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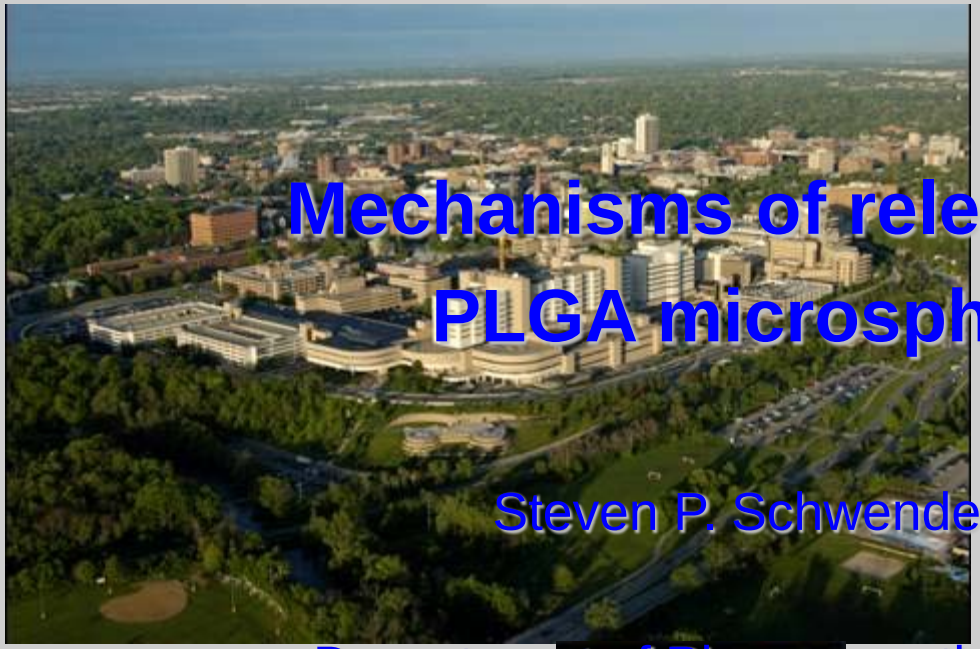

Mechanisms of release from PLGA microspheres

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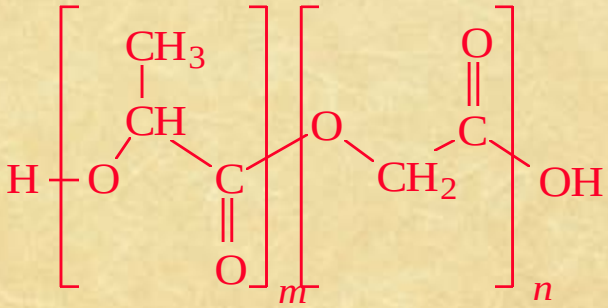
Poly(lactic-*co*-glycolic acid) (PLGA) for controlled drug delivery

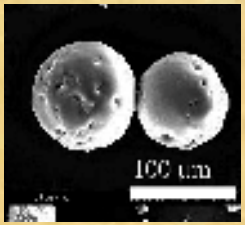
- Structure



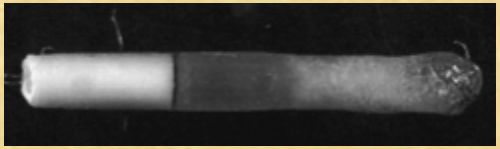
e.g., PLGA 50/50, m/n = 1
PLGA 75/25, m/n = 3

Mw ~ 10 kDa - 100 kDa

- Advantages
  - wide range of properties
  - ease of processing and biodegradable
  - predictable *in vivo* degradation kinetics
  - used in numerous FDA approved products
  - No daily injections
  - Control release rate
  - Lower systemic toxicity
  - Reduce booster doses (vaccines)
- Major configurations of injectable devices



microspheres (1 - 100 μm)



millicylinders (∅ = 0.8-1.5 mm)

in-situ forming implants

Examples of PLGA microspheres used clinically

	Name	*Company*	*Disease*
Peptides	Sandostatin© LAR©	Novartis	Acromegaly
	Lupron©	TAP	Prostate and breast cancer
	Decapeptyl© Depot	Ferring	Prostate cancer, endometriosis
	Trelstar©	Pfizer	Prostate cancer
	Pamorelin ©	Ipsen	Prostate cancer
	Somatuline© LA	Ipsen	Acromegaly
	Suprecur MP© (Japan)	Mochida	Endometriosis
Proteins	Nutropin Depot®	Genentech	Pediatric GH deficiency
Small Mol.	Vivitrol©	Cephalon	Alcoholism
	Risperidal© Consta©	Janssen	Schizophrenia
	Arestin©	OraPharma	Peridontal disease
	Parlodel LA ©	Sandoz	Parkinson's, acromegaly

Wischke, C.; Schwendeman, S. P., *International Journal of Pharmaceutics* 2008, 364, (2), 298-327.

No generic PLGA-based drug products approved by US FDA

FDA's Regulatory Science Program for Generic PLA/ PLGA-Based Drug Products

Wednesday, June 15, 2016

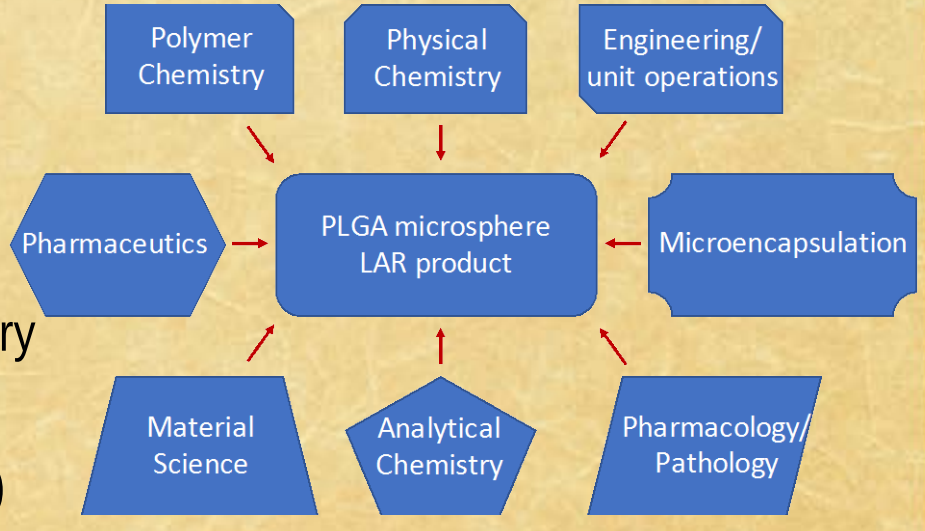
Yan Wang
Center for Drug Evaluation and Research
Wen Qu
Center for Drug Evaluation and Research
Stephanie H. Choi
Center for Drug Evaluation and Research

- FDA

"Currently, no PLA/PLGA-based generic drug products have been approved."

Challenges for PLGA science

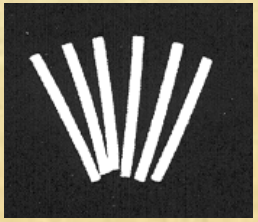
- Strong diversity of scientific disciplines needed (polymer chemistry, material science, pharmaceutics, engineering/unit ops, p-chemistry, etc)
- Most research focused on delivering a drug and not understanding mechanism
- Tendency to oversimplify complex physical chemistry (e.g., small vs. large molecular drug, role of microsphere size)
- Shortage of specialized assays and insufficient research equipment in academic labs
- Key development science proprietary (manufacturing, scale up, composition-equivalent formulation)

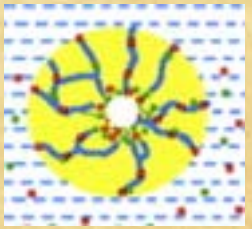


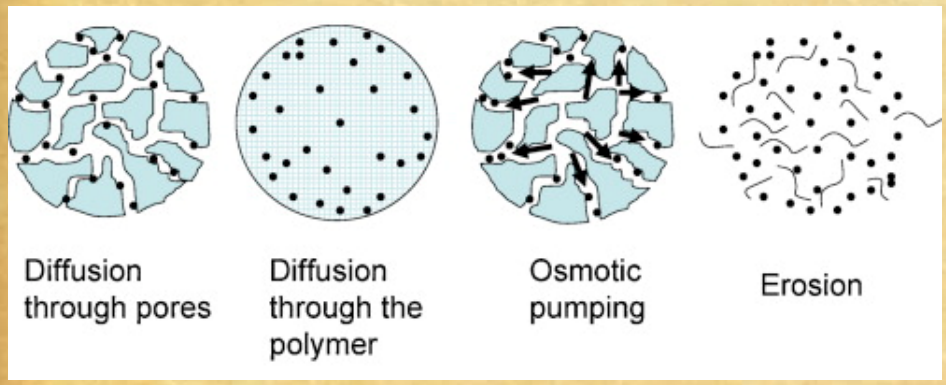
How is slow release commonly achieved from PLGA?

Combination of 3 basic phenomena —

- ✧ Diffusion
- ✧ Osmotic pressure/swelling
- ✧ Bioerosion when polymer chains become small enough to give way to stresses and/or dissolve







Fredenberg et al., *Int. J. Pharm.*, 415, 34–52 (2011)

Additional mechanisms that influence drug release from PLGA?

