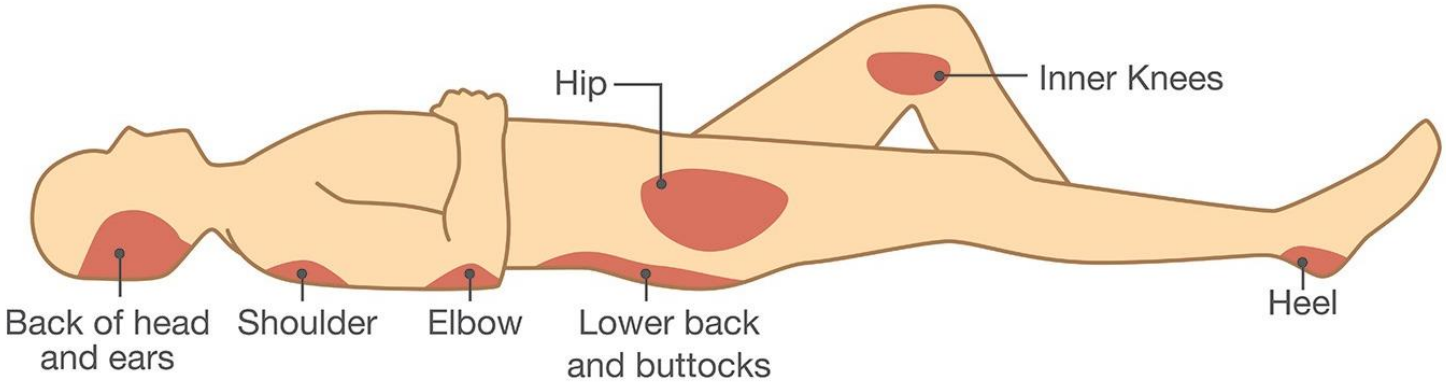
Diabetic Ulcer

Due to three main pathologies that are associated with diabetes

- Peripheral neuropathy
- Peripheral arterial disease
- Infection

Pressure Ulcers

Commonly caused by restricted blood flow to soft tissue



Treatment of Chronic Wounds

Problems

- Heterogeneous
- Underlying conditions
- Age – elderly

Common courses of treatment

- Debridement
- Dressings
- Compression / pressure bandages, negative pressure therapy
- Skin grafts

As biomedical engineers what new technologies have been or can be developed for wound healing?

Wound Dressings

Dressing type	Advantages	Disadvantages
Films (elastic sheets of polyurethane)	•Adherent •Transparent •Forms a bacterial barrier but is gas permeable	•Fluid collection •Difficult to remove, which may disturb new keratinocytes
Foams (bilaminate sheets containing polyurethane and often silicone)	•Absorbent •Moist healing environment •Conforms to body contours	•May require secondary dressing to place •Can adhere to wound if exudate dries
Hydrogels (96% water, cross-linked hydrophilic polymer)	•Comfortable •Absorbent •Promotes autolytic debridement	•Nonadherent •Maceration of skin around wound
Hydrocolloids (carboxymethylcellulose in adhesive base)	•Improved healing •Easy to use •Waterproof •Promote granulation tissue	•Unpleasant odor •Yellow brown, gel-like fluid drainage •May overstimulate granulation •Difficulty to use in cavities
Alginates (natural polysaccharides from kelp and algae)	•Absorbent •Useful in sinuses •Hemostatic properties	•Not useful for dry wounds •May require frequent dressing change if lots of exudate

