# Rudiments of vaccine design

---

**Last Time:** continued discussion of stealth particles
basic immunobiology underlying vaccination

**Today:** basics of vaccine design and vaccine immune responses

**Reading:** Raychaudhuri and Rock, 'Fully mobilizing host defense: building better vaccines,' *Nat. Biotech*. **16** 1025-1031 (1998)

**Supplementary Reading**:

---

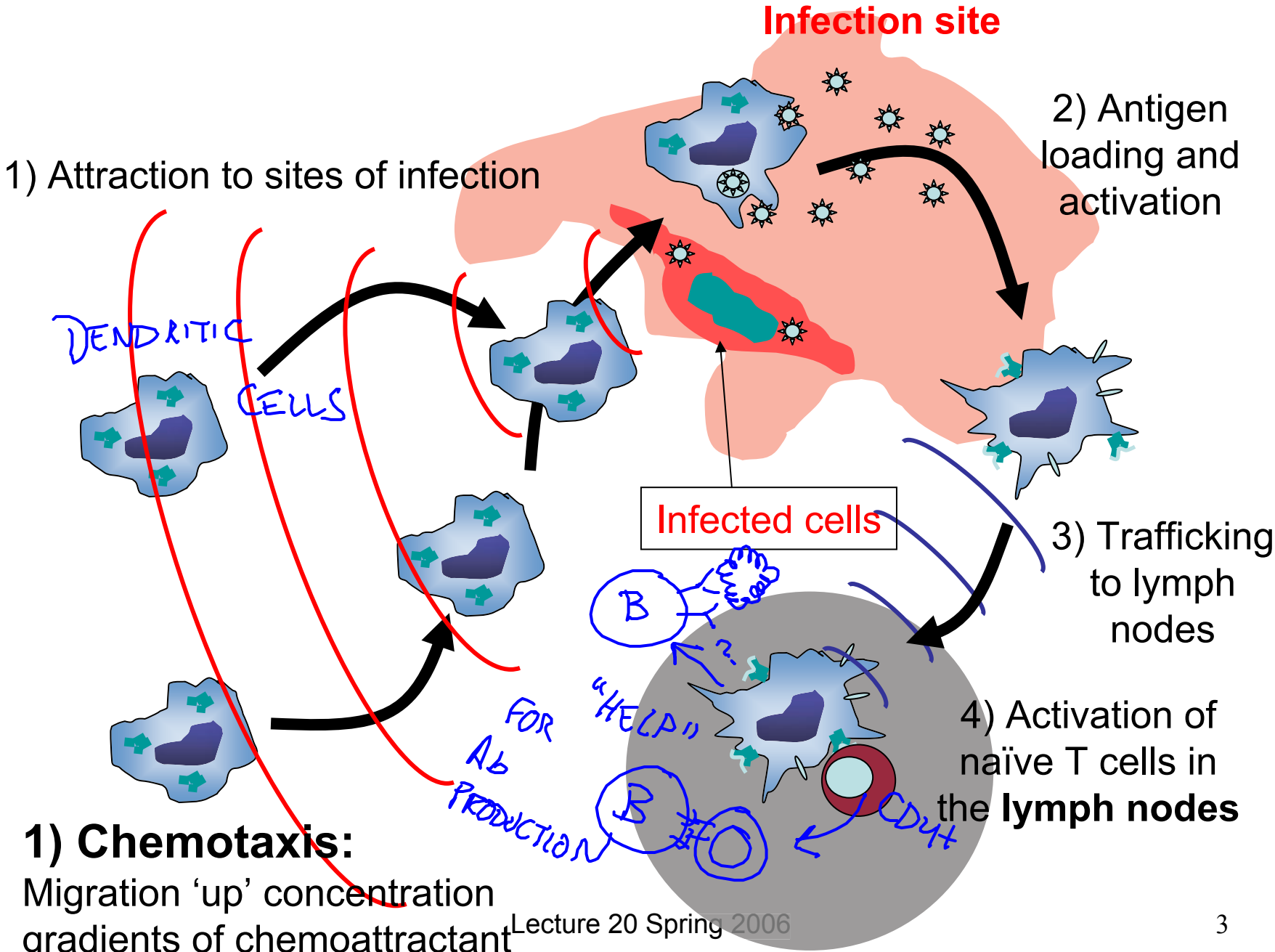
**ANNOUNCEMENTS:**

Note on take-home exam: 6-page limit includes any schematics or figures from the literature (1/3 of space max)

# KEY EFFECTORS OF ADAPTIVE IMMUNITY

Image removed due to copyright reasons.

Please see: Abbas, A. K., and A. H. Lichtman. *Cellular and Molecular Immunology*. San Diego, CA: Elsevier, 2005. ISBN: 1416023895.