3 other phenomena not normally discussed —

- ✧ Passive healing (i.e., spontaneous pore closing)
- ✧ Desorption (or decoupling polymer/drug interactions)
- ✧ Dynamic polymer microstructural changes

4 cases of diffusion through non-degradable polymer matrices

- Case 1
  - $C_s > C_d$; transport in polymer phase
- Case 2
  - $C_s < C_d$; transport in polymer phase

C_s = drug solub. In polymer
C_d = drug loading (mass/vol.)
L = thickness of matrix
D_{im} = drug diffusion coef in polymer

Case 1: The drug is molecularly dissolved in the polymer matrix and drug diffusion occurs via a solution-diffusion mechanism.

$$\frac{dM_i}{A dt} = 2c_d\left(\frac{D_{im}}{\pi}\right)^{1/2} t^{-1/2} \qquad (9)$$

Early-time solution

Case 2: The drug is dispersed in the polymer matrix (i.e., it is loaded above its solubility limit) and diffusion occurs via a solution-diffusion mechanism.

$$\frac{dM_i}{A dt} = \frac{1}{2}\left[D_{im} c_s (2c_d - c_s)\right]^{1/2} t^{-1/2} \qquad (10)$$

Higuchi solution

(Langer & Peppas, *Biomaterials*, 1981)

Exact solutions

$$\frac{1}{A}\frac{dM}{dt} = \frac{8 D_{im} C_d}{L}\sum_{n=0}^{\infty} e^{-D_{im}(2n+1)^2 \pi^2 t/L^2} \qquad (2 \text{ faces})$$

$$\frac{1}{A}\frac{dM}{dt} = C_s\sqrt{\frac{D_{im} t}{\pi}}\frac{1}{erf\left(\eta^R\right)}$$

where

$$F\left(\eta^R\right) = \left(\frac{C_d}{C_s} - 1\right)\eta^R - \frac{1}{\sqrt{\pi}}\frac{e^{-(\eta^R)^2}}{erf(\eta^R)} = 0$$

defines the root, η^R

4 cases of diffusion through non-degradable polymer matrices

- Case 3
  - $C_s < C_d$; transport in pores
- Case 4
  - $C_s > C_d$; transport in pores

C_s = drug solub. In polymer
C_d = drug loading (mass/vol.)
L = thickness of matrix
D_{iw} = drug diffusion coef in water
C_{sw} = drug solub in water
ε = matrix porosity
τ = matrix tortuosity

Case 3: The drug is dissolved in the polymer matrix and diffusion occurs through water-filled pores in the matrix

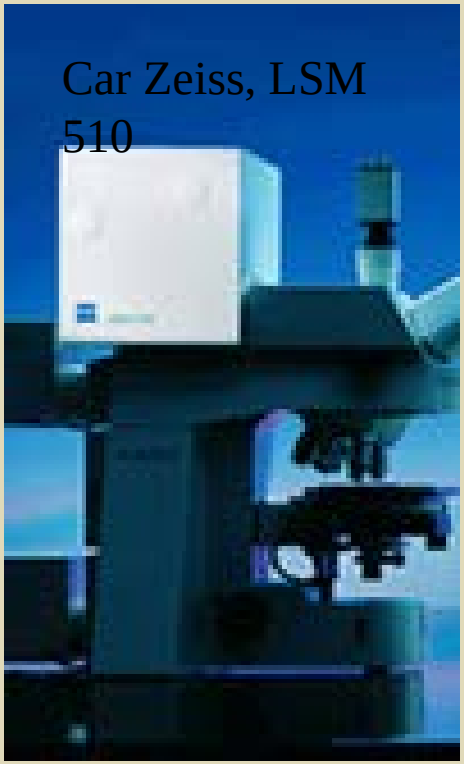
$$\frac{dM_i}{A\,dt} = 2c_d\left(\frac{D_{iw}\epsilon}{\tau\pi}\right)^{1/2} t^{-1/2} \qquad (11)$$

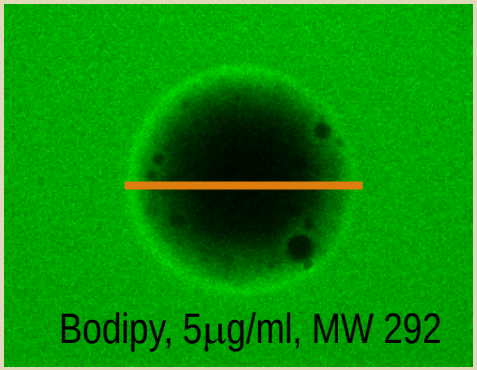
Case 4: The drug is dispersed in the polymer matrix and diffusion occurs through water-filled pores in the matrix

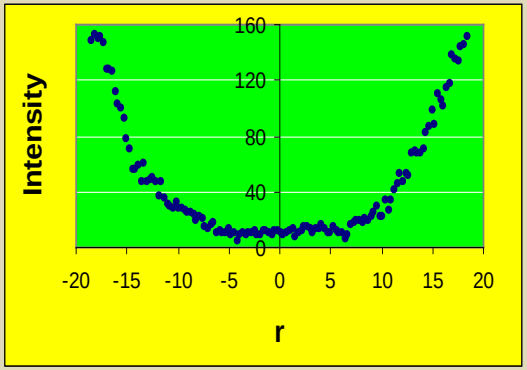
$$\frac{dM_i}{A\,dt} = \frac{1}{2}\left[\frac{D_{iw}\epsilon}{\tau}c_{sw}(2c_d - \epsilon c_{sw})\right]^{1/2} t^{-1/2} \qquad (12)$$

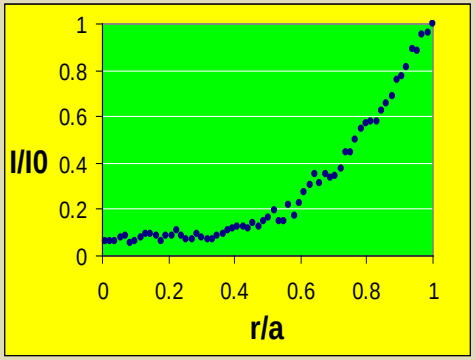
(Langer & Peppas, *Biomaterials*, 1981)

Monitoring polymer diffusion of pH-independent dye by LSCM







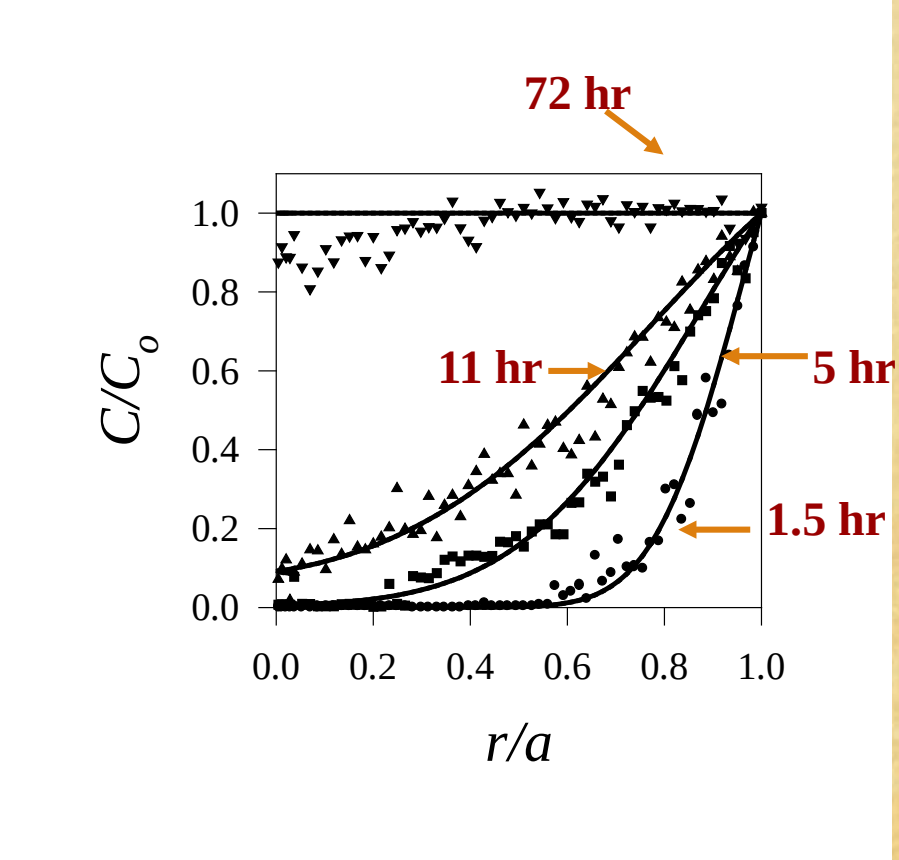


$$\frac{C}{C_0} = \frac{1}{r/a}\sum_{n=0}^{\infty}\left(erfc\frac{(2n+1)(1-r/a)}{2\sqrt{Dt/a^2}} - erfc\frac{(2n+1)(1+r/a)}{2\sqrt{Dt/a^2}}\right)$$

$$\rightarrow D_{eff}$$

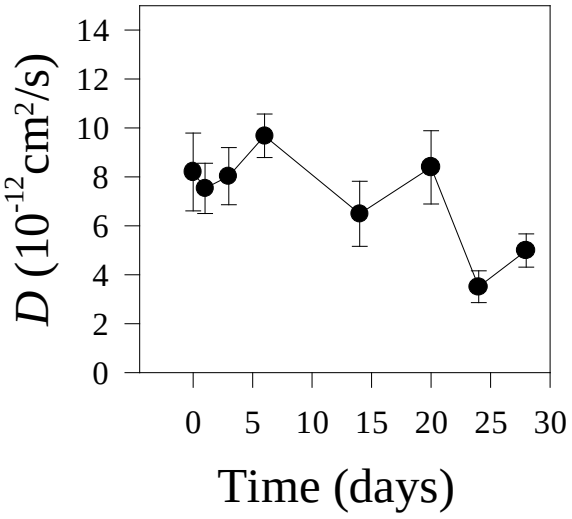
Examples of measured and fitted probe concentration profile inside microparticles