Wound Dressings of the Future

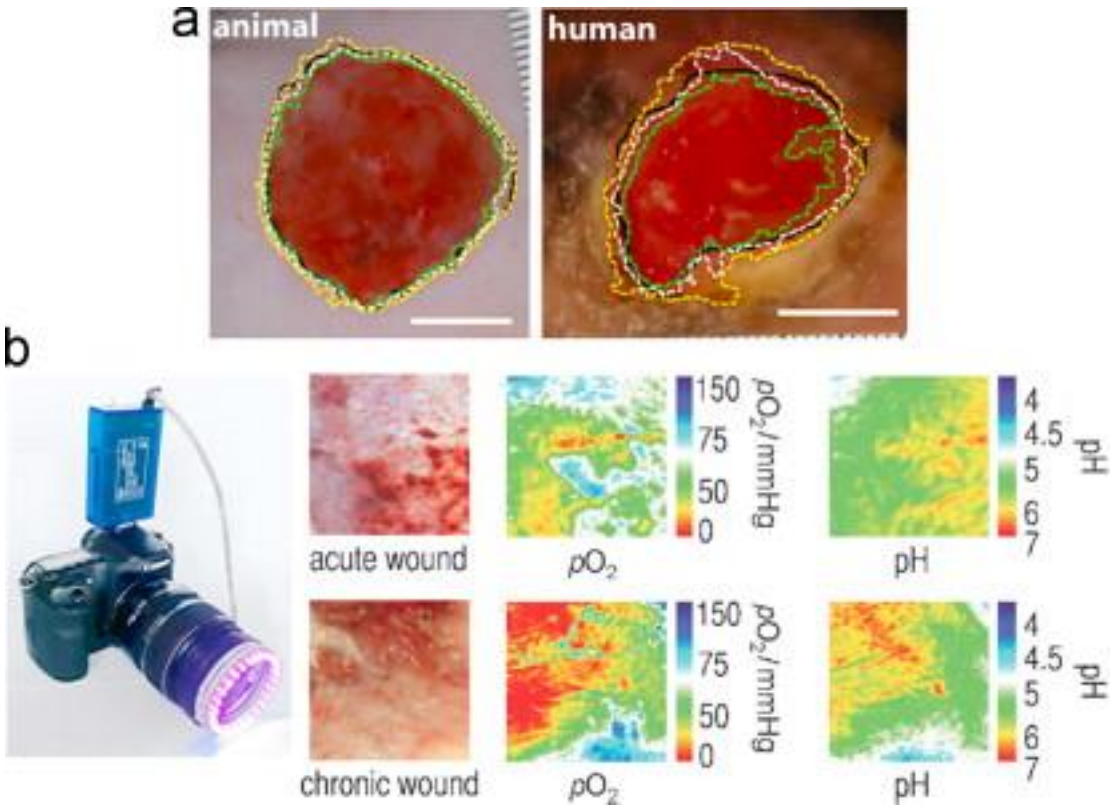
Dargaville et al. **Biosensors and Bioelectronics** Volume 41, 15 March 2013, Pages 30-42

Wound Dressings of the Future

List of potential markers for wound healing

- Bacterial load/specific microbial species/biofilms
- Cytokine release in response to specific microbial antigens
- Enzymes and their substrates—e.g. matrix metalloproteinases and extracellular matrix components
- Growth factors and hormones—e.g. platelet-derived growth factor (PDGF),thyroid hormones
- Inflammatory mediators—e.g. cytokines and interleukins to monitor healing status and guide use of anti-inflammatory treatments
- Nitric oxide
- Nutritional factors—e.g. zinc, glutamine ,vitamins
- pH of wound fluid
- Reactive oxygen species
- Temperature

