

# Direct Delivery

## A Gentle Polymer for RDEB Skin

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**UCD Charles Institute of  
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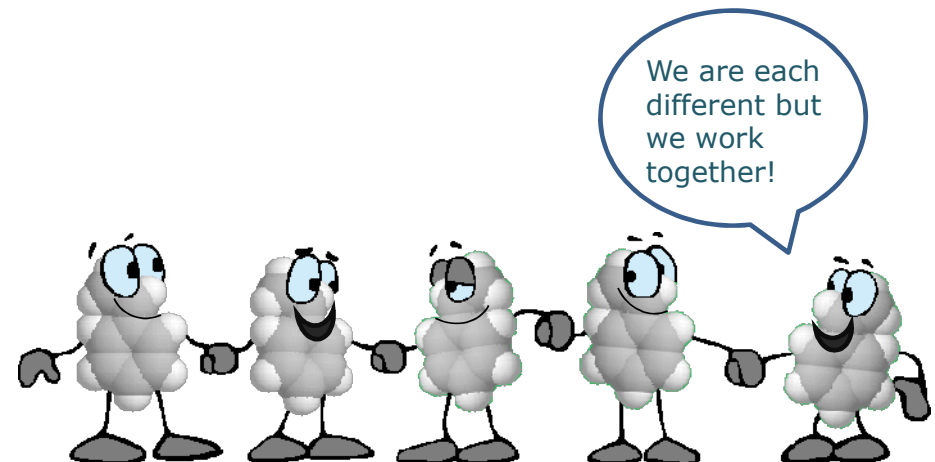
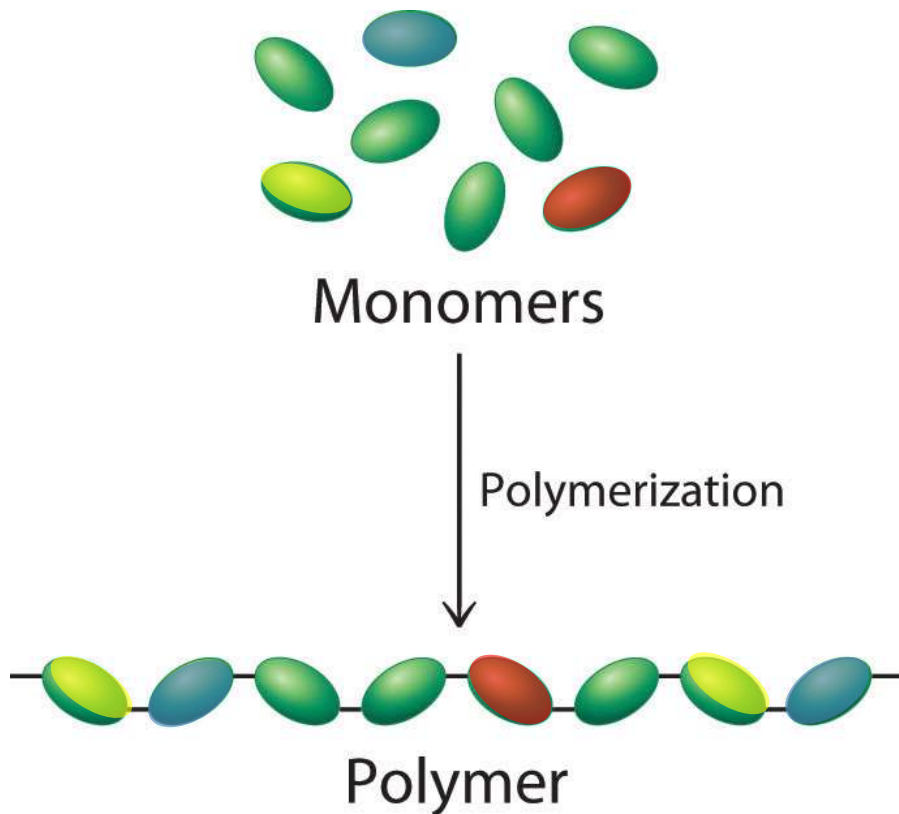
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# Polymers

Polymers are molecules made up from lots of small molecules called monomers.

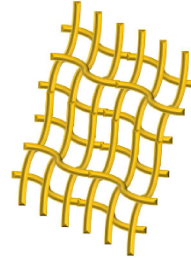
There is a huge range of possible monomers you can use and the polymers produced can have simple or very complex shapes.

Linear or Branched Polymers.