**1) Chemotaxis:**
Migration 'up' concentration gradients of chemoattractant

# PAMP recognition of microbes by dendritic cells

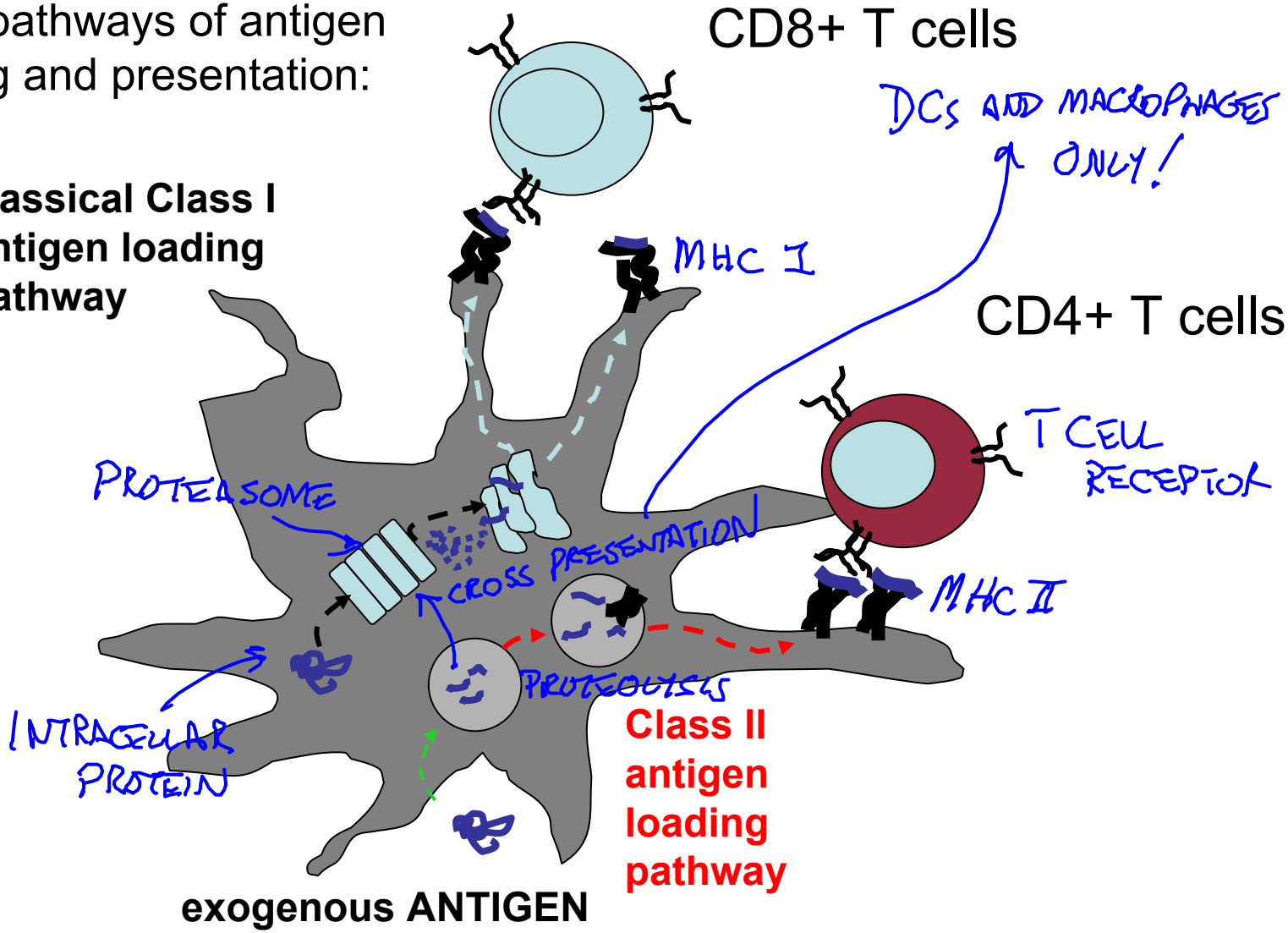
Immune cells integrate many signals to ‘fingerprint’ pathogens:

Image removed due to copyright restrictions.
Please see: Huang, et al. *Science* 294 (2001): 3870.

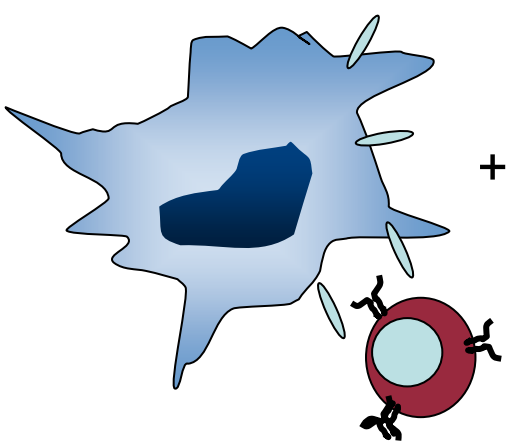
Image removed due to copyright restrictions.
Please see: Kawai, and Akira. *Curr Opin Immunol* 17 (2005): 338-344.

# Biology of dendritic cells in T cell activation

Classical pathways of antigen processing and presentation:

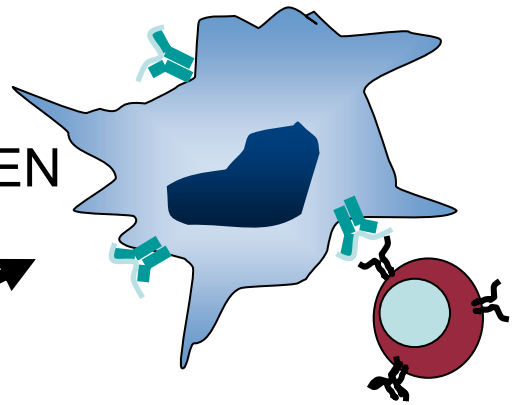


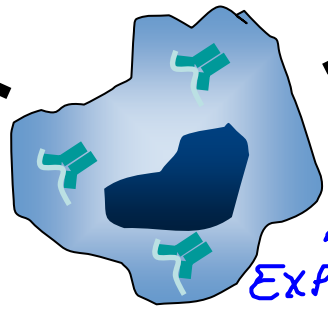
# Antigen is *one* of (at least) *two* signals that must be delivered by a vaccine



+DC ACTIVATION

+ANTIGEN





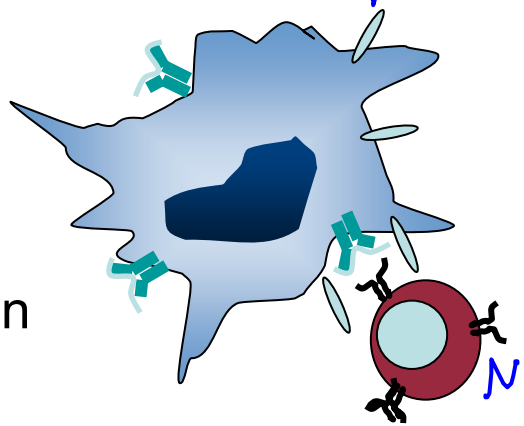
•**NO T CELL ACTIVATION**

•**T CELLS TOLERIZED**

ACTIVATION DRIVES EXPRESSION OF COSTIMULATORY RECEPTORS CD80, CD86, CD40, ...