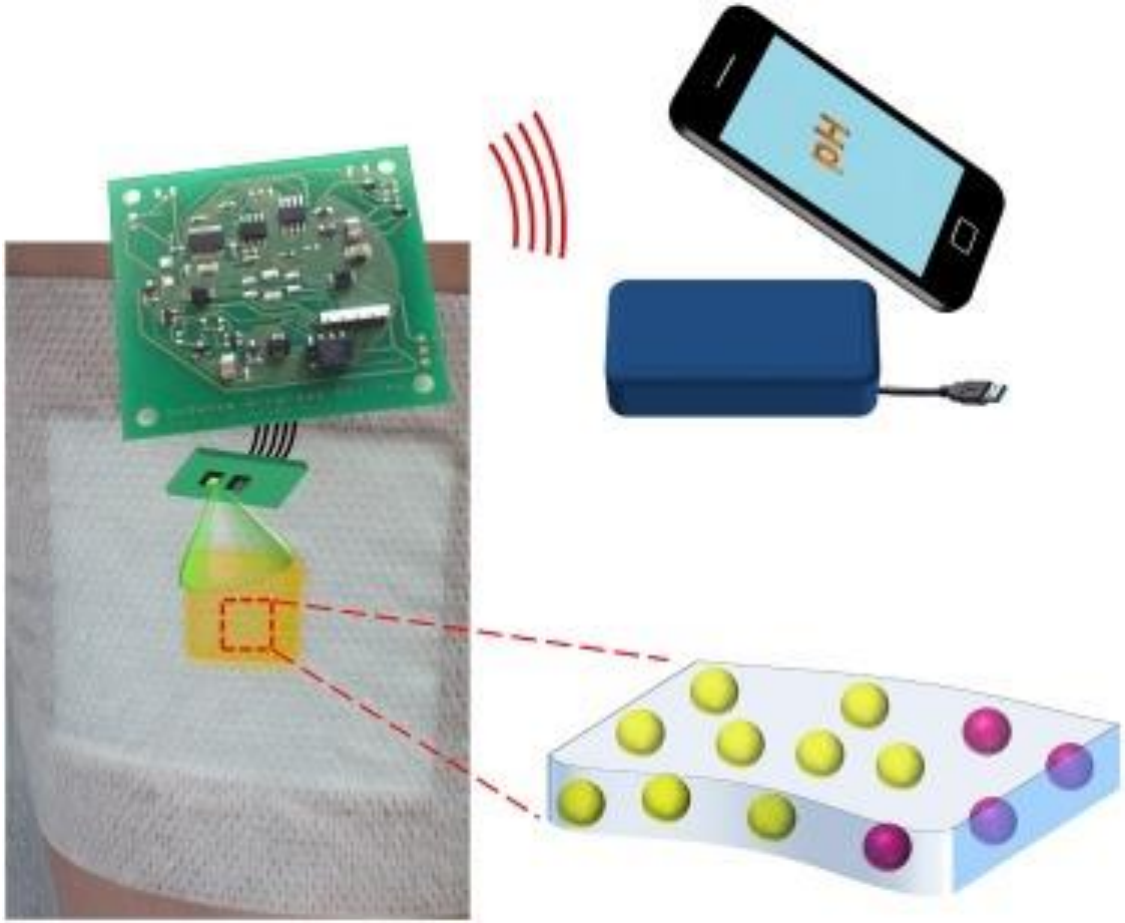
Wound Dressings of the Future



(a) Animal and human wounds traced manually (black line) and using image analysis (green, white, yellow lines). Scale bar=1 cm and (b) Left: a digital camera fitted with a 405 nm-LED ring light and an emission filter for photographing wounds covered in a sensor film. Right: visible light pictures, pO_2 and pH maps comparing acute and chronic wounds.

Dargaville et al. **Biosensors and Bioelectronics** Volume 41, 15 March 2013, Pages 30-42