# Tuneable Conformation for Different Applications

Networked polymers



Hydrogel dressings

Surgical tissue adhesives

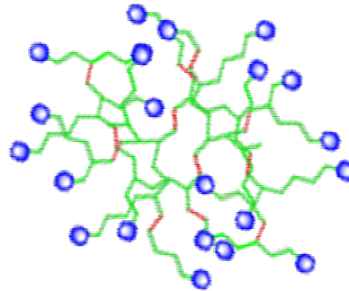
Cyclised Knot Polymers



Anti-microbial agents

RNA delivery vectors

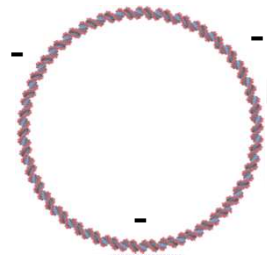
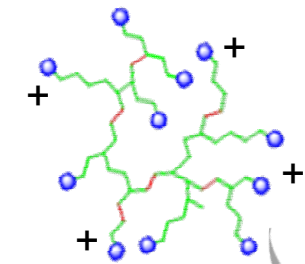
Highly Branched Polymers



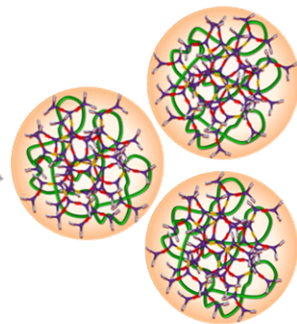
DNA delivery vectors

# Polymer Gene Therapy

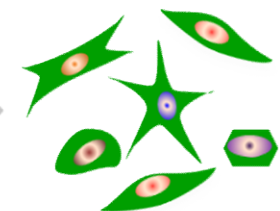
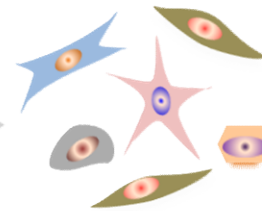
Highly Branched Polymer