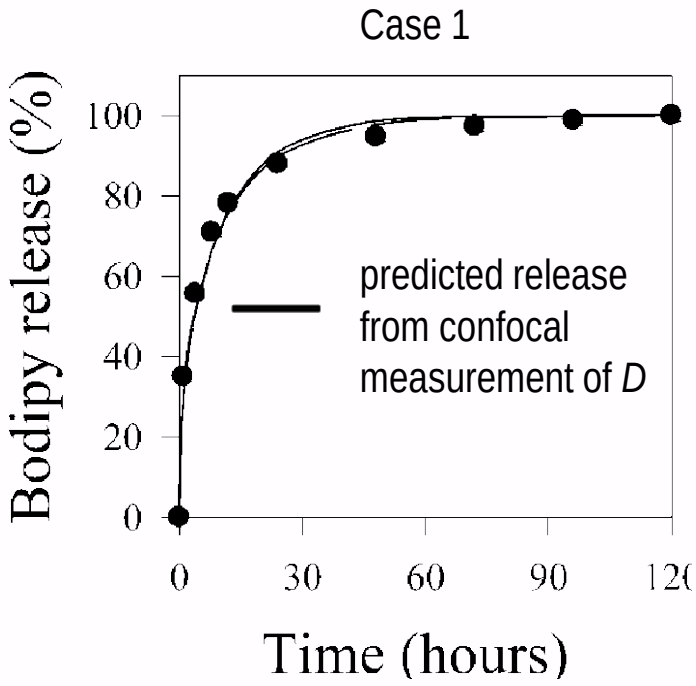
time (hr)	D_{eff} (cm^2/s)
1.5	$6.7 \pm 2.0 \times 10^{-12}$
5.0	$6.5 \pm 1.3 \times 10^{-12}$
11.0	$5.7 \pm 1.2 \times 10^{-12}$



(Kang & Schwendeman, *Macromolecules*, 2003)

Kinetics of Bodipy effective diffusivity and predictability of Dye release



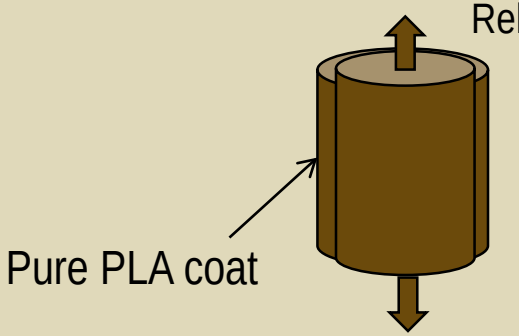


(Kang & Schwendeman, *Macromolecules* 2003)

Release by diffusion with loading >> percolation threshold

Rationale:

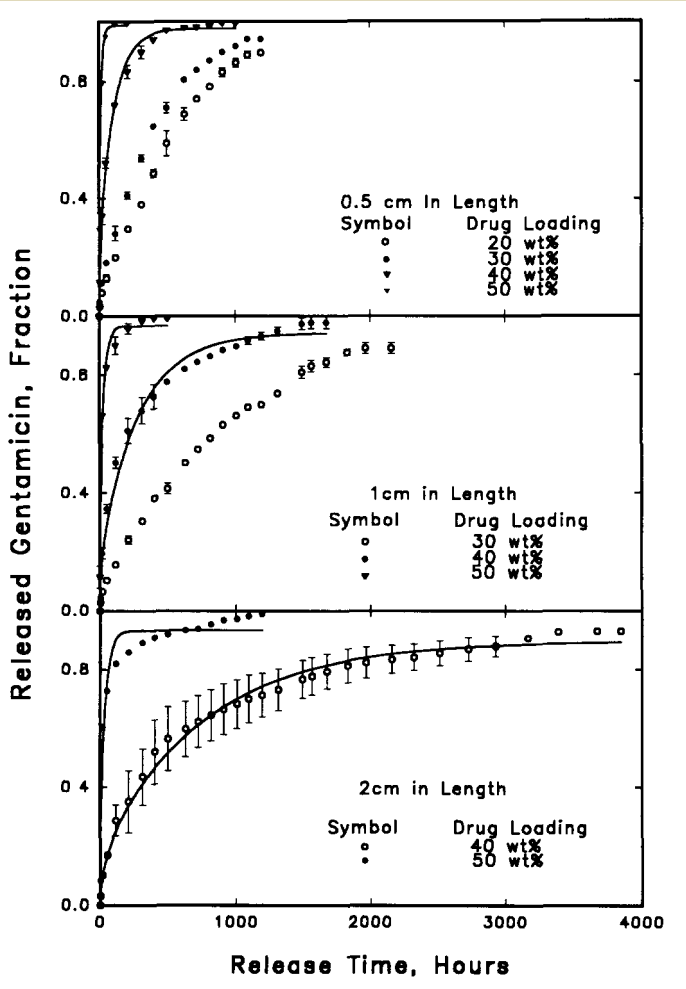
- If diffusion in polymer is much more rapid than polymer degradation, get cases 1 or 2
- If use very high loading >> percolation threshold, then get cases 3 or 4



Case 2
> 40% load

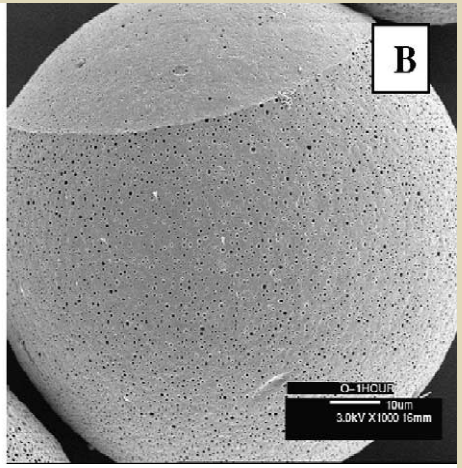
PLA, Mv = 32.6 kD

(Zhang *et al.*, *J. Cont. Rel.*, 1994)



Burst release of octreotide from PLGA microspheres

- Observations
  - Initial pores, pore creation, and swelling
  - floating microspheres allowed contrast of time frames
  - pore closing with loss of permeability



(Wang *et al.*, *J. Cont. Rel.*, 2002)

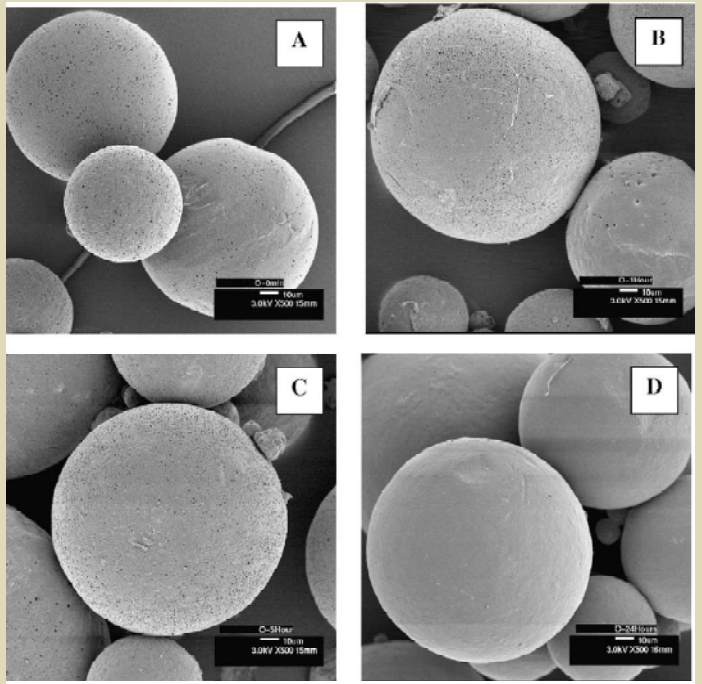
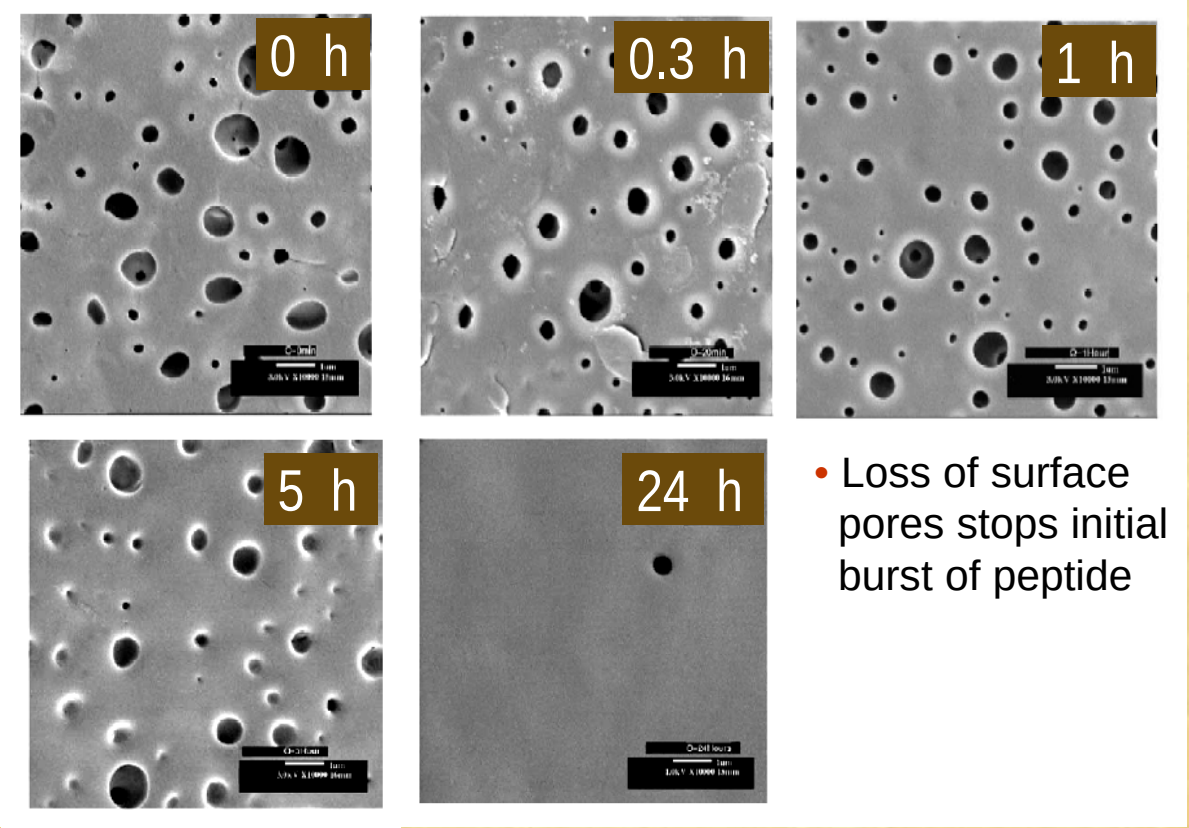


Fig. 6. Overview of external morphology of octreotide acetate-loaded PLGA biodegradable microspheres after 0-h (A), 1-h (B), 5-h (C) and 24-h (D) incubation times in the release medium at 500× magnification by SEM.