+ANTIGEN
+DC ACTIVATION

PAMPS

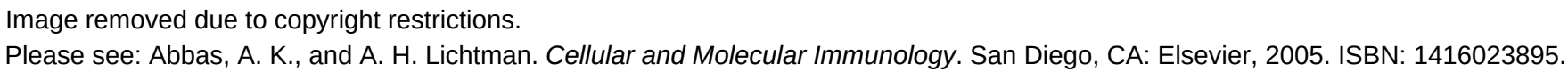


•**MAXIMAL T CELL PROLIFERATION**
•**GENERATION OF FULL EFFECTOR FUNCTIONS**
•**GENERATION OF MEMORY T CELLS**

NAIVE T CELL

Signal 1 - antigen
Signal 2 - costimulation

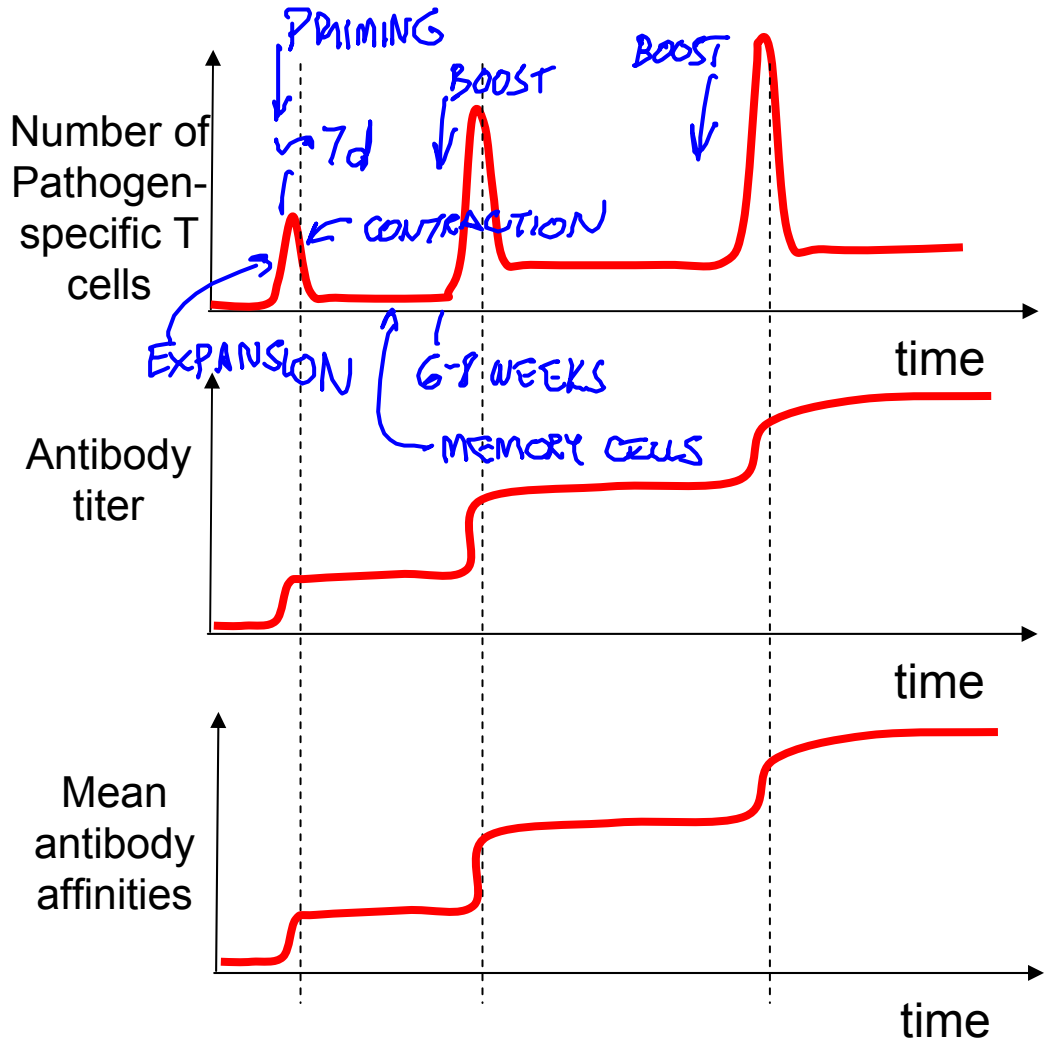
# B cell activation



# Induction of immunological memory (the basis of vaccination)

PROPHYLACTIC

Image removed due to copyright restrictions.
Please see: Ahmed. *Science* 300 (2003): 263-264.



# OBJECTIVES OF VACCINATION

T + B MEMORY:

MEM. T CELLS => RESPOND QUICKLY DIRECTLY AT INFECTION SITE

→ POPULATE PERIPHERAL TISSUES (NAIVE T STAY IN CIRCUIT LNS ⟷ BLOOD)

MEM. B CELLS → BONE MARROW / PERIPHERAL TISSUES

Image removed due to copyright restrictions.
Please see: Neutra, and Kozlowski. *Nat Rev Immunol* 6 (2006): 148-158.

# Prophylactic vs. therapeutic immunization

**Two situations where vaccination is of interest:**

(1) Therapeutic vaccine: TREAT AN ONGOING CONDITION

CANCER
HIV
⋮

↓

GENERATE EFFECTOR CELLS AGAINST PATHOGEN/TUMOR

↓

MADE CHALLENGING BY ONGOING SUBVERSIVE FUNCTIONS OF MICROBES/TUMORS

(2) Prophylactic vaccine:

↳ PREPARE MEMORY CELLS AGAINST FUTURE EXPOSURE

→ MADE CHALLENGING BY NEED FOR SAFETY

# ROUTES OF IMMUNIZATION

Image removed due to copyright restrictions.
Please see: Neutra, and Kozlowski. *Nat Rev Immunol* 6 (2006): 148-158.