Plasmid DNA



Polyplexes

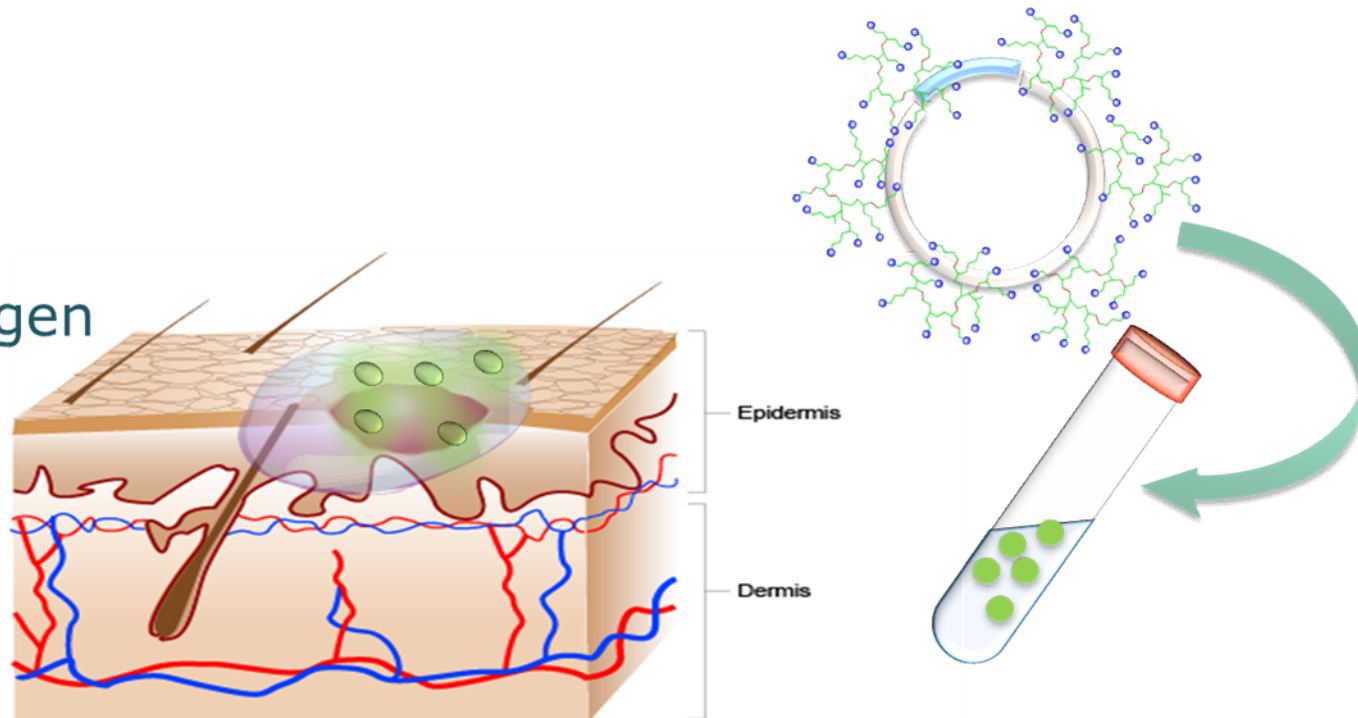




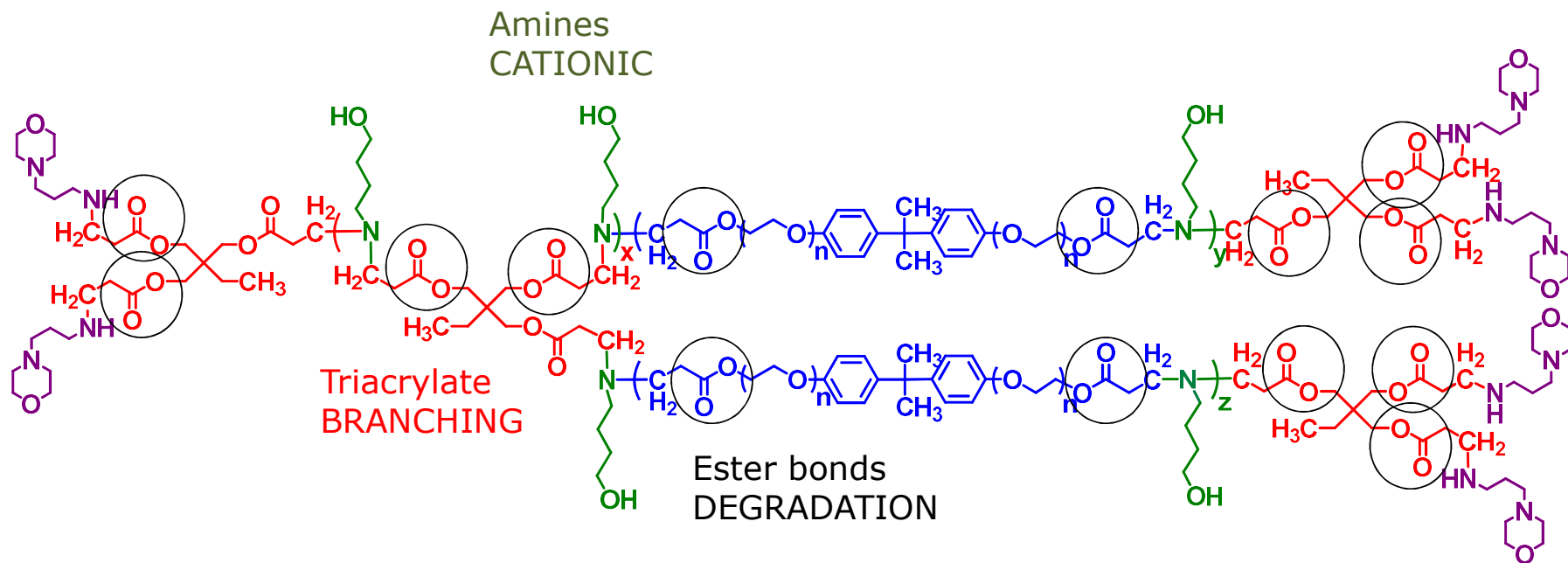
# Polymer Gene Therapy

## HPAE-COL7A1 Polyplexes

Type VII Collagen  
restoration



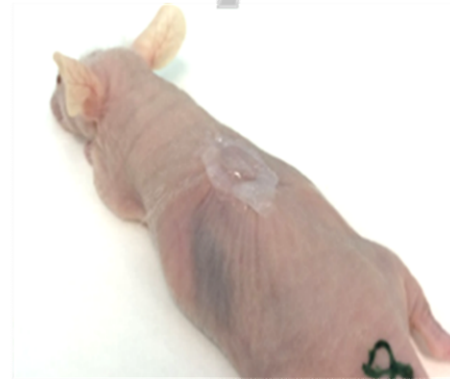
# Original Highly branched Poly ( $\beta$ -amino ester) - HPAE