Pore network for diffusion during release not static

Polymer healing (spontaneous pore closing)



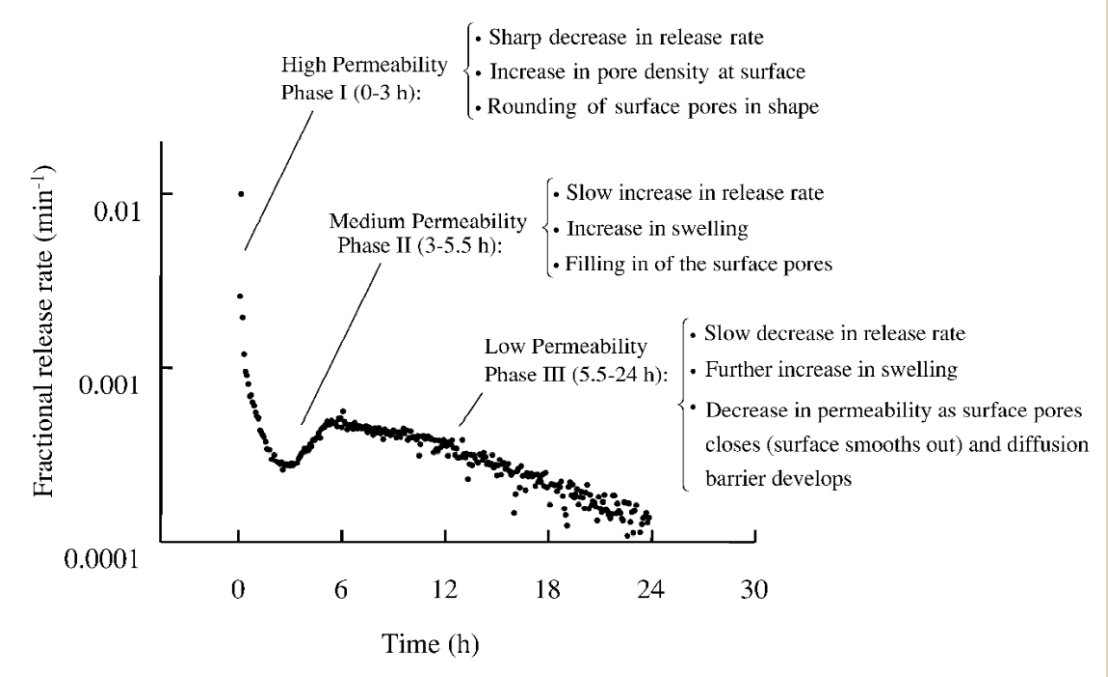
- Loss of surface pores stops initial burst of peptide

Morphology of octreotide acetate-loaded PLGA microspheres after incubation at 37˚C in acetate buffer solution.

(Wang, et al., *J. Cont. Rel.*, *82*, 289-307 (2002))

Burst release of octreotide from PLGA microspheres

- Three phases identified
  1. High permeability (initial holes and rapid pore opening)
  2. Medium permeability (swelling overcomes shrinking diffusion path)
  3. Pore closing



(Wang *et al.*, *J. Cont. Rel.*, 2002)

Pore closing in PLGA-Glucose microspheres captured as a function of temperature

- Observations
  - Higher temp increases pore closing rate
  - Both dextran and BSA obey same behavior

Table 2. Fitted and Calculated Parameters of Macromolecular Release from PLGA- Glu Microspheres

temperature (°C)		4	25	37	45
D_{eff} (× 10^{-11} cm^2/s)	BSA	2.6	4.1	22	30
	dextran (70 kDa)	1.1	2.2	24	31
	dextran (70 kDa) calculated[a]		2.2	2.9	3.5
releasable fraction *p*	BSA	0.45	0.48	0.20	0.11
	dextran (70 kDa)	0.45	0.45	0.15	0.08

[a] Calculated using Stokes- Einstein equation assuming D_{eff} at 4 °C is 1.1 × 10^{-11} cm^2/s.

(Kang & Schwendeman, *Molec. Pharm.*, 2007)

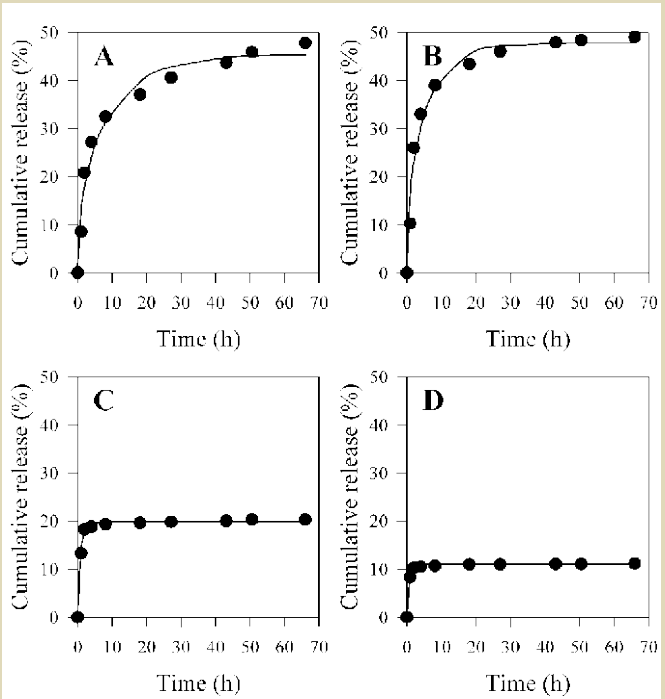


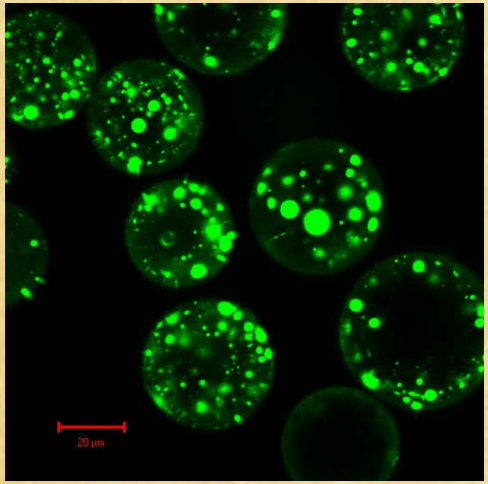
Figure 6. Curve-fitting of BSA release from PLGA- Glu microspheres at 4 (A), 25 (B), 37 (C), and 45 °C (D) by Crank's solution. The experimental data were represented by symbols and the fitted curve was by line. The adjusted coefficient of multiple determination (Ra^2) > 0.97.