# Rudimentary components of vaccines

- Antigen: FRAGMENT FROM ORIGINAL PATHOGEN/TUMOR, OFTEN PROTEIN (PEPTIDE FOR T CELLS)

ALUM DRIVES "Th2" RESPONSE → GENERATES Abs, BUT T CELL RESPONSE IS POOR
(ALLERGIC REACTION)

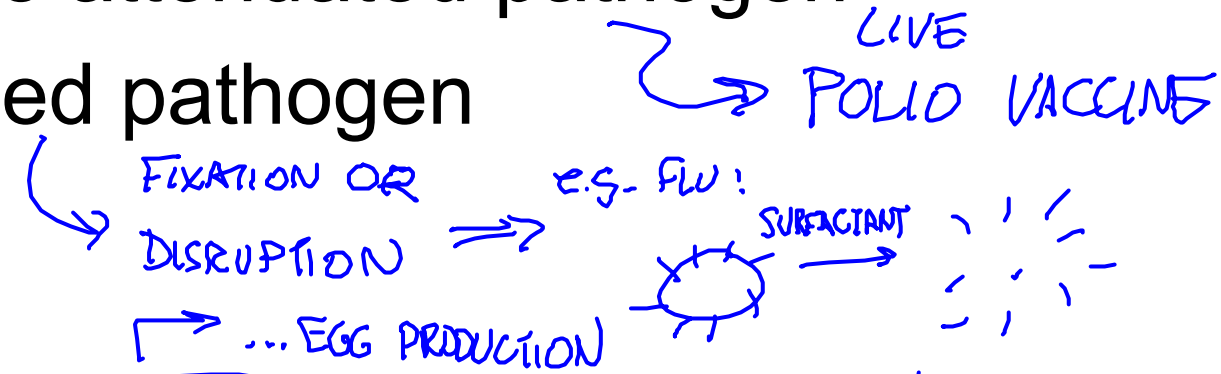
- Adjuvant: "2ND SIGNAL" THAT ACTIVATES DCs, PROMOTES EFFECTOR AND MEMORY LYMPHOCYTE DEVELOPMENT

DEPOT/ANTIGEN

↳ ONLY 2 FDA-APPROVED ADJUVANTS:
① ALUM (ALUMINUM HYDROXIDE) ↗ PROTEIN ADSORBS TO ALUM
② MF59 (SQUALENE (OIL) + SURFACTANTS)

# Compositions of vaccines- clinical and experimental


- Live attenuated pathogen
- Killed pathogen




— MANUFACTURE + CHARACTERIZATION IS CHALLENGING

— SAFETY IS AN ISSUE (e.g., HIV)