# In Vivo Assessment of HPAE Original

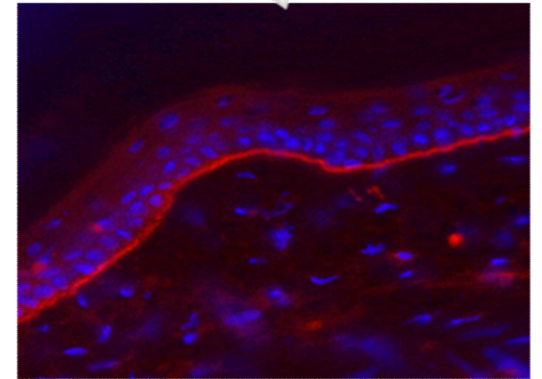
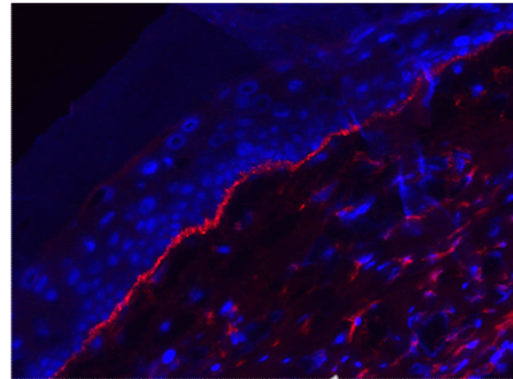
RDEB human: murine full thickness skin graft model

**HPAE+C7 Injection**



**Topical HPAE+C7**

**After 4 weeks**

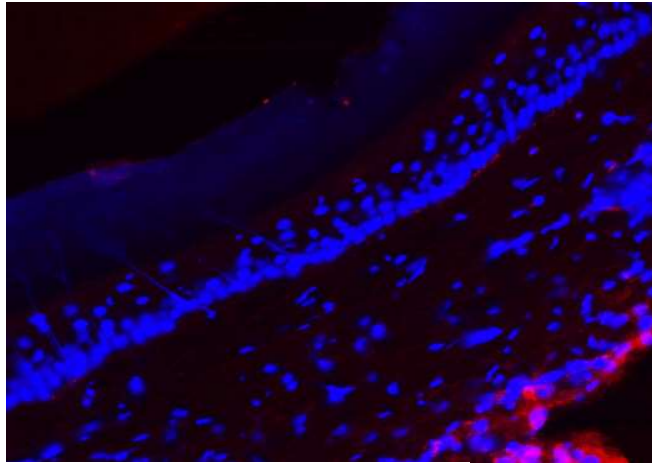


**After 10 weeks**

# Topical delivery of rtype VII Collagen

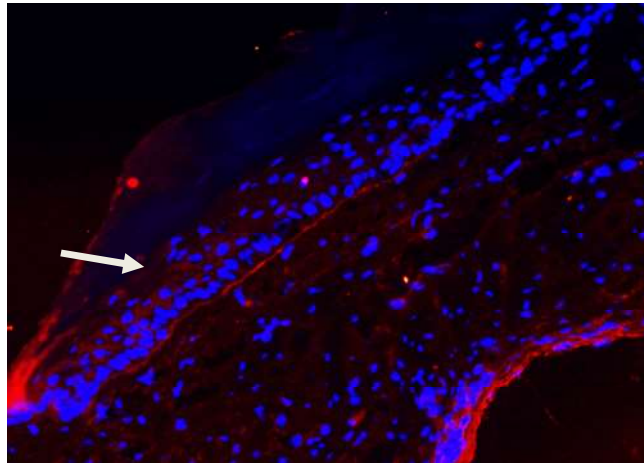
- ✓ Type VII Collagen Expression, after topical application

Control RDEB Skin No C7



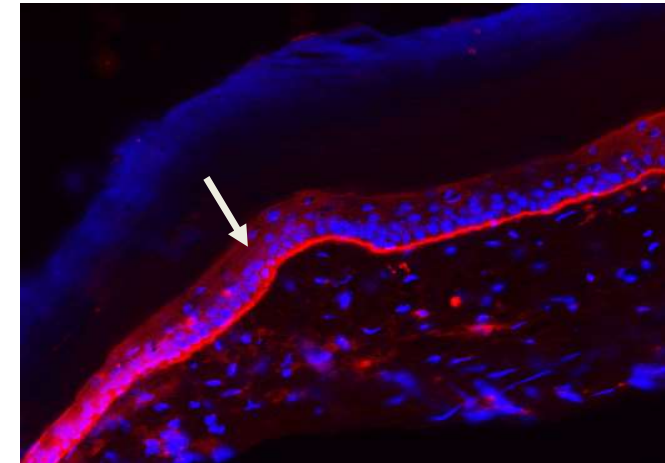
Images taken at 20x

1xHPAE-COL7A1 Topically



4 weeks

3xHPAE-COL7A1 Topically



10 weeks

- ✓ Of the 9 animals treated in total by intradermal injection and topical application, 7 were seen to be positive for recombinant for human type VII collagen
- ✓ With a clear advantage seen to repeated application