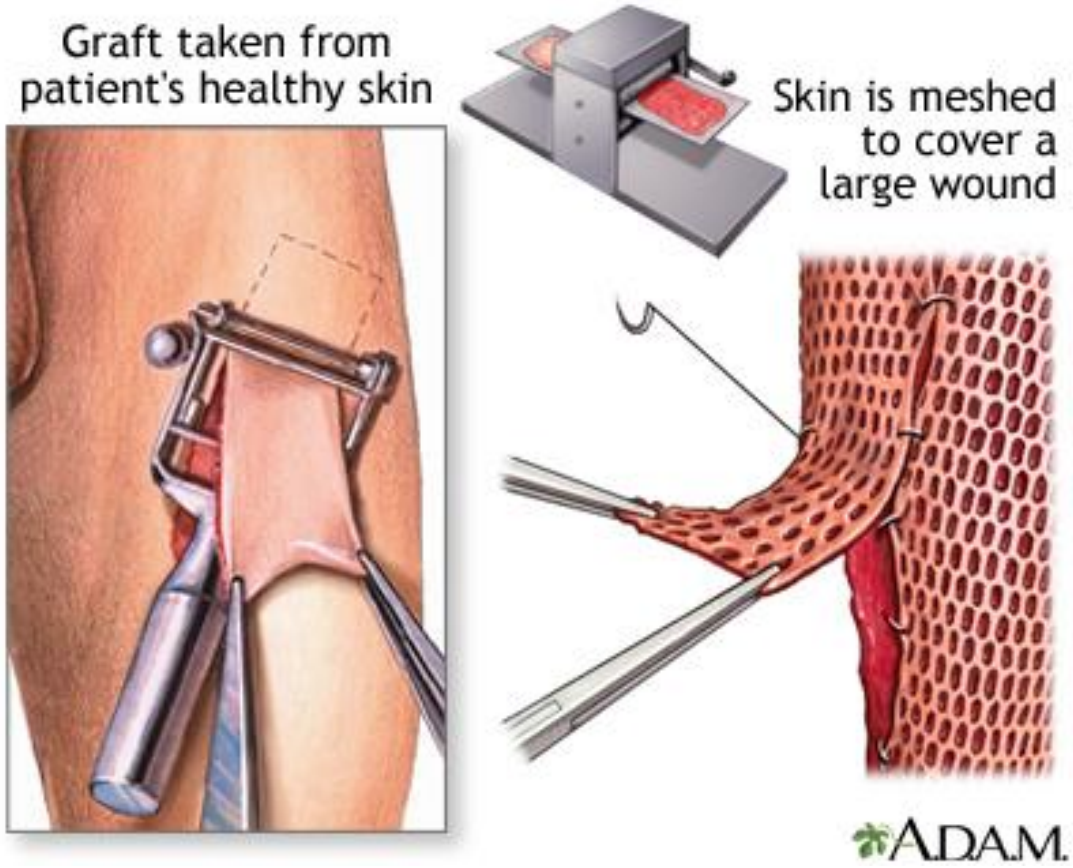
Wound Dressings of the Future



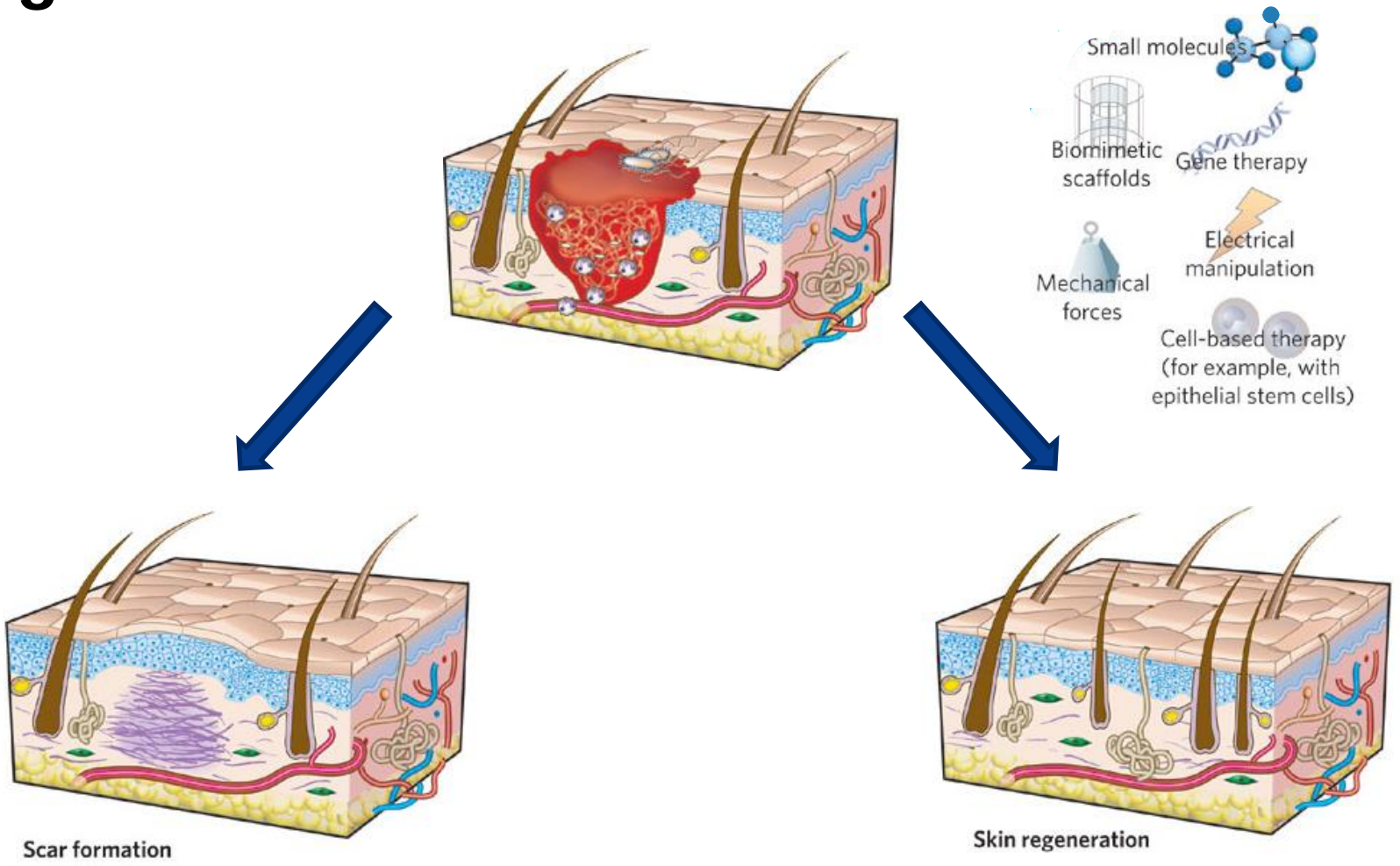
Schematic showing operation of the wireless smart bandage.
The pH-induced colour change of cellulose particles is measured and sent by contactless readout to a remote unit.

