Nanoparticles for Cancer Detection and Treatment

Wole Soboyejo

US/Africa Materials Institute (USAMI)
Princeton Institute of Science and Technology of Materials (PRISM)
and
Department of Mechanical and Aerospace Engineering
Princeton University

The Structure of USAMI

- NSF funded virtual institute operated from Princeton
- Princeton setting **(usami.princeton.edu)** includes
  - PRISM (Jim Sturm, Kim Hegelbach, BarbaraVarga, Dan Steinberg, Shannon Swilley)
  - MAE (Maureen Hickey, Jenny Kokini, Vocaturo, Whitehead)
  - Institute for Advanced Studies (Arlen Hastings & Phillip Griffiths)
  - Carl Fields Center/Third World Center (Makeba Clay)
- Administrative/technical staff members
  - Dale Grieb (Administrator)
  - Laura Ceritto (Administrative Assistant)
  - Betty Adam (Secretary)
  - Eric Paul (Web Support)

Princeton Scientists & Students

- Faculty – Scherer, Prud'homme, Schwartz, Ong, Register, Arnold, Young, Haataja
- Senior Scientists – Yao, Steinberg, Vocaturo
- Graduate Students – Zong, Meng, Yang, Chen, Akogwu, Fan, Oni, Chi
- Undergraduate Students – Theriau, Pawlowski, Sud, Rogers, Davis, Cohen, Huang, Li, Chaco, McFarland, Farias, Mandecki

U.S. Institutions/Collaborators

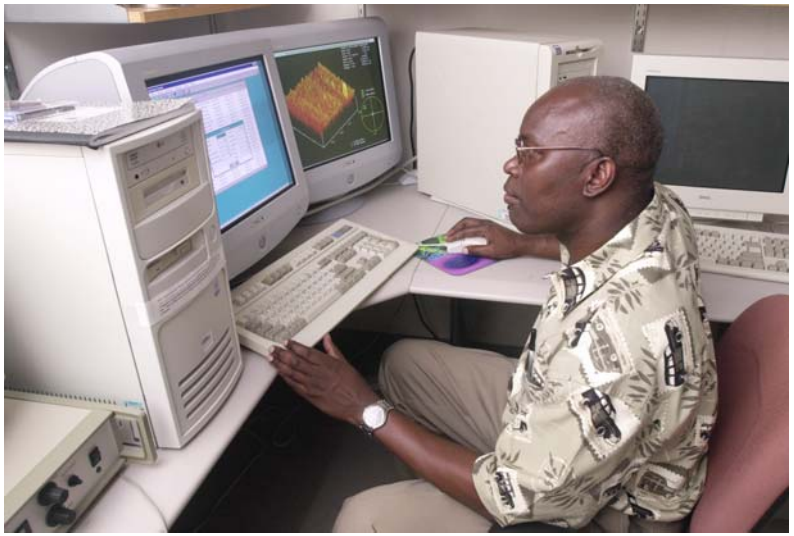
• Ohio State University	Soboyejo
• Harvard University	Vlassak, Hutchinson
• Howard	Mitchell
• Brown	Needleman
• Yale University	Ramirez
• Columbia	Kysar
• Duke	Warren
• University of Michigan	Forrest
• LSU/CAMD	Hormes, Kumar
• UIUC	Paulino
• Rutgers University	Cuitino
• Sandia	Buchheit, Boyce
• Pennington Biomedical	Leuschner