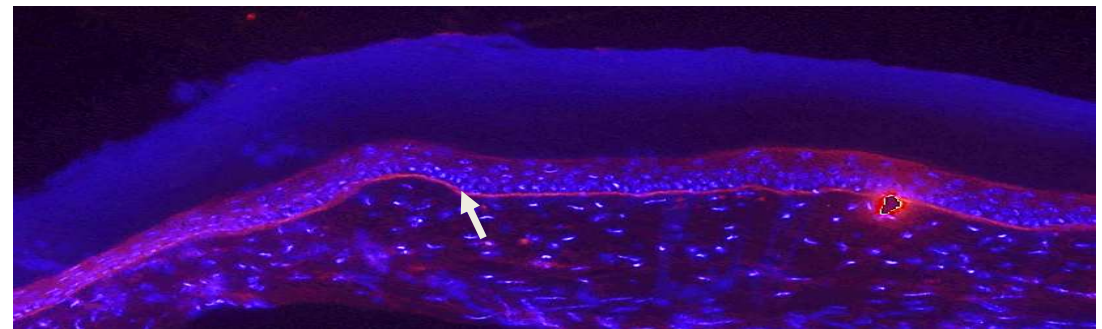
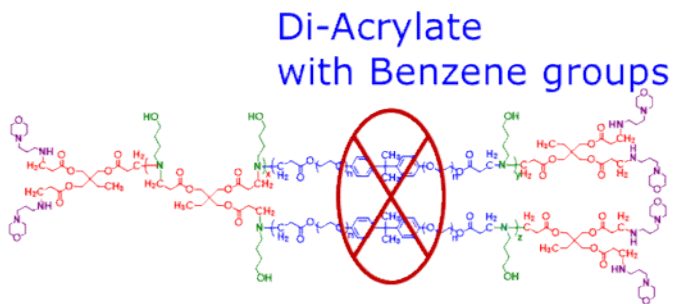