— STRONGEST IMMUNE RESPONSE

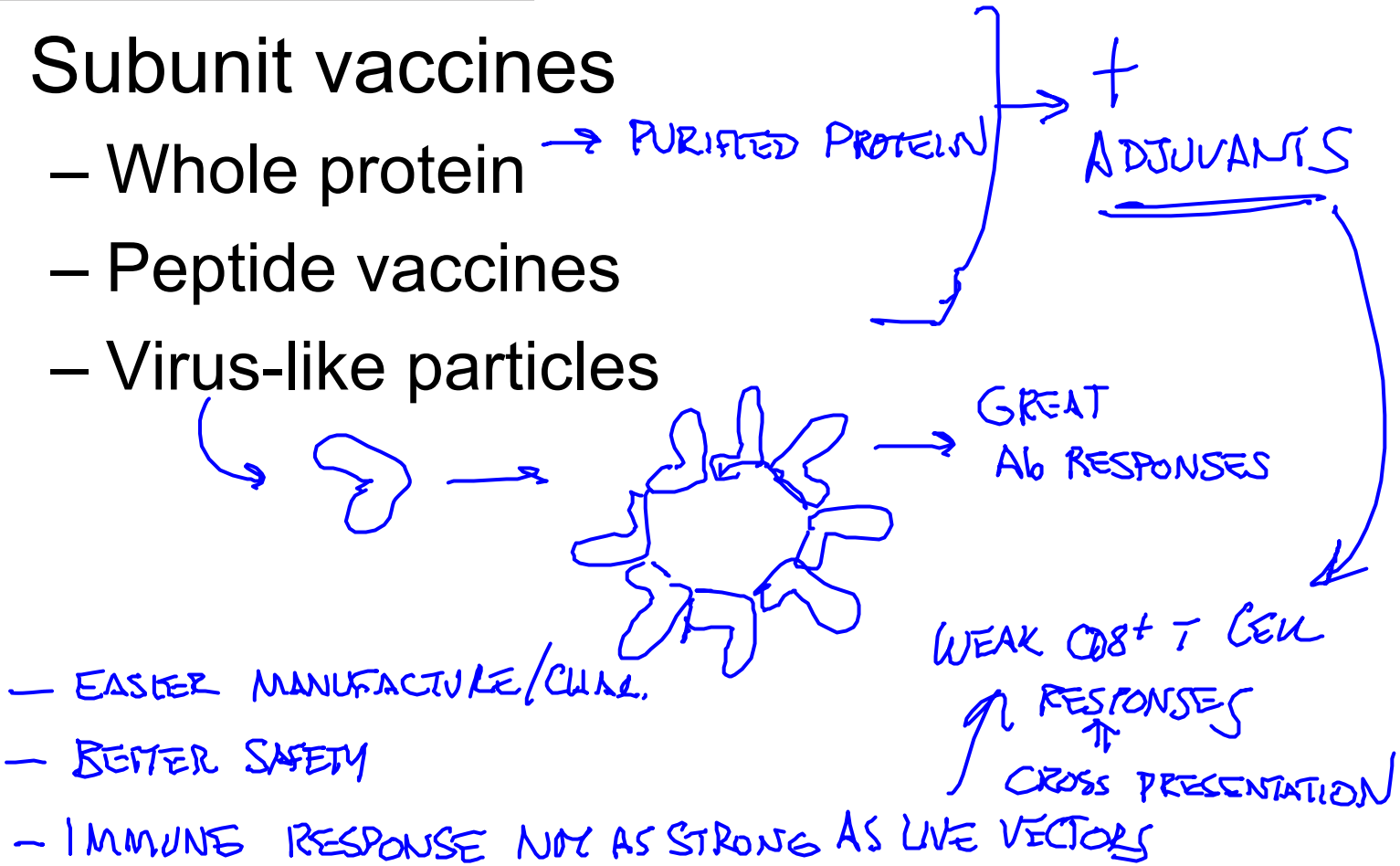
↳ BUILT-IN ADJUVANT → PAMPS

SELF-REPLICATING ANTIGEN

# Compositions of vaccines- clinical and experimental

## 'engineered' vaccines:

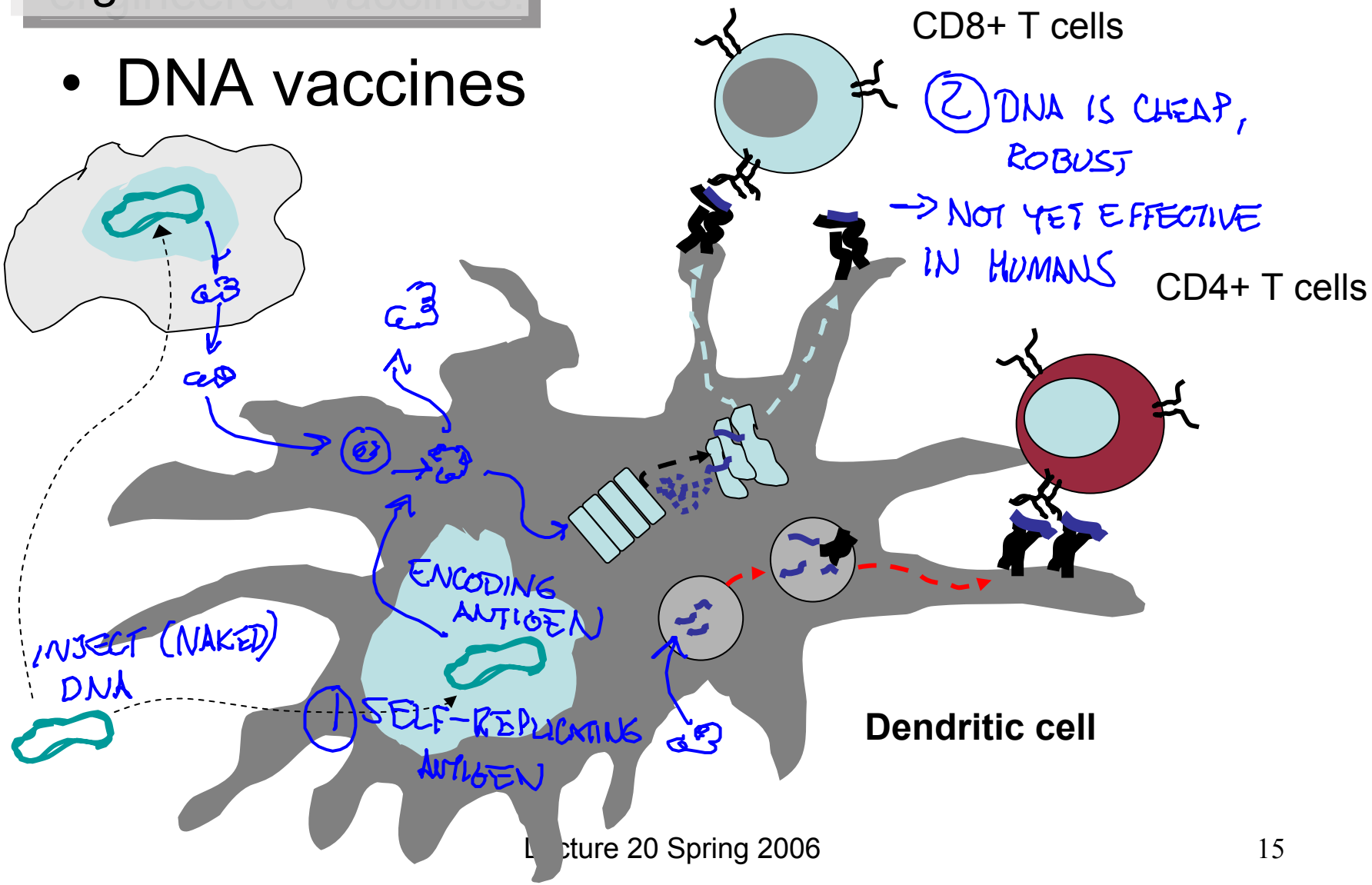
- Subunit vaccines
  - Whole protein
  - Peptide vaccines
  - Virus-like particles



# Compositions of vaccines- clinical and experimental

## 'engineered' vaccines:

- DNA vaccines



# Compositions of vaccines- clinical and experimental

## ‘engineered’ vaccines:

- DNA vaccines

# Existing vaccines

Table removed due to copyright restrictions.
Please see: Table 1 in Ada, G. "Advances in Immunology - Vaccines and Vaccination." *New England Journal of Medicine* 345 (2001): 1042-53.