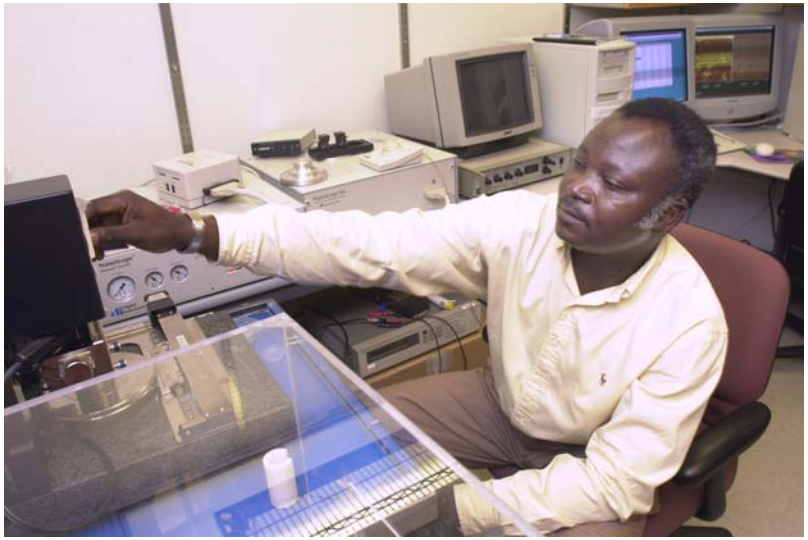
The Americas Program

- A complementary initiative of the NSF and other American agencies (Holmer Savastano et al.)
- Emerging U.S./South America collaboration in infrastructure materials
  - Focus on natural fiber reinforcement
  - Supports 5 South American researchers (per year) to visit the U.S. for 9 weeks per year
  - Current collaborators from Brazil, Argentina and Guadeloupe

International Collaborators

- Eastern Africa – Kenya, Uganda, Ethiopia, Tanzania
- Northern Africa – Egypt, Tunisia, Algeria, Morocco
- Western Africa – Senegal, Nigeria, Ghana, Burkina Faso
- Central Africa – Rwanda
- Southern Africa – Zambia, Botswana, Mozambique, South Africa, Namibia
- South America - Brazil
- Caribbean - Guadeloupe





Approach of The IMI Program

- 16 international researchers visit the U.S. to work with U.S. collaborators for 9 weeks
- They then return to their home countries to continue their work
- Many return over the next few years to do a complete piece of work
- A systems based approach - must work in one of the four areas of focus

The Areas of Focus of the IMI

- Advanced Materials/Small Structures
  - MEMS/thin films and organic electronics
  - Biomaterials
- Materials for Societal Development
  - Materials for affordable infrastructure
  - Thermostructural materials

Acknowledgments

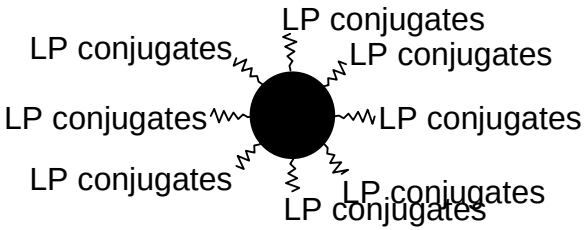
- Post-docs – Jikou Zhou, Craig Steeves
- Students - C. Milburn, S. Mwenifumbo, R. Weissbard, L. Ionescu, L. Hayward, O. Bravo, J. Meng, C. Thieraux
- Colleagues - Challa Kumar (CAMD), Josef Hormes (CAMD), Carolla Leuschner (Pennington), Jeff Schwartz (Princeton), Julie Young (Princeton), Aboubaker Beye (Cheikh Anta Diop), Tom Otiti (Makerere), Warren Warren (Princeton), Bob Prud'homme (Princeton)
- MRI Technician - Silvia Cenzano (Princeton)
- Financial Support - Carmen Huber (NSF) and Princeton University

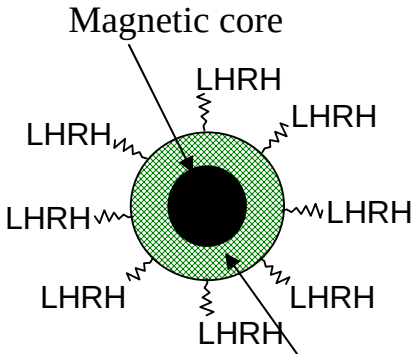
Background and Introduction

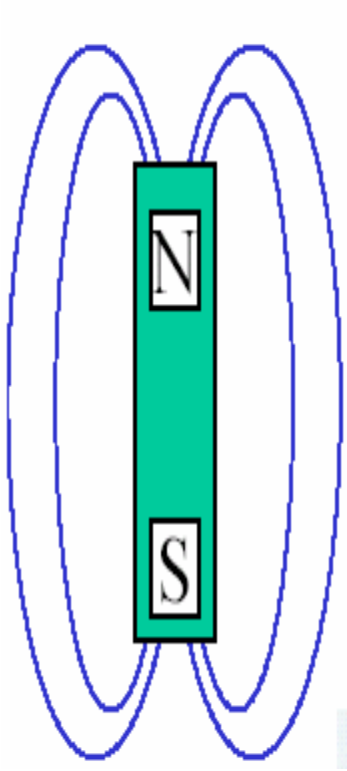
- Several people could benefit from implantable or injectable systems for disease detection and treatment
- This class examines nanotechnology for disease detection and treatment (with a focus on cancer)
  - Breast cancer
  - Prostate cancer