Kassal et al. Sensors and Actuators B: Chemical Volume 246, July 2017, Pages 455-460

Skin Grafts


Graft taken from patient's healthy skin
Skin is meshed to cover a large wound
A.D.A.M.

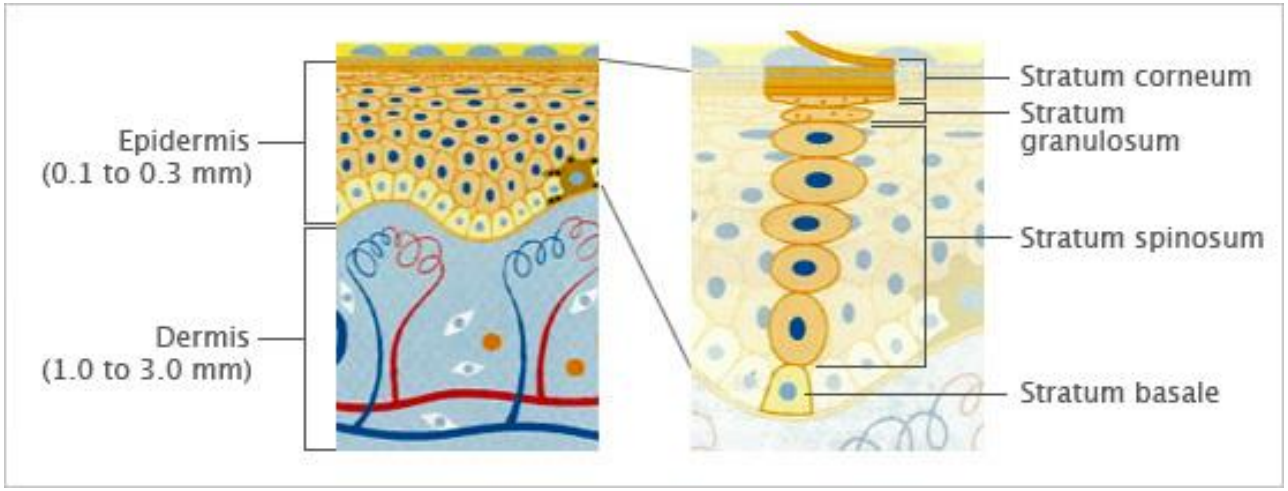
Regenerative Medicine



Gurtner et al. Nature 453, 314-321(15 May 2008) doi:10.1038/nature07039