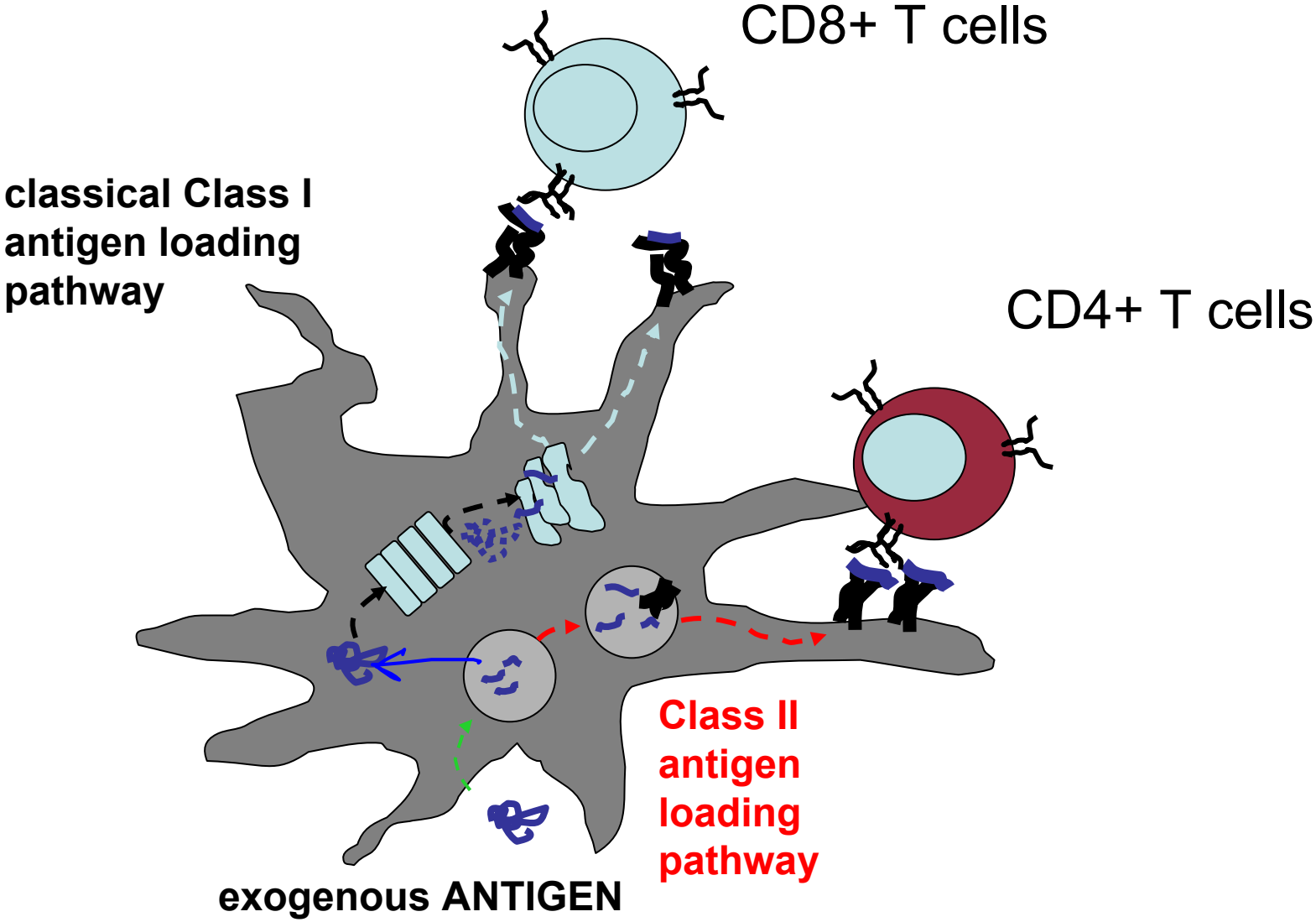
# Existing vaccines

Table removed due to copyright restrictions.
Please see: Table 1 in Ada, G. "Advances in Immunology - Vaccines and Vaccination." *New England Journal of Medicine* 345 (2001): 1042-53.

## Biomaterials to adjuvant subunit vaccines:

**intracellular drug delivery** and the design of protein and peptide vaccines that stimulate cytotoxic T cell responses

# Cross presentation and Particulate antigen delivery



# Pathways of intracellular import

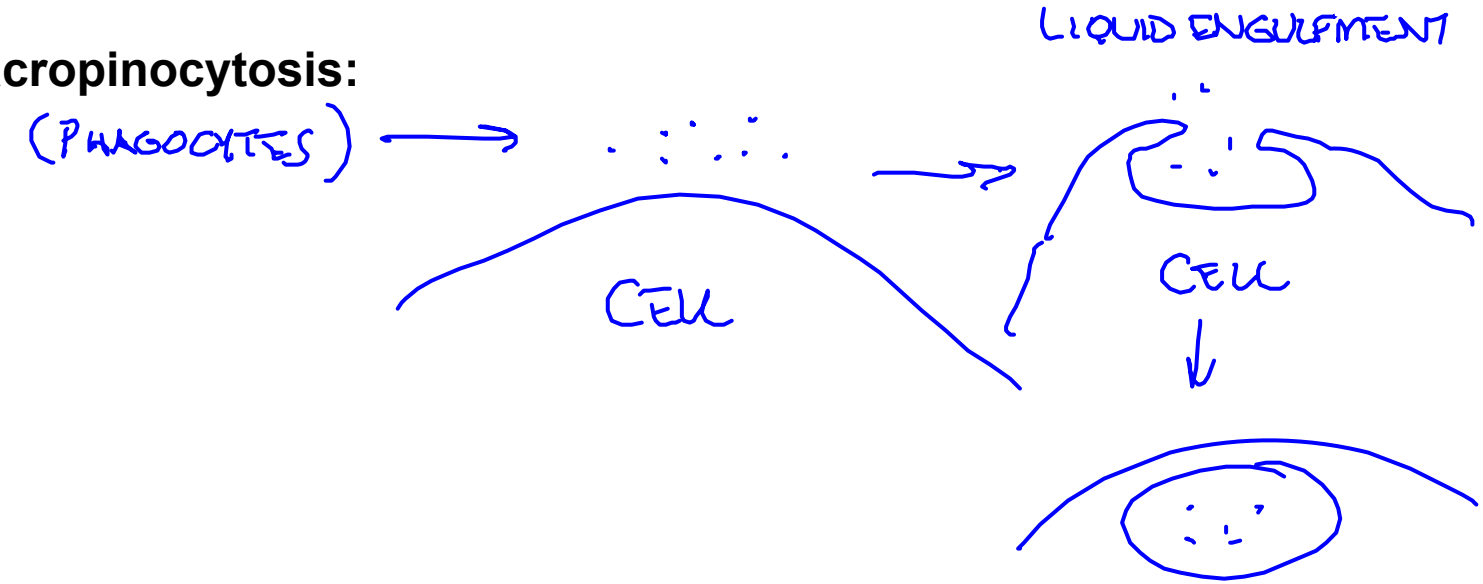
**Endocytosis:**
(nearly all cells)

Image removed due to copyright restrictions.
Please see: Figure 13-46 in Bruce, Alberts, et al. *Molecular Biology of the Cell.* New York, NY: Garland, 2004.