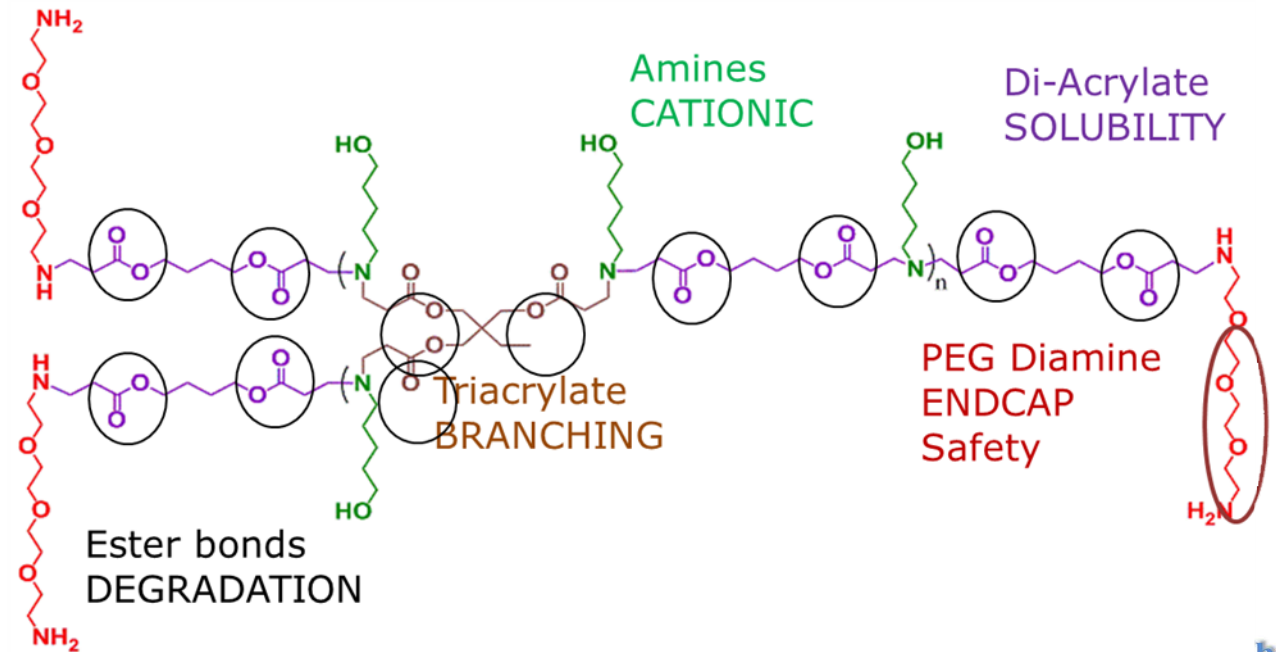
Image taken at 10x

# The New Generation of HPAE for EB

## Original Highly branched PAE

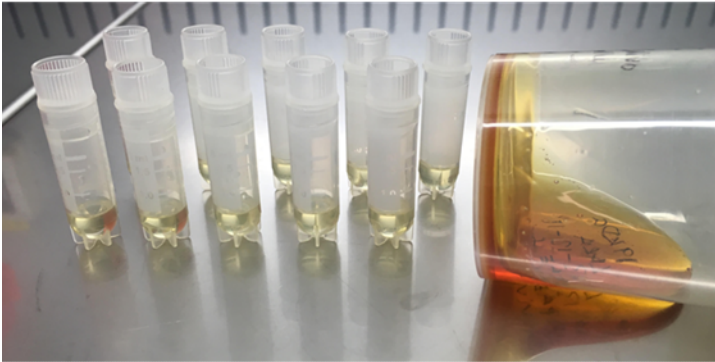


## New Generation Highly branched PAE-EB (HPAE-EB)





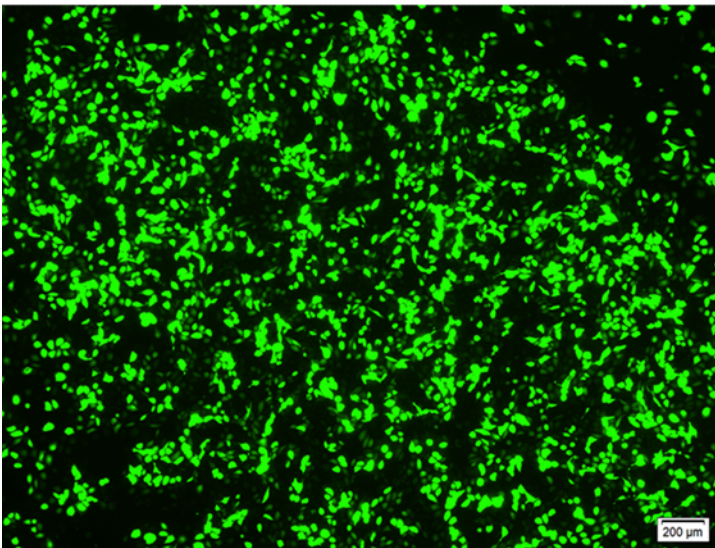
## New HPAE-EB



✓ Binds almost 100% of DNA at 30:1.

✓ Polyplexes are positively charged  
 $10-12\text{mV}\pm 4$ .

✓  $100\text{nm}\pm 11$  size range.



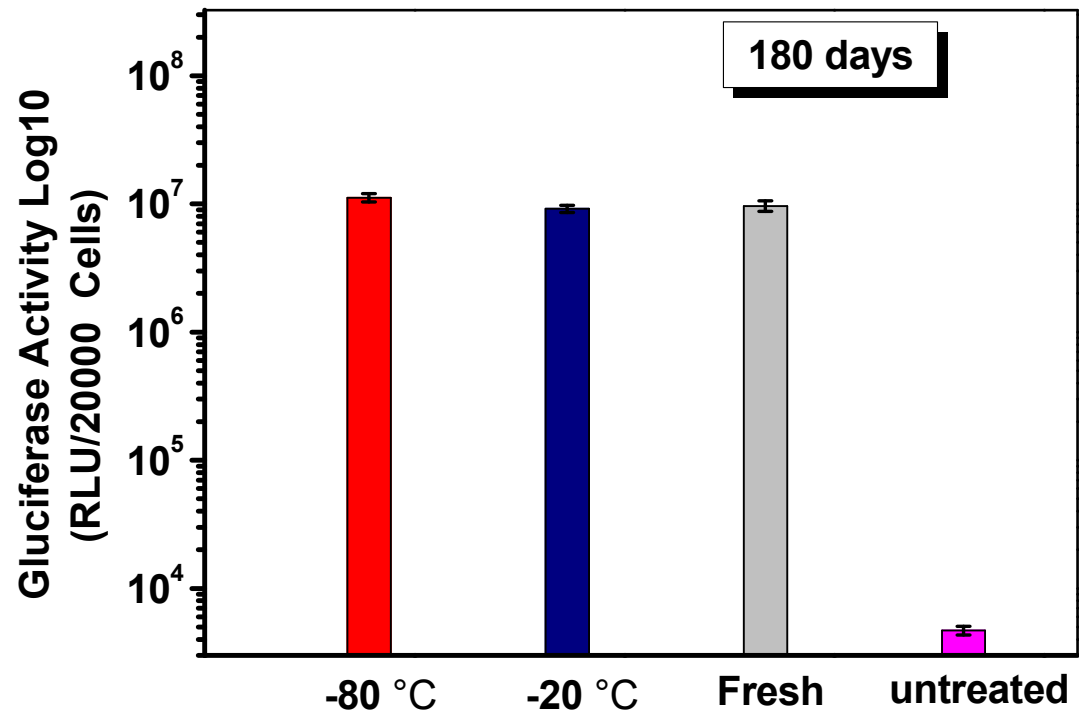
✓ Highly efficient at delivering DNA to  
TA4-RDEBKs, primary keratinocytes and  
primary fibroblasts to  $\sim 90\%$  GFP+.  
(Scale bar  $200\mu\text{m}$ )