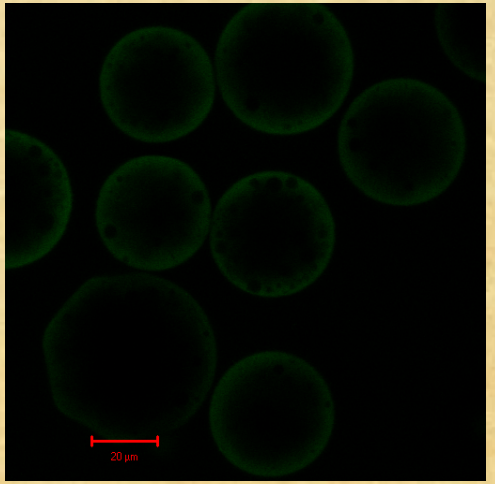
Pores close at 37 °C—stay open at 4 °C

No pre-incubation

Pre-incubation: 4 °C, 2 days

Pre-incubation: 37 °C, 2 days

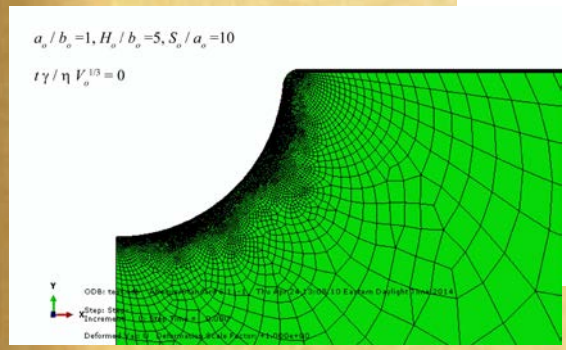


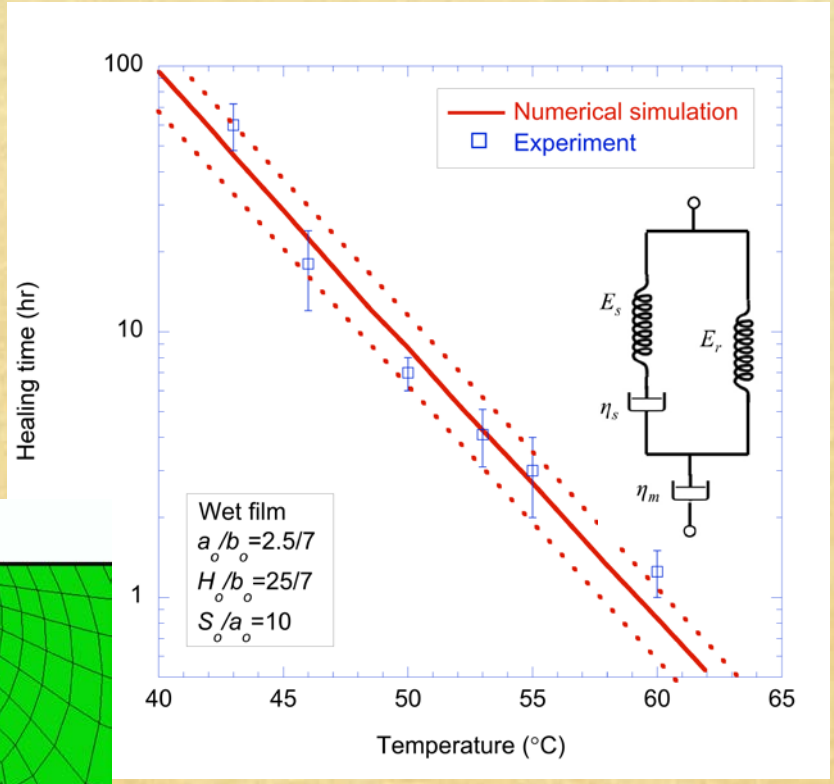


- Monitoring open pores by 12-h uptake of fluorescent dextran

Mol. Pharm., **4**, 104-118 (2007)

Simulated healing times can match well with experimental data



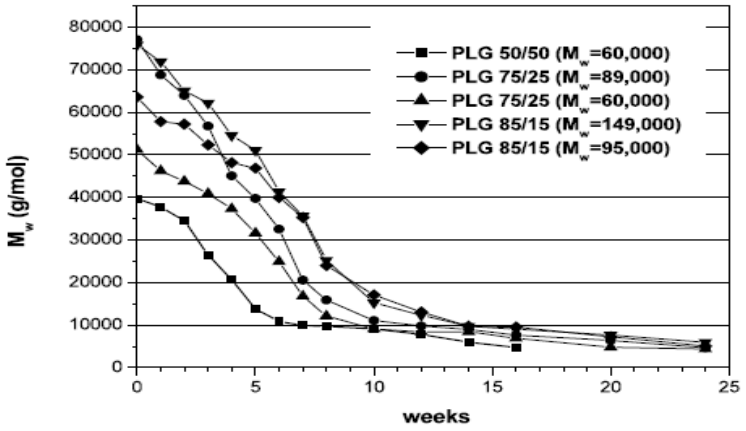


5 μm pores

Left - Courtesy of Jessie Huang and Michael Thouless, UM Mechanical Engineering

Mazzara *et al. J. Control. Rel.*, 171, 172-177 (2013) Huang et al. *J. Control. Rel.*, 206, 20-29 (2015)

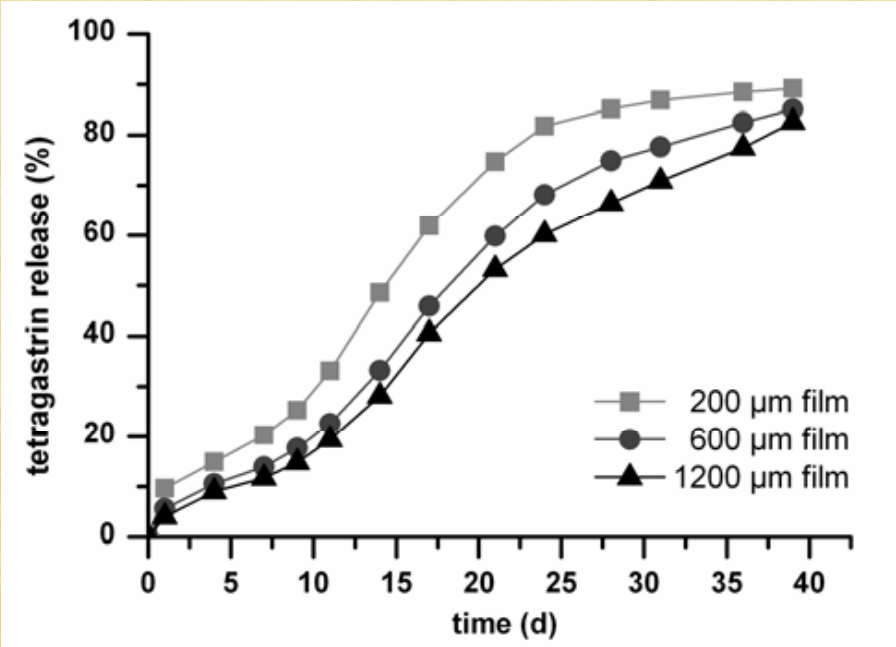
PLGA mass loss occurs at a critical MW providing a means to control drug release



Husmann, M., Schenderlein, S., Luck, M., Lindner, H., and Kleinebudde, P. 2002. Polymer erosion in PLGA microparticles produced by phase separation method. Int. J. Pharm. 242: 277-280.

Common method to accomplish continuous release of peptides and small molecules –

use low molecular weight PLGA



low MW fraction (as a blend or 100%) helps to eliminate induction time of mass loss for sustained erosion-controlled release

PLGA 50/50, Mw = 15.2 kD

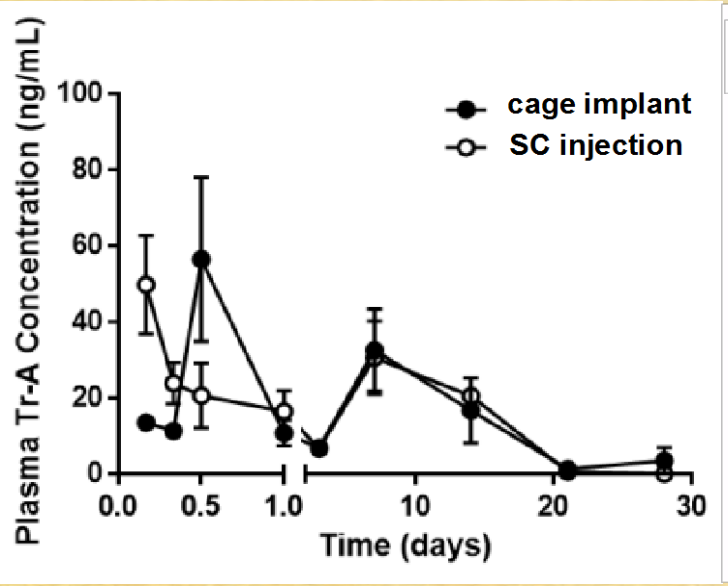
(Hutchinson, EP 058481, 1982)

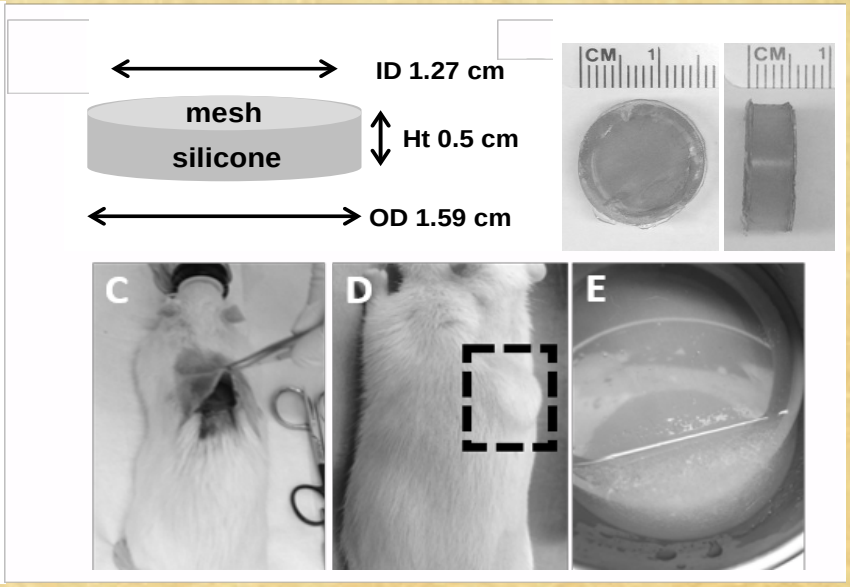
Cage Model for Evaluation of Microsphere Performance

Cages:

- ✧ Surgical grade stainless steel mesh (37 µm opening)
- ✧ Silicone tubing for injection into cage
- ✧ Vulcanize and autoclave
- ✧ Validated by PK vs. SC injection

Amy Doty





PK of the steroid triamcinolone acetonide (Tr-A)/PLGA 50/50 microspheres

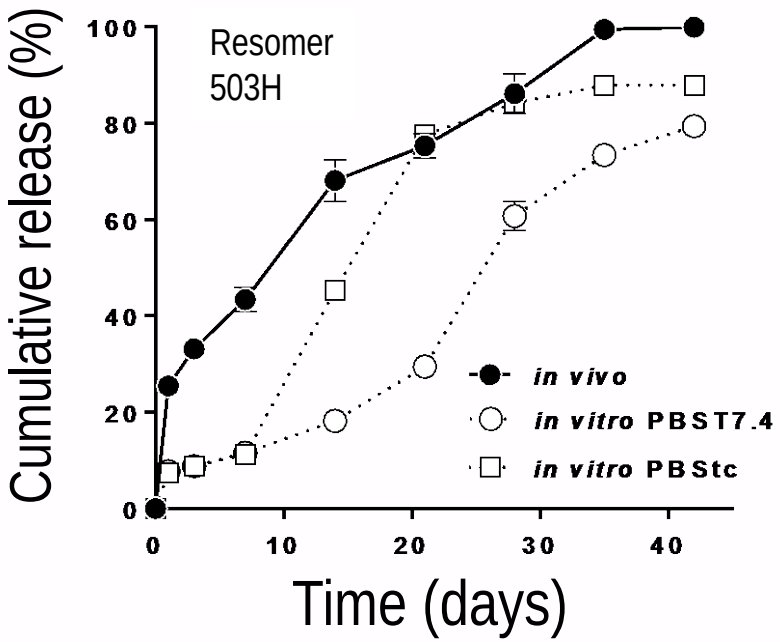
Doty *et al. Biomaterials*, **109**, 88-96 (2016)