# Pathways of intracellular import

Image removed due to copyright restrictions.
Please see: http://www.cellsalive.com

**macropinocytosis:**



# How do exogenous antigens get presented on class I MHC?

Image removed due to copyright restrictions.
Please see: Figure 13-46 in Bruce, Alberts, et al. *Molecular Biology of the Cell*. New York, NY: Garland, 2004.

# Particle-stimulated cross presentation

Graph removed due to copyright restrictions.
Please see: Kovacs-Bankowski, et al. *PNAS* 90 (1993): 4942-4946.

Image removed due to copyright restrictions.
Please see: Lehner, and Cresswell. *Curr Opin Immunol* 16, no. 82 (2004).

# Particle-stimulated cross presentation

Images and graph removed due to copyright restrictions.
Please see: Rodrigues, et al. *Nat Cell Biol* 1 (1999): 362.

# ENDOSOMAL ESCAPE:

Enhancing cross presentation
cytosolic delivery of large macromolecules

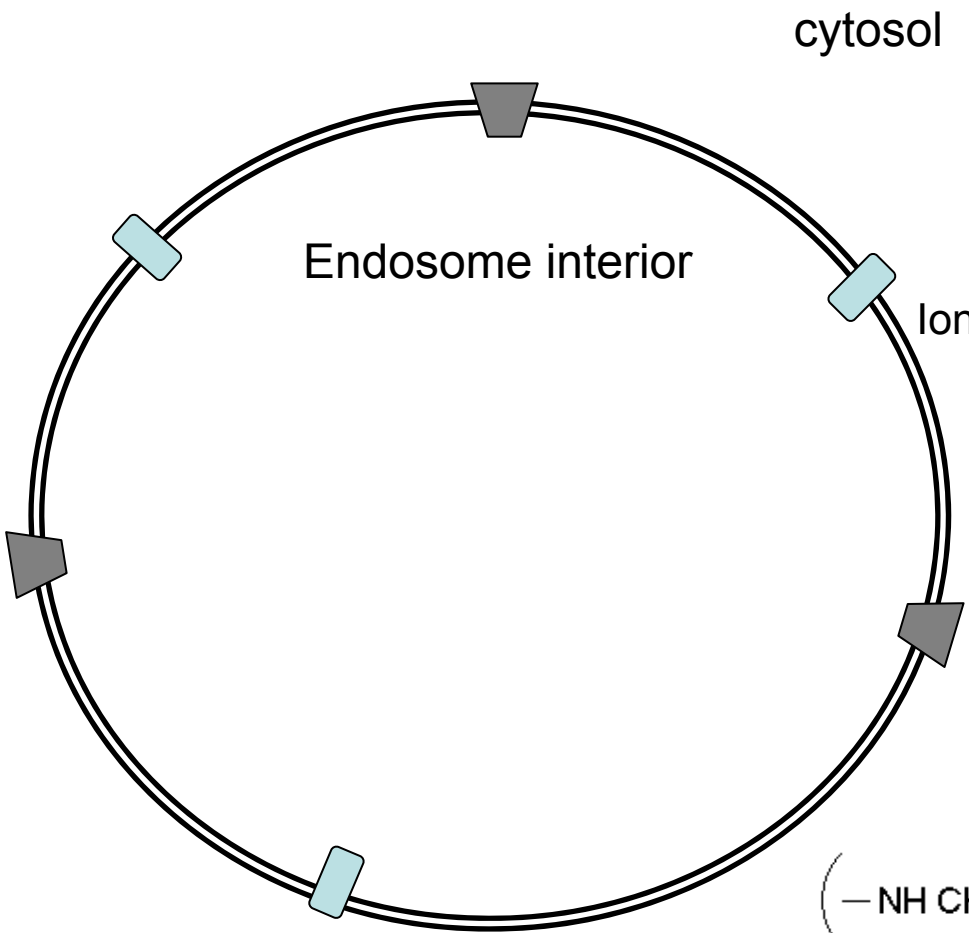
# Mechanisms for endosomal escape by polymeric carriers