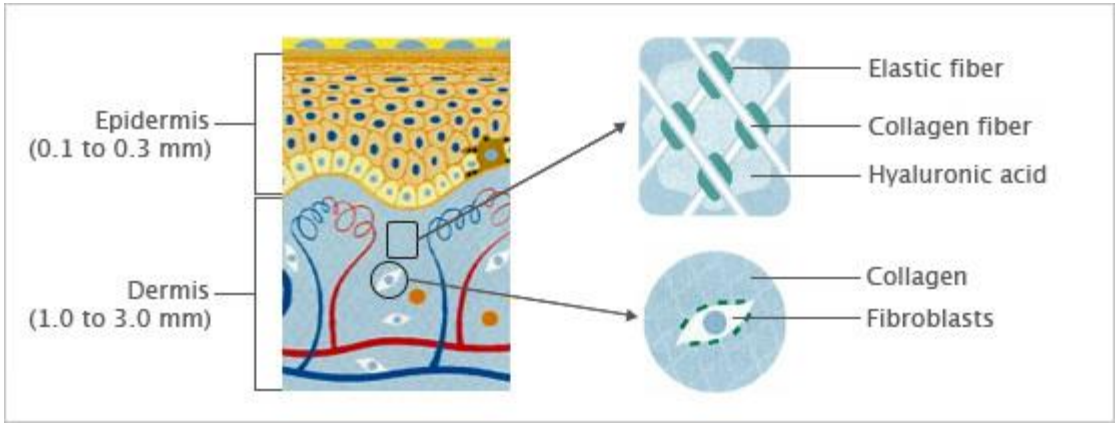
Epidermis

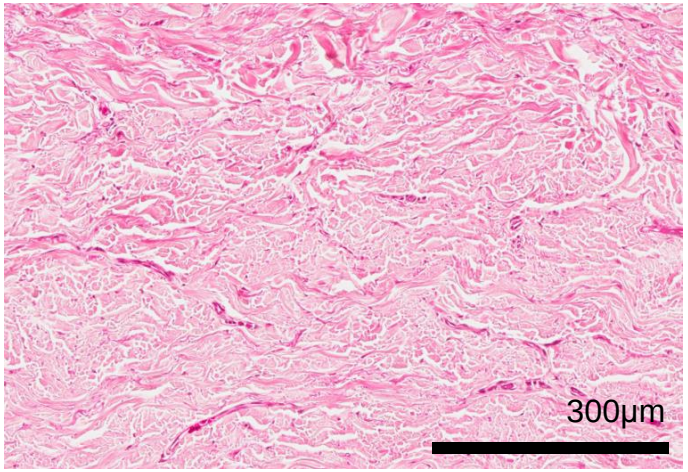
- Multiple layers of keratinocytes
- Continual turnover of cells (40–56 days)
- No ECM



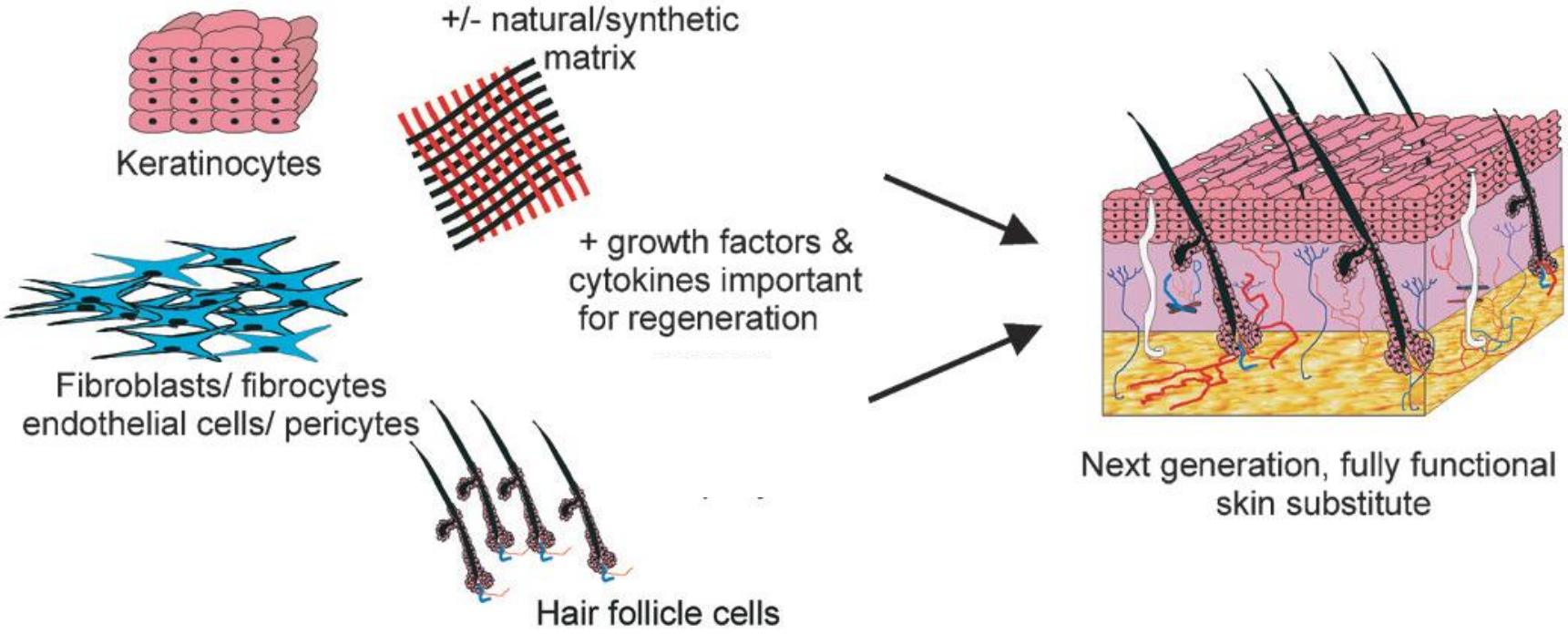
Dermis

- Major cell type - fibroblasts
- Collagen, elastic fibres, and extrafibrillar matrix





What are we striving to achieve?