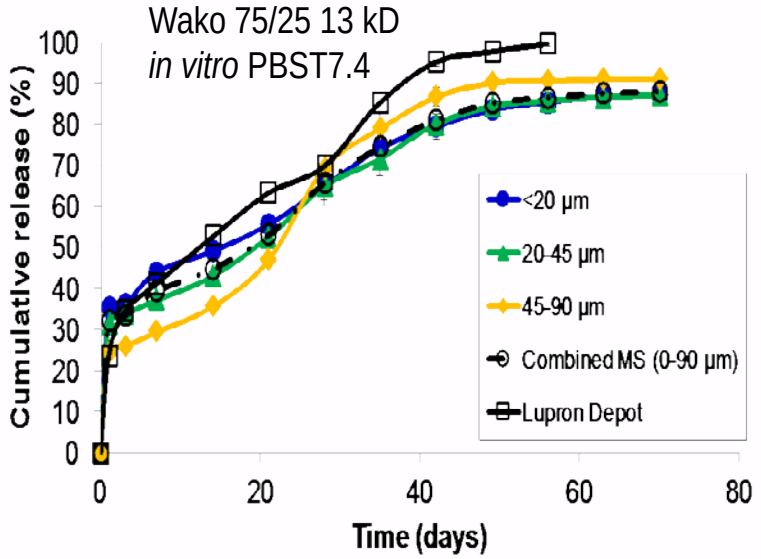
Keiji Hirota

Continuous release of leuprolide from PLGA microspheres

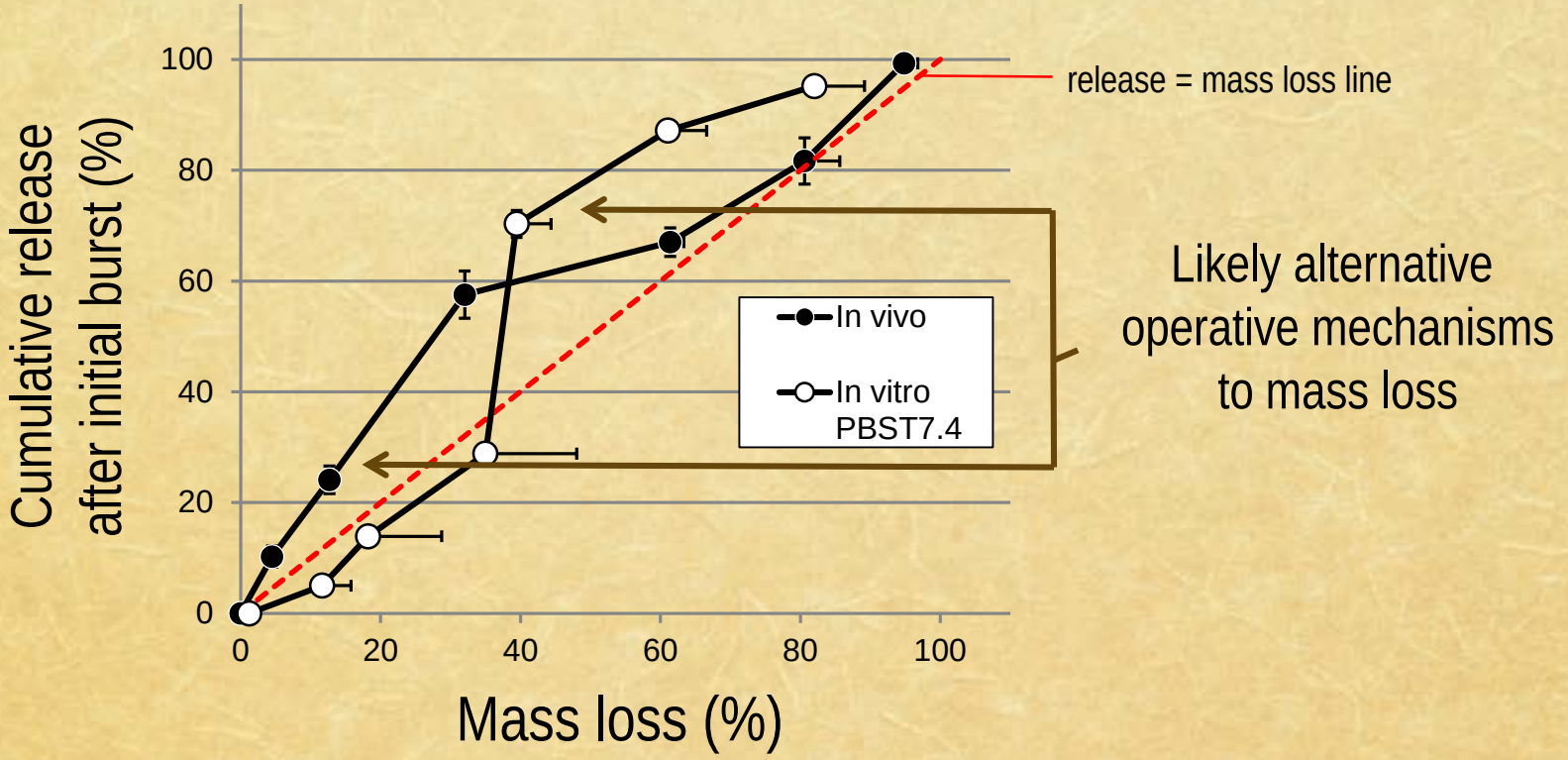
(use low molecular weight PLGA)

Jia Zhou





Comparing mechanistic signatures *in vitro* and *in vivo* for leuprolide from R503H



- *In vitro*: release = mass loss at *late times*
- *In vivo*: release = mass loss at *early times (from cage model)*

1-day absorption of cationic peptides in R502H in neutral pH buffer solution

leuprolide

octreotide

Table 1
Langmuir model fitted parameters (see Eq. (1)) and estimated fraction of acids occupied at maximal sorption.

Peptide	Polymer	K [μM^{-1}]	Γ_0 [μmol/g PLGA]	Γ_{max}[a] [μmol/g PLGA]	Total acids [μmol/g PLGA]	FA[b]
Leuprolide	RG 502H	0.77	1.5	229	185	1.24
Octreotide	RG 502H	1.6	8.5	163	185	0.88
Octreotide	RG 503H	1.2	2.9	81	94	0.86
Octreotide	RG 504H	n.d	n.d	~6[c]	53	n.d

[a] $\Gamma_{max} = \Gamma_0 + \Gamma_1$.
[b] Fraction of acids occupied (Γ_{max}/Total acids).
[c] Γ_{max} estimated visually from isotherm.

(Sophocleous *et al.*, *J. Cont. Rel.* 2013)

Proposed desorption mechanism for in vitro leuprolide release from 503H

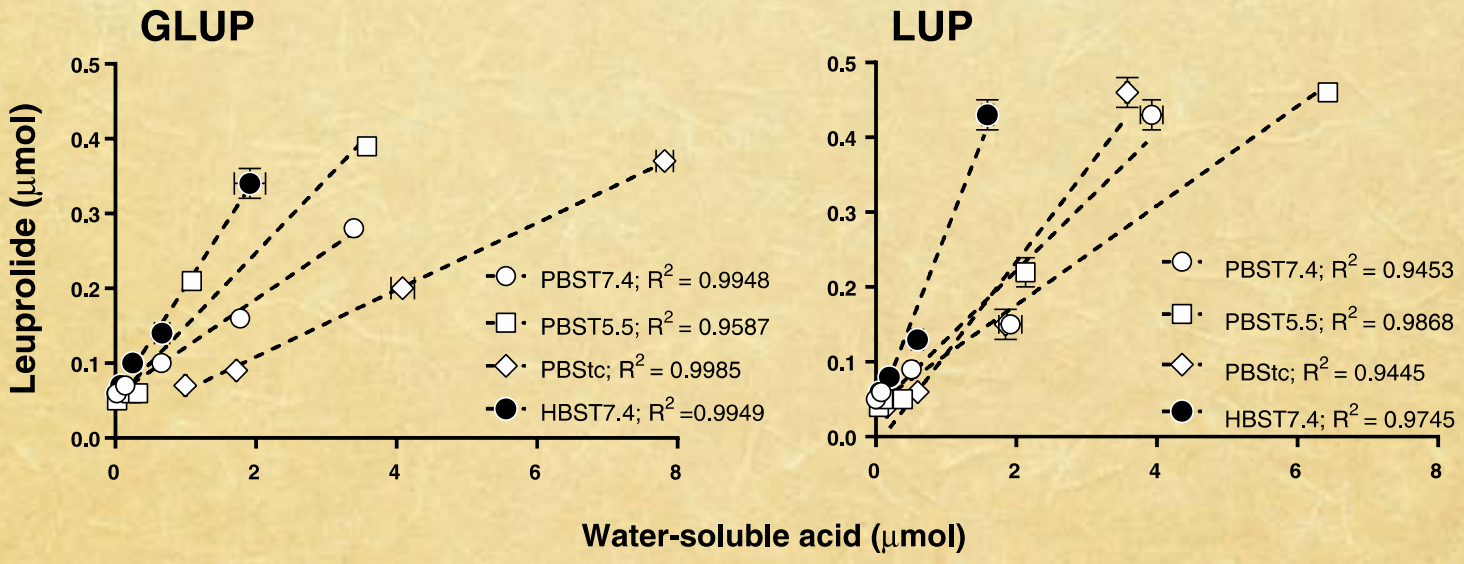
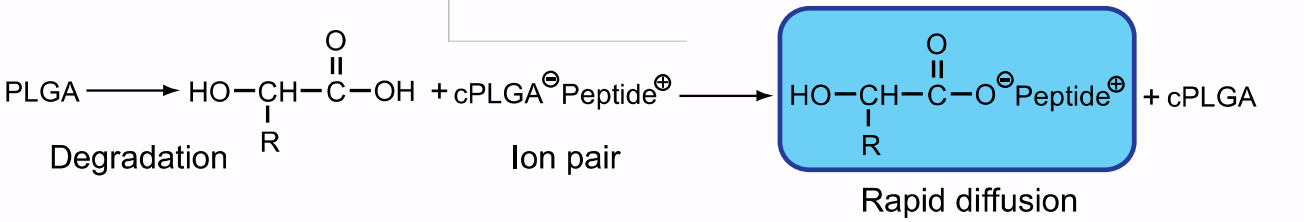