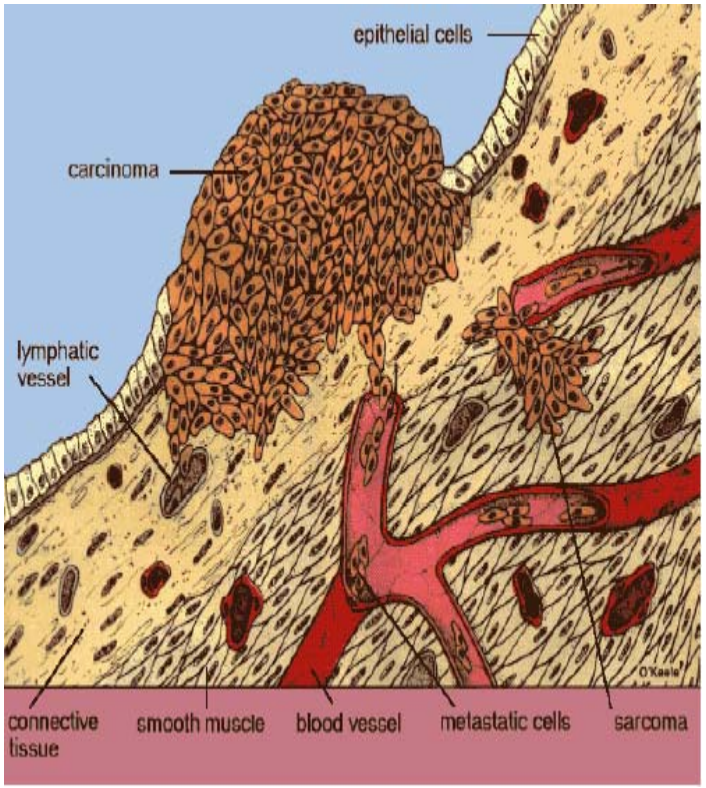
Our Approach to Early Cancer Detection and Treatment!

A novel use of magnetic fields and magnetic particles to deliver therapeutic drugs at the desired time in the correct dosage to the correct site in the human body.