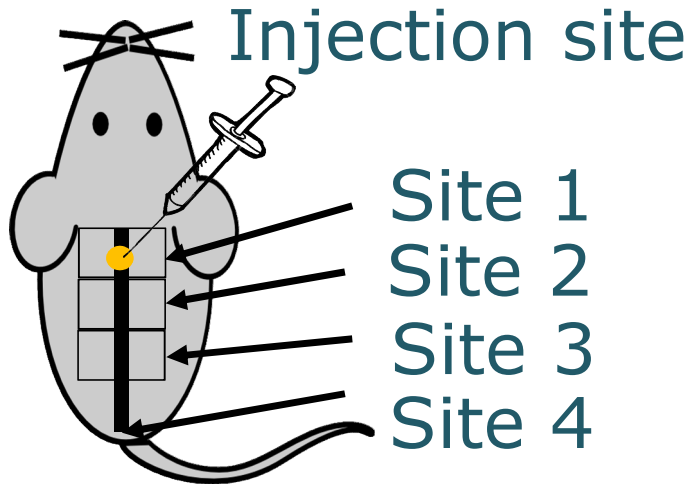
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## New HPAE-EB

- ✓ Polyplexes form in 15 mins.
- ✓ Polyplexes stable at -20°C for at least 6 months.
- ✓ Monomers are cheap, “One-Pot” synthesis, Total cost per gram of HPAE = €13.36.



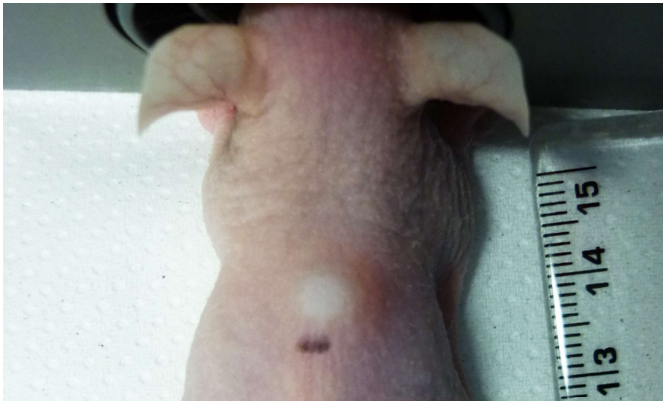
# Safety Assessment of HPAE-EB



Intradermal injections of PEI+COL7A1 and HPAEA-EB+C7 polyplexes in healthy SKH1 mice were carried out every 2nd day for 12 days.

Six injections in total per group,  
n=11, 8 Groups

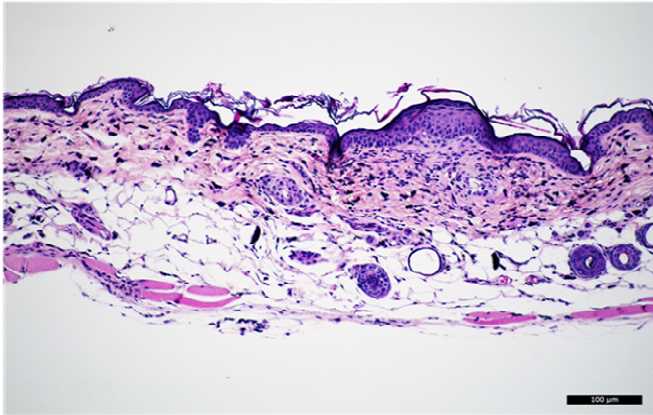
Tissues harvested:  
Skin x4, Serum, Liver and kidneys.