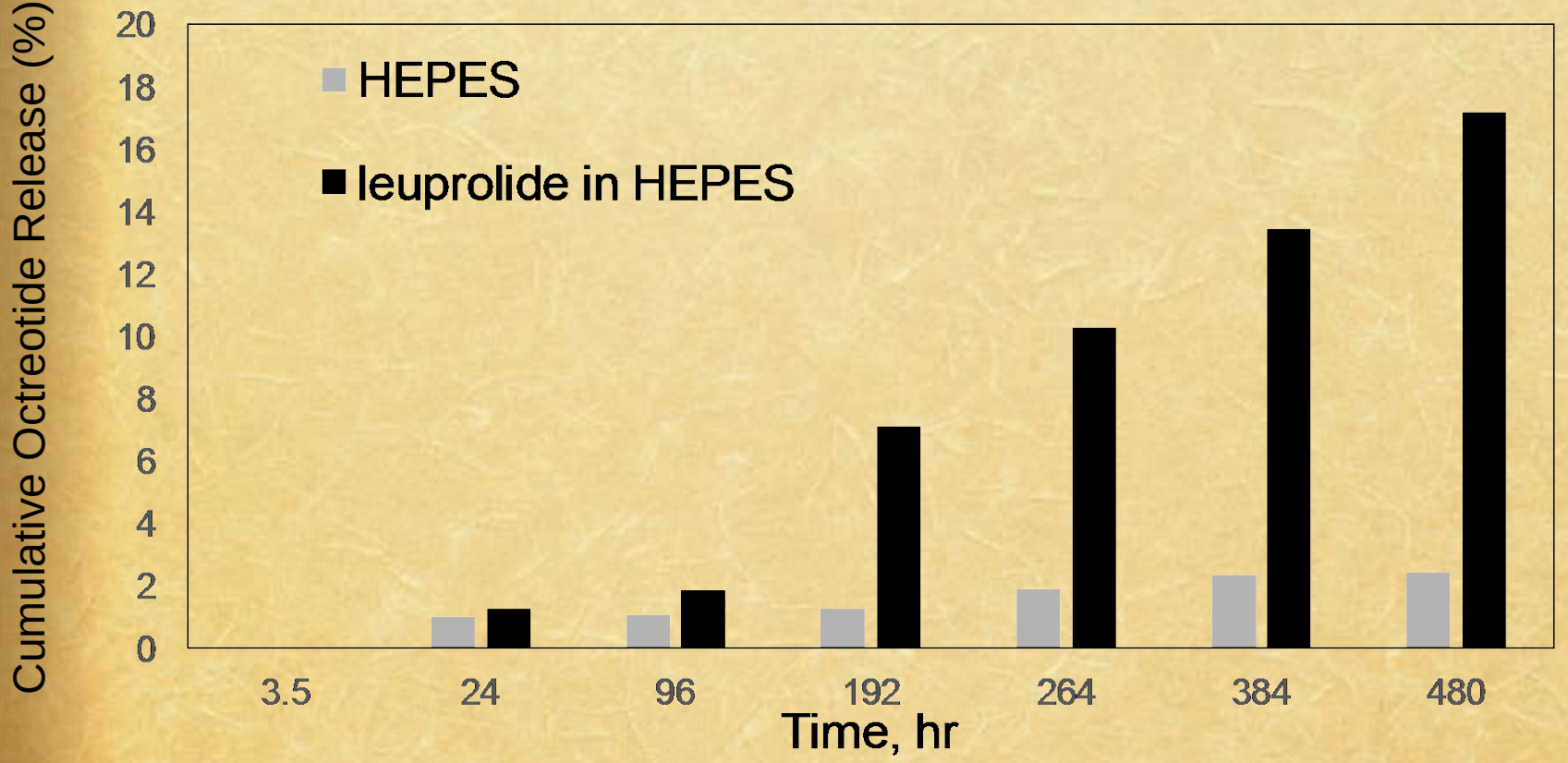
Fig. 8. Correlation of leuprolide release with water soluble acid levels in incubation media, PBST7.4 (○), PBST5.5 (□), PBStc (◇), and HBST7.4 (●) at days 1, 3, 7, 14, and 21 except for PBStc which was examined until day 14. Curve fitting results for each sample are indicated by dotted lines with R^2 values shown in legend notes. Each plot represents mean ± S.E.M (n = 3).



(Hirota *et al*, *J. Cont. Rel.*, 2016)

Release of octreotide from SLAR accelerated in the presence of leuprolide

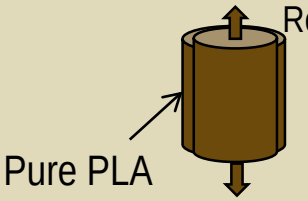
Avital Beig



Case for water-mediated processes controlling drug release

Rationale:

- If diffusion slower than osmotic pumping (e.g., by increasing cylindrical length) then strong dependence on external π



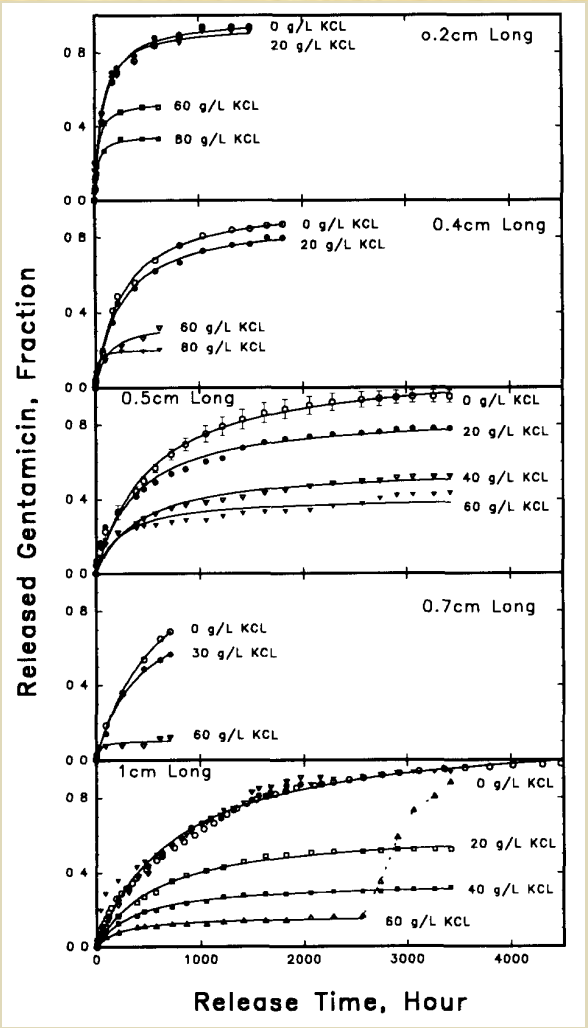
30% load

d Release 31 (1994) 129–144 1

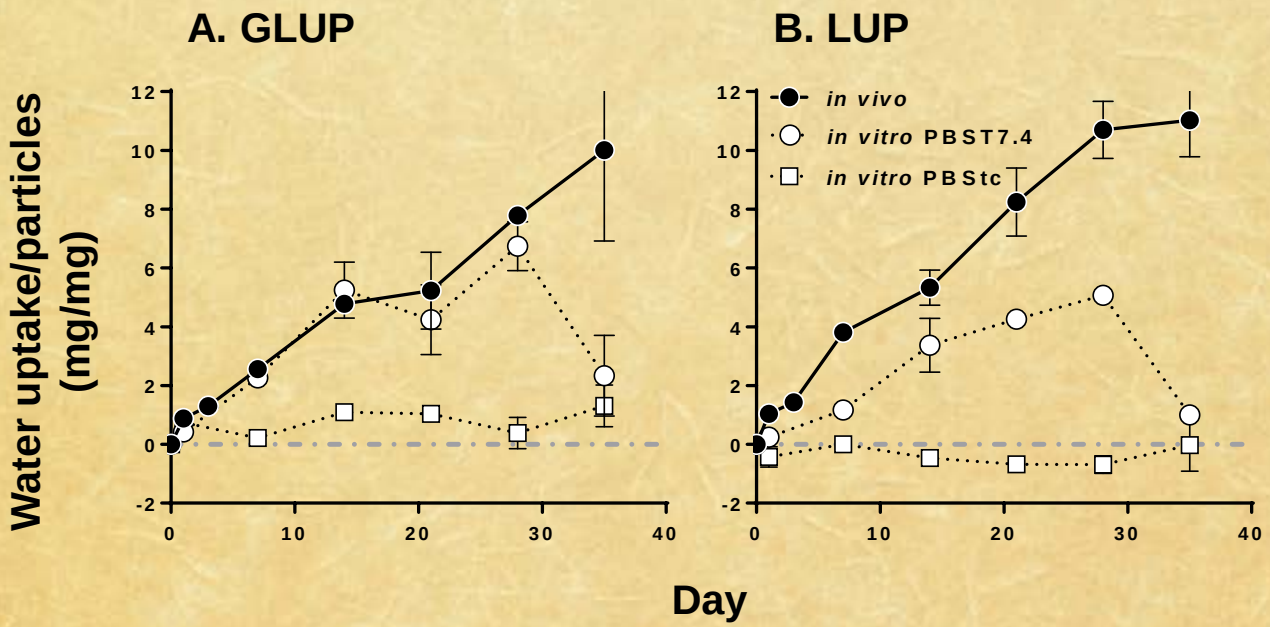
pumping. After the osmotic pumping effect, drug di
fusion through the capsule openings will become don
nant. Since water can still permeate through polym