Commercially available epidermal constructs for clinical use

Brand name/manufacturer	Graft type		Cell source	Biomaterial	Life-span
	Cell-based	Cell-seeded scaffold (TE)			
Cell Spray Clinical Cell Culture (C3), Perth, Australia	x		Autologous keratinocytes	–	Permanent
Epicel Genzyme Biosurgery, Cambridge, MA, USA	x, cell sheet		Autologous keratinocytes	–	Permanent
EpiDex Modex Therapeutiques, Lausanne, Switzerland	x, cell sheet		Autologous keratinocytes	–	Permanent
EPIBASE Laboratoires Genevrier, Sophia-Antipolis, Nice, France	x, cell sheet		Autologous keratinocytes	–	Permanent
MySkin Cell Tran Ltd, Sheffield, UK		x	Autologous keratinocytes	Synthetic, silicone support layer with a specially formulated surface coating	Permanent
Laserskin or VivodermFidia Advanced Biopolymers, Padua, Italy		x	Autologous keratinocytes	Recombinant, (HAM)	Permanent
Bioseed-S BioTissue Technologies GmbH, Freiburg, Germany		x	Autologous keratinocytes	Allogeneic, fibrin sealant	Permanent

HAM: hyaluronic acid membrane (microperforated).

doi:10.1016/j.addr.2011.01.005

Cell Spray

[Dr Fiona Wood on spray on skin]

My Skin

Plasma polymerization can be used to produce surfaces that are either nonadherent or keratinocyte adherent. (a) Plasma rig. The sample to be coated is placed within the vacuum chamber and a high radiofrequency energy is used to break the material into fragments under vacuum. This introduces the material to be coated as a gas, which becomes attached to the surface. (b) This technique is used to produce a background surface coated with octadiene, which is nonadhesive for the majority of cells. Onto this is placed a template of the letters 'TONY' coated with acrylic acid. Skin cells from Tony Ryan of the University of Sheffield, UK, were expanded in the laboratory and placed on this surface. They adhered to the acrylic acid-coated surface but failed to adhere to the octadiene-coated surface. (c) The clean room conditions under which patients' skin cells are expanded from a small skin biopsy. (d) An acrylic acid plasma polymerized silicone carrier (Myskin™). Patients' cells are placed on the acrylic acid-coated surface and kept in media for transport to the patient. The clinician or dressings nurse picks up the Myskin carrier and places it with the cells facing downwards onto the wound bed.

Biomaterials for tissue engineering of skin

doi:10.1016/S1369-7021(08)70087-7

Commercially available dermal constructs for clinical use

Brand name/manufacturer	Graft type		Cell source	Biomaterial	Life-span
	Cell-free	Cell-seeded scaffold			
AlloDermLifeCell Corporation, USA	x		–	Allogeneic human acellular lyophilized dermis	Permanent
Karoderm Karocell Tissue Engineering AB, Sweden	x		–	Allogeneic human acellular dermis	Permanent
Matriderm Dr Suwelack Skin and HealthCare Germany	x		–	Xenogeneic bovine non-cross-linked lyophilized dermis, coated with α-elastin hydrolysate	Permanent
Integra Dermal Regeneration Template Integra NeuroSciences, USA	x		–	Xenogeneic and synthetic: polysiloxane, bovine cross-linked reconstituted	Semi-permanent
Terudermis Olympus Terumo Biomaterial Corp, Japan	x		–	Xenogeneic and synthetic: silicone, bovine lyophilized cross-linked collagen sponge made of heat-denatured collagen	Semi-permanent
Pelnac Standard/Pelnac Fortified Gunze Ltd, Medical Materials Center, Japan	x		–	Xenogeneic and synthetic: silicone/silicone fortified with silicone gauze TREX, atelocollagen derived from pig tendon	Semi-permanent
Biobrane/Biobrane-LUDL Laboratories, Inc, USA	x		–	Xenogeneic and synthetic: silicone film, nylon fabric, porcine collagen	Temporary
Hyalomatrix PAFidia Advanced Biopolymers, Italy	x		–	Allogeneic and synthetic: HYAFF layered on silicone membrane	Semi-permanent
TransCyte (DermagraftTC) Advanced BioHealing, IncUSA		x	Neonatal allogeneic fibroblasts	Xenogeneic and synthetic: silicone film, nylon mesh, porcine dermal collagen	Temporary
Dermagraft Advanced BioHealing, Inc USA		x	Neonatal allogeneic fibroblasts	Allogeneic and synthetic: PGA/PLA, ECM	Temporary
Hyalograft 3D Fidia Advanced Biopolymers, Italy		x	Autologous fibroblasts	Allogeneic: HAM	Permanent

PGA: polyglycolic acid (Dexon).
PLA: polylactic acid (Vicryl).
ECM: extracellular matrix.
HAM: hyaluronic acid membrane (microperforated).