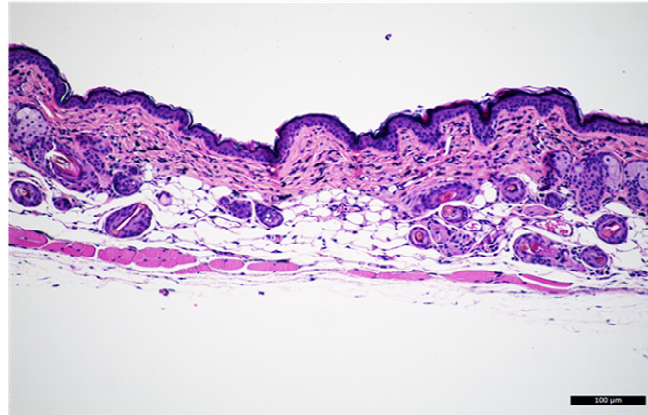


# Results of HPAE-EB Safety Study

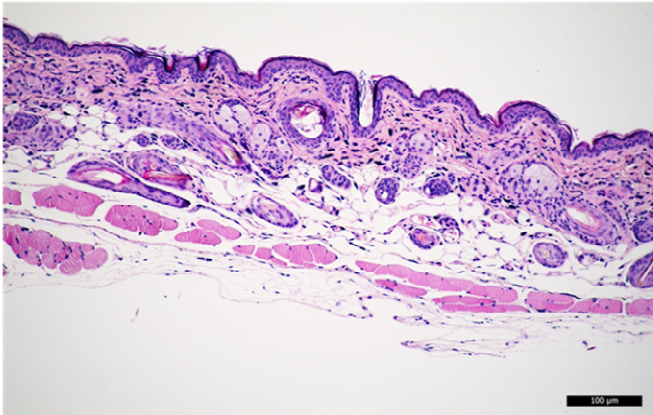
Control



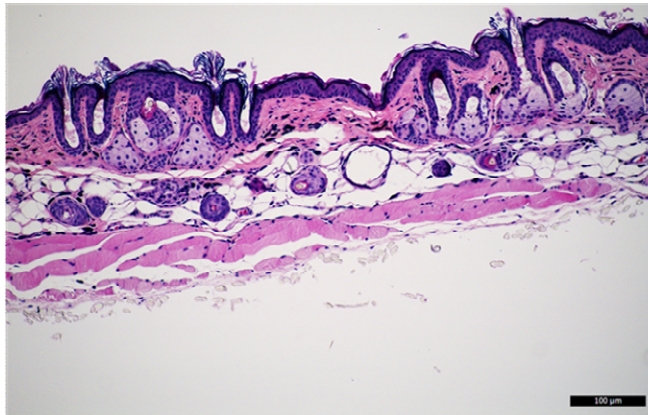
6xHPAE-EB+C7 Intradermal Inj. MD



6xHPAE-EB Only Intradermal Inj.

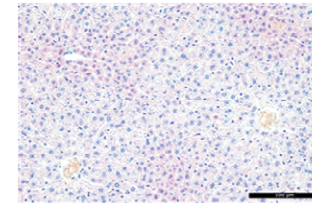


6xHPAE-EB+C7 Intradermal Inj. HD

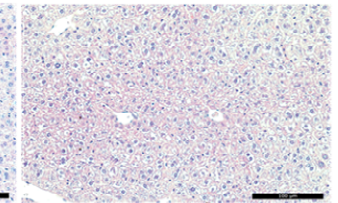


Scale bar 100µm

Control

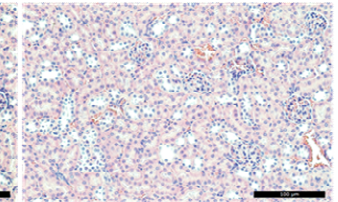
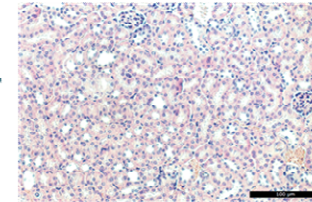


6xHPAE-EB+C7 Intradermal Inj.



Liver

Kidneys



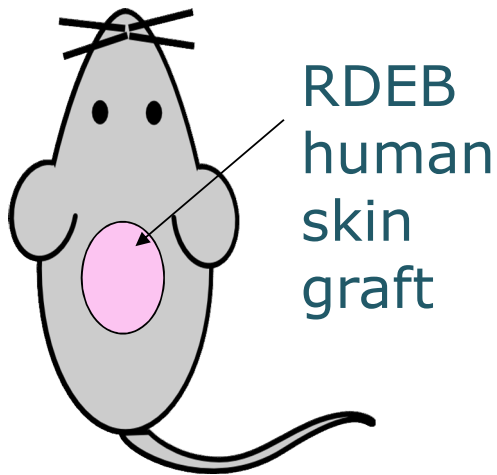
Scale bar 100µm



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# Next Steps For HPAE-EB Assessment

- 1. Find** Minimum Effective Dose (**MED**) with RDEB graft model.



## **Pilot topical HPAE-EB efficacy assessment:**

Nude mice have been grafted with human RDEB skin.

Different HPAE-EB-C7 polyplex doses will be applied topically (x3).

Detection of type VII collagen after 7 days and 4 weeks.

- 2. Safety** assessment of **repeated topical** HPAE-EB-C7 applications on immunocompetent mice.
- 3. Scaling up production** of the polyplexes.







Thank you,  
for your time and attention!



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