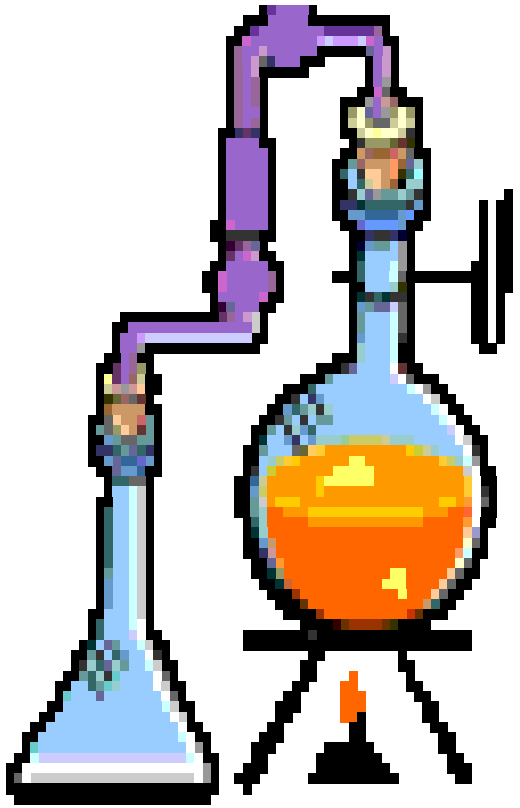








Wet Chemical Synthesis of Nano-particles

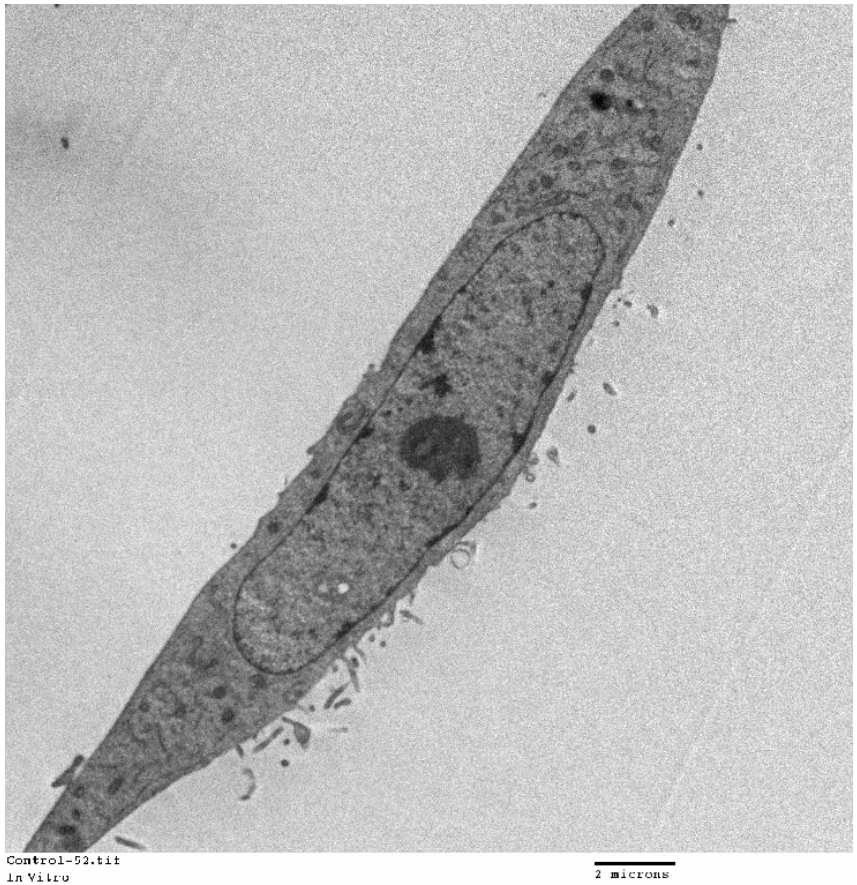


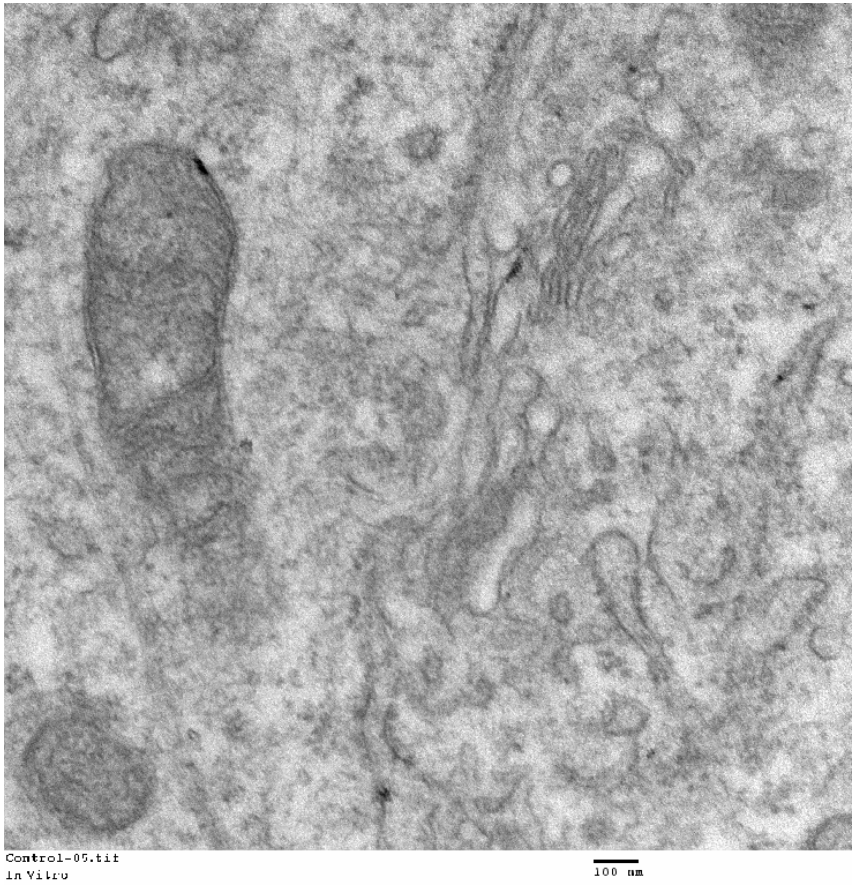
- Metallic, polymeric and metal-polymer Nano-particles using bottom-up approaches
- Novel Micro reactor technology for scale-up and controlled synthesis
- Synchrotron radiation based X-ray absorption Spectroscopic characterization
- Capability to attach bio-molecules