AlloDerm LifeCell

Allogeneic human acellular lyophilized dermis

Commercially available epidermal/dermal constructs for clinical use

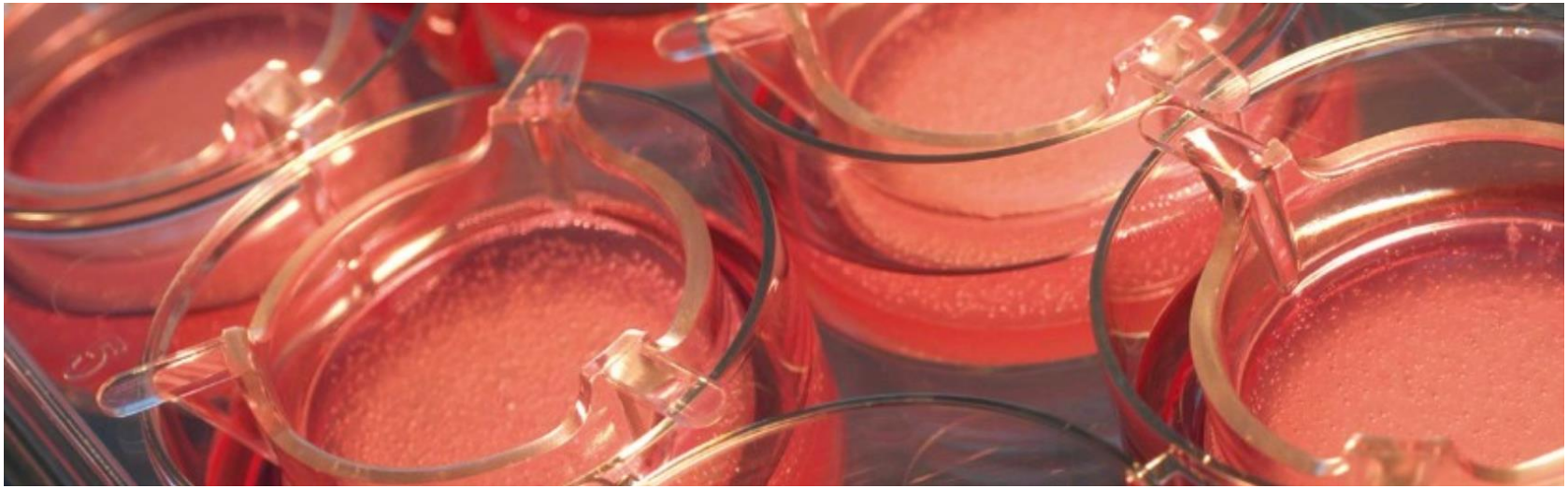
Brand name/manufacturer	Graft type			Cell source	Biomaterial	Life-span
	Cell-free	Cell-based	Cell-seeded scaffold (TE)			
Apligraf Organogenesis Inc., Canton, USA			x	Allogeneic keratinocytes and fibroblasts	Bovine collagen	Temporary
OrCel Ortec International, Inc. USA			x	Allogeneic keratinocytes and fibroblasts	Bovine collagen sponge	Temporary
PolyActive HC Implants BV, Leiden, The Netherlands			x	Autologous keratinocytes and fibroblasts	Synthetic, … (PEO/PBT)	Temporary
TissueTech Autograft System (Laserskin and Hyalograft 3D) Fidia Advanced Biopolymers, Italy			x	Autologous keratinocytes and fibroblasts	Recombinant, HAM	Temporary

PEO: polyethylene oxide terephthalate.
PBT: polybutylene terephthalate.
HAM: hyaluronic acid membrane (microperforated).

Alipgraf

Alipgraf Manufacturing video

Alternative uses – Product Testing



In vitro skin model

- Cosmetics – L'Oreal
- The Tissue Factory
- Wound healing therapeutics
  - Animal model limitations

What have we learnt

What is a wound -

The 4 main phases of wound healing –

When does a chronic wound occur –

Three main types of chronic wounds -

Why are chronic wounds hard to treat –

Common treatments for chronic wounds –

In the future what could wound dressings monitor –

What is a disadvantage of skin grafts –

The main cell type in the (i) epidermis and (ii) dermis –

The main cell source for epidermal constructs –

An advantage of decellularised tissue –

A disadvantage of epidermal/constructs -