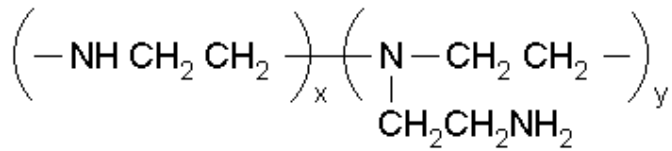
(1) ‘proton sponge’ effect

(2) Direct membrane interaction/destabilization

(3) pH-activated CPPs

# Proton sponge effect




polyethyleneimine

# Further Reading


1. Moghimi, S. M., Hunter, A. C. & Murray, J. C. Long-circulating and target-specific nanoparticles: theory to practice. *Pharmacol Rev* **53**, 283-318 (2001).
2. Hawiger, J. Noninvasive intracellular delivery of functional peptides and proteins. *Curr Opin Chem Biol* **3**, 89-94 (1999).
3. Derossi, D. et al. Cell internalization of the third helix of the Antennapedia homeodomain is receptor-independent. *J Biol Chem* **271**, 18188-93 (1996).
4. Falnes, P. O. & Sandvig, K. Penetration of protein toxins into cells. *Curr Opin Cell Biol* **12**, 407-13 (2000).
5. Joliot, A. & Prochiantz, A. Transduction peptides: from technology to physiology. *Nat Cell Biol* **6**, 189-96 (2004).
6. Schwarze, S. R., Ho, A., Vocero-Akbani, A. & Dowdy, S. F. In vivo protein transduction: delivery of a biologically active protein into the mouse. *Science* **285**, 1569-72 (1999).
7. Snyder, E. L. & Dowdy, S. F. Cell penetrating peptides in drug delivery. *Pharm Res* **21**, 389-93 (2004).
8. Thoren, P. E. et al. Membrane binding and translocation of cell-penetrating peptides. *Biochemistry* **43**, 3471-89 (2004).
9. Asokan, A. & Cho, M. J. Exploitation of intracellular pH gradients in the cellular delivery of macromolecules. *J Pharm Sci* **91**, 903-13 (2002).
10. Sandgren, S., Cheng, F. & Belting, M. Nuclear targeting of macromolecular polyanions by an HIV-Tat derived peptide. Role for cell-surface proteoglycans. *J Biol Chem* **277**, 38877-83 (2002).
11. Yatvin, M. B., Kreutz, W., Horwitz, B. A. & Shinitzky, M. Ph-Sensitive Liposomes - Possible Clinical Implications. *Science* **210**, 1253-1254 (1980).
12. Lee, K. D., Oh, Y. K., Portnoy, D. A. & Swanson, J. A. Delivery of macromolecules into cytosol using liposomes containing hemolysin from Listeria monocytogenes. *J Biol Chem* **271**, 7249-52 (1996).
13. Bhakdi, S. et al. Staphylococcal alpha-toxin, streptolysin-O, and Escherichia coli hemolysin: prototypes of pore-forming bacterial cytolysins. *Arch Microbiol* **165**, 73-9 (1996).
14. Raychaudhuri, S. & Rock, K. L. Fully mobilizing host defense: building better vaccines. *Nat Biotechnol* **16**, 1025-31 (1998).
15. Falo, L. D., Jr., Kovacsovics-Bankowski, M., Thompson, K. & Rock, K. L. Targeting antigen into the phagocytic pathway in vivo induces protective tumour immunity. *Nat Med* **1**, 649-53 (1995).
16. Murthy, N., Campbell, J., Fausto, N., Hoffman, A. S. & Stayton, P. S. Bioinspired pH-Responsive Polymers for the Intracellular Delivery of Biomolecular Drugs. *Bioconjug Chem* **14**, 412-9 (2003).
17. Shi, G., Guo, W., Stephenson, S. M. & Lee, R. J. Efficient intracellular drug and gene delivery using folate receptor-targeted pH-sensitive liposomes composed of cationic/anionic lipid combinations. *J Control Release* **80**, 309-19 (2002).

# Further Reading


1. Ada, G. Advances in immunology - Vaccines and vaccination. New England Journal of Medicine 345, 1042-1( (2001).
2. Donnelly, J. J., Wahren, B. & Liu, M. A. DNA vaccines: progress and challenges. J Immunol 175, 633-9 (2005
3. Eldridge, J. H. et al. Controlled Vaccine Release in the Gut-Associated Lymphoid-Tissues .1. Orally-Administe Biodegradable Microspheres Target the Peyers Patches. Journal of Controlled Release 11, 205-214 (1990).
4. Ermak, T. H., Dougherty, E. P., Bhagat, H. R., Kabok, Z. & Pappo, J. Uptake and transport of copolymer biodegradable microspheres by rabbit Peyer's patch M cells. Cell Tissue Res 279, 433-6 (1995).
5. Finn, O. J. Cancer vaccines: between the idea and the reality. Nat Rev Immunol 3, 630-41 (2003).
6. Foged, C., Sundblad, A. & Hovgaard, L. Targeting vaccines to dendritic cells. Pharm Res 19, 229-38. (2002).
7. Garcea, R. L. & Gissmann, L. Virus-like particles as vaccines and vessels for the delivery of small molecules. Opin Biotechnol 15, 513-7 (2004).
8. Letvin, N. L. Strategies for an HIV vaccine. Journal of Clinical Investigation 110, 15-20 (2002).
9. Letvin, N. L., Barouch, D. H. & Montefiori, D. C. Prospects for vaccine protection against HIV-1 infection and A Annu Rev Immunol 20, 73-99 (2002).
10. Levine, M. M. & Sztein, M. B. Vaccine development strategies for improving immunization: the role of modern immunology. Nat Immunol 5, 460-4 (2004).
11. Mackay, I. R. & Rosen, F. S. Vaccines and Vaccination. New England Journal of Medicine 345, 1042-1053 (2
12. Murthy, N. et al. A macromolecular delivery vehicle for protein-based vaccines: acid-degradable protein-loade microgels. Proc Natl Acad Sci U S A 100, 4995-5000 (2003).
13. Mutwiri, G. et al. Induction of mucosal immune responses following enteric immunization with antigen delivere alginate microspheres. Vet Immunol Immunopathol 87, 269-76 (2002).
14. Mutwiri, G., Bowersock, T. L. & Babiuk, L. A. Microparticles for oral delivery of vaccines. Expert Opin Drug De 791-806 (2005).
15. O'Hagan, D. T., Singh, M. & Ulmer, J. B. Microparticles for the delivery of DNA vaccines. Immunol Rev 199, 1 (2004).
16. Pinto, A. R., Fitzgerald, J. C., Gao, G. P., Wilson, J. M. & Ertl, H. C. Induction of CD8+ T cells to an HIV-1 ant upon oral immunization of mice with a simian E1-deleted adenoviral vector. Vaccine 22, 697-703 (2004).
17. Shalaby, W. S. Development of oral vaccines to stimulate mucosal and systemic immunity: barriers and nove strategies. Clin Immunol Immunopathol 74, 127-34 (1995).
18. Singh, M. & O'Hagan, D. Advances in vaccine adjuvants. Nat Biotechnol 17, 1075-81 (1999).
19. Stevenson, F. K. DNA vaccines and adjuvants. Immunol Rev 199, 5-8 (2004).