Cell Blebs for Intracellular Protein/Drug Delivery

James Espeleta
Susan Daniel Research Group
Protein/Drug Delivery

Protein/Drug → Cell
Protein/Drug Delivery: Therapeutics

Other pathways:
- Diabetes
- Osteoporosis
- Arthritis
- Anticoagulant
What are blebs?

Protrusions that are formed when the cell membrane detaches from its cytoskeleton.
Blebbing Buffer: .075% formaldehyde, .075% Dithiothreitol in 4 mL GPMV solution
Blebs infect target cells
How do they get in?

Blebs infect target cells

How do they get out of the bleb?
Solution: Virus Coat Proteins

Protein coats bind to receptors and are engulfed via endocytosis.
Putting the Protein/Drug into the Bleb
Transfection Step

293T Cells

+ 

5 µg VSV-G
(Virus Coat)

5 µg eGFP
(Protein/Drug to be Delivered)
Blebbing

Add Blebbing Buffer

VSV-G
eGFP
Delivering eGFP with VSVG Coated Blebs into MDCK cells
Infection

293-T Blebs (VSV-G / eGFP) → MDCK Cells
Results

(Above: eGFP + VSVG treated MDCK cells post-infection at 40x magnification with fluorescence microscope)
Delivering eGFP with Ebola GP Coated Blebs into MDCK cells
Infection

293-T Blebs (Ebola GP /eGFP) → MDCK Cells
Results

Brightfield

Brightfield + GFP

eGFP + Ebola GP

(Above: eGFP + Ebola GP treated vero cells post-infection at 40x magnification with FRAP microscope)
eGFP is soluble protein, so it is everywhere in the cell and near the cell membrane when transfected
eGFP is soluble protein, so it is diffuse and near the cell membrane when transfected.

But what if the protein is not soluble?
Solution: CRY2/CIBN

Implications of Research

1. Blebbing is versatile
   - Ebola GP and VSV-G

1. Blebbing is time efficient
   - 24–36 hours vs. 48-72 hours
Acknowledgements

**Susan Daniel Research Group**

*Principal Investigator*
Susan Daniel

*Mentor*
Hung-Lun Hsu

*Graduate Students*
Rohit Singh, Lakshmi Nathan, Han-Yuan Liu, Johana Uribe,
Zeinab Mohamed, Tiffany Tang