PLA, Mv = 32.6 kD

(Zhang *et al.*, *J. Cont. Rel.*, 1994)

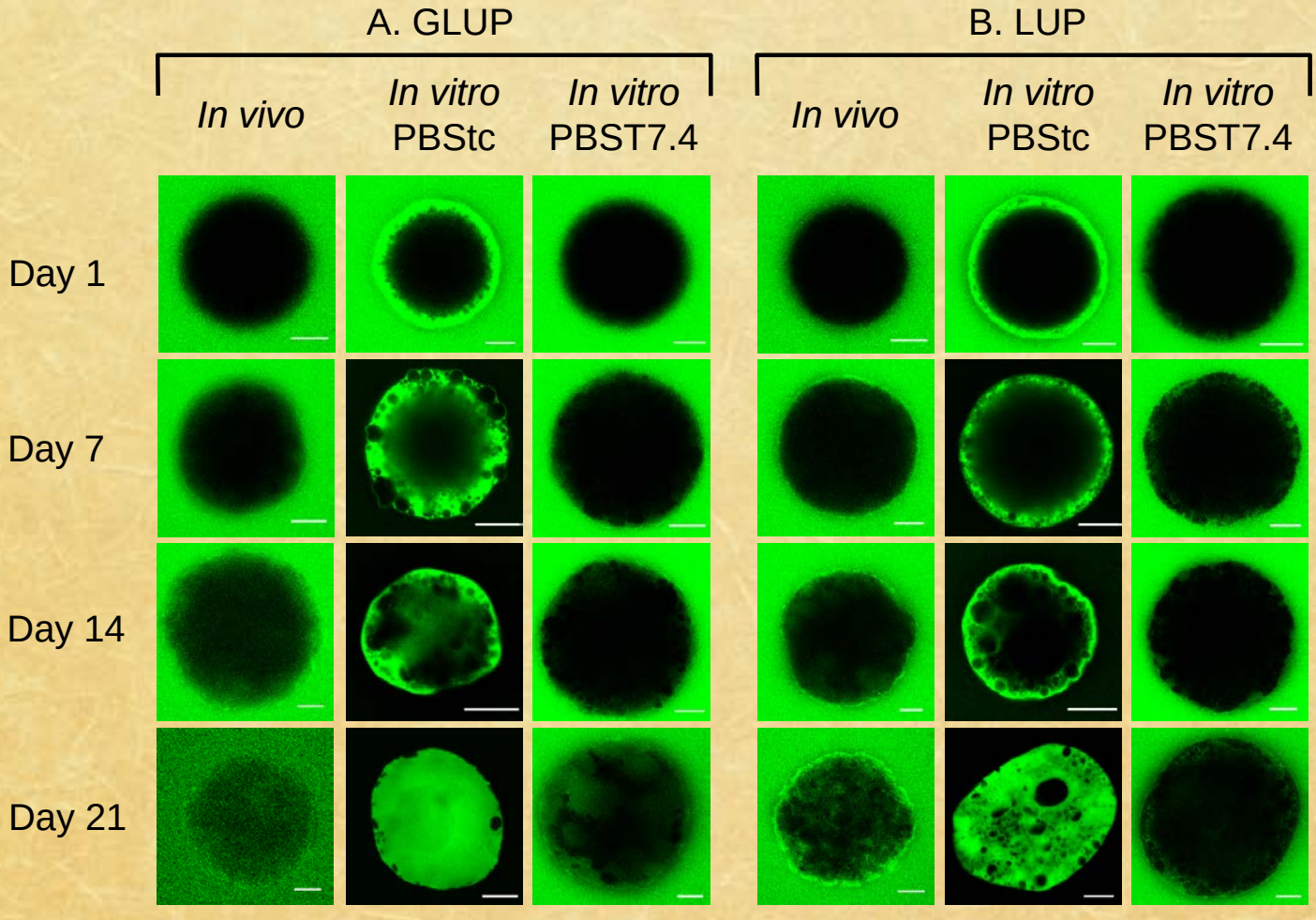


Water uptake continuous and extensive for leuprolide release from R503H



GLUP = + gelatin with leuprolide
LUP = – gelatin with leuprolide

Microstructural changes detected by confocal mapping of bodipy uptake in R503H

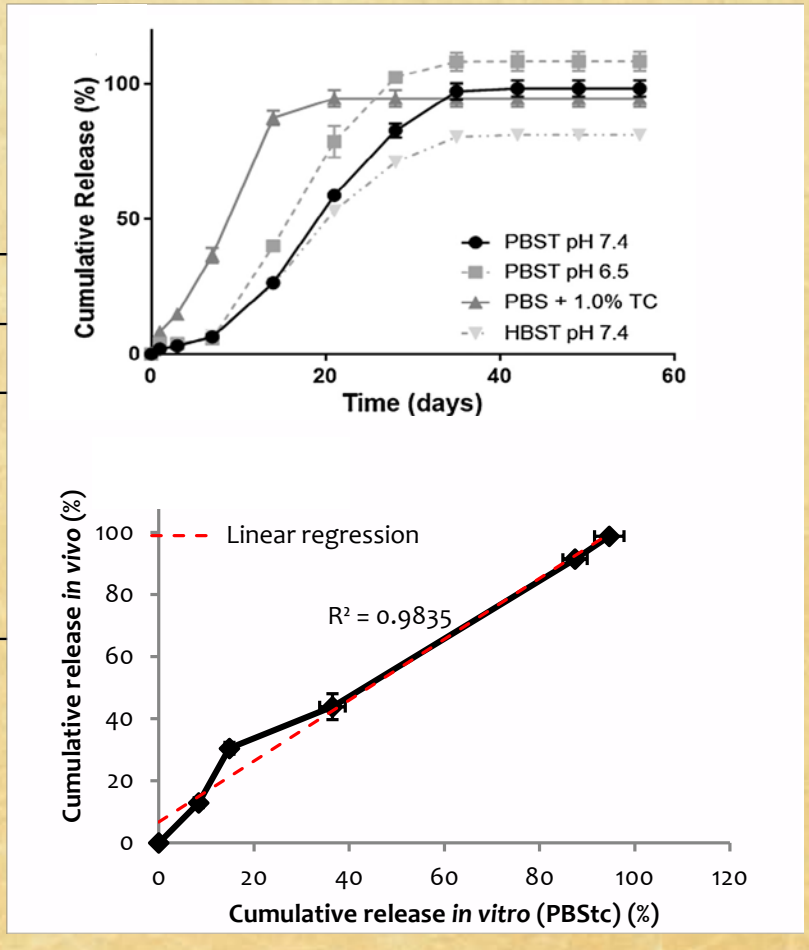


Characterizing release mechanisms and identifying biorelevant in vitro release media

Compare time-scales to understand release mechanisms (acid-capped PLGA 50/50)

Characteristic times (in days) of release and erosion

	Tr-A_1	
	in vitro	***in vivo***
$t_{50,release}$	19.0 ± 0.4	7.9 ± 0.8
$t_{50,erosion}$	25 ± 8	11 ± 1
$t_{50,release} / t_{50,erosion}$	**0.77**	**0.72**



Doty *et al.*, *Eur. J. Pharm. Biopharm*, 113, 24-33 (2017)

Summary

- Several mechanisms contribute to the release of drugs from PLGA microspheres *in vitro* and *in vivo*
- In addition to erosion, diffusion, and water-mediated processes, pore healing, drug-polymer interactions, and other dynamic microstructural changes to the polymer may affect the release mechanism
- Development of a cage model has provided utility to facilitate mechanistic analysis of *in vivo* release by recovery of the microspheres
- Further refinement of methods to evaluate mechanistic effects *in vitro* and *in vivo* for existing products would appear useful to compare incoming generic drug products
- Continued focus on mechanistic research and related opportunities may reduce barriers to new PLGA microsphere products

Acknowledgements

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Tianhong Zhou *
Gaozhong Zhu *
Juan Wang *
Wenlei Jiang *
Julia Marinina *
Chengji Cui *
Jichao Kang *
Longsheng Lai *
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Brittany Bailey *
Karthik Pisupati *
Amy Doty *
Max Mazzara *
Kellisa Hansen *

* former lab members

Financial support

NIH, Novartis, Merck, Dow, Coulter, FDA, FHI360

Collaborators

James Moon (U of M)
Susan R. Mallery (OSU)
John F. Carpenter (Univ Colorado, Denver)
Anna Schwendeman (U of M)
Ji-Xin Cheng (Purdue)
Michael Thouless (U of M)
Mark Prausnitz (Georgia Tech)
Mark Meyerhoff (U of M)
Don Schwendeman (RPI)
Dave Antonetti (U of M)

Yan Wang (FDA)
Stephanie Choi (FDA)
Jeff Jiang (FDA)

Acknowledgements