In-Vitro Experiments

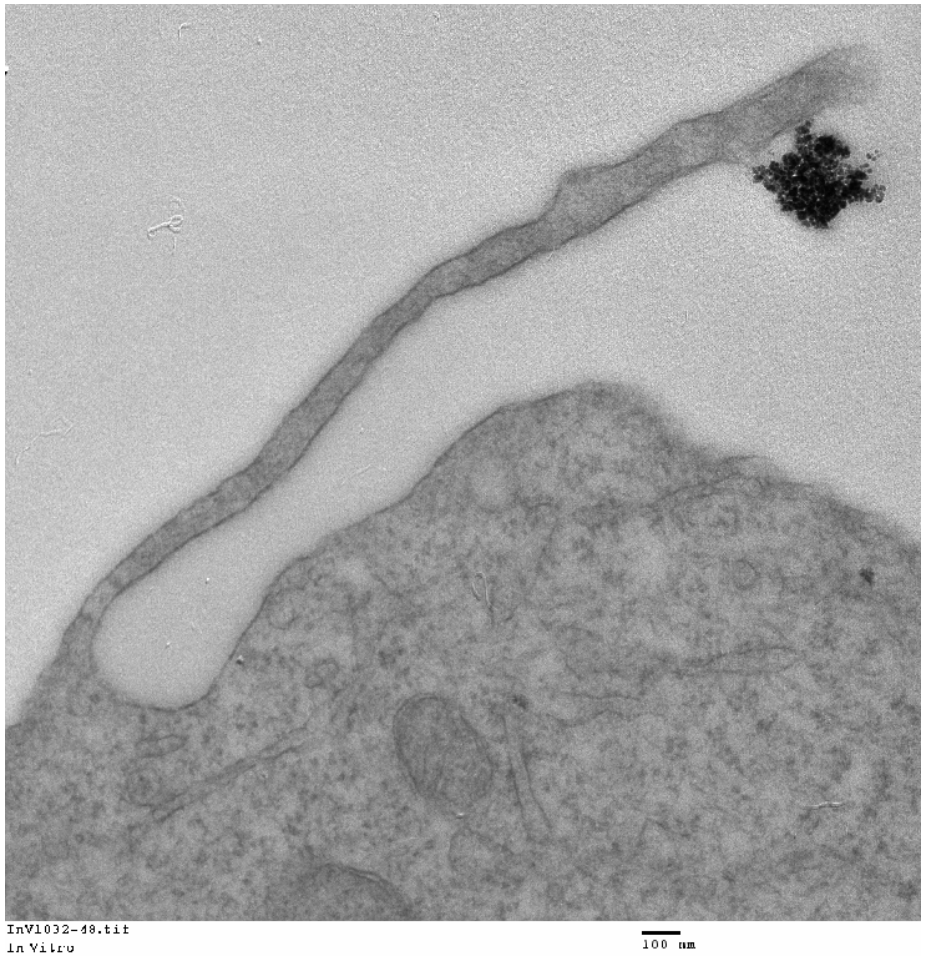
- Studied attachment of nano-particles in cell culture experiments
- Studied effects of temperature and time
- Imaging done using TEM after fixing
- Studies conducted on breast cancer cells with LHRH receptors
  - Unconjugated nanoparticles
  - LHRH-coated nanoparticles

TEM Images of Breast Cancer Cells (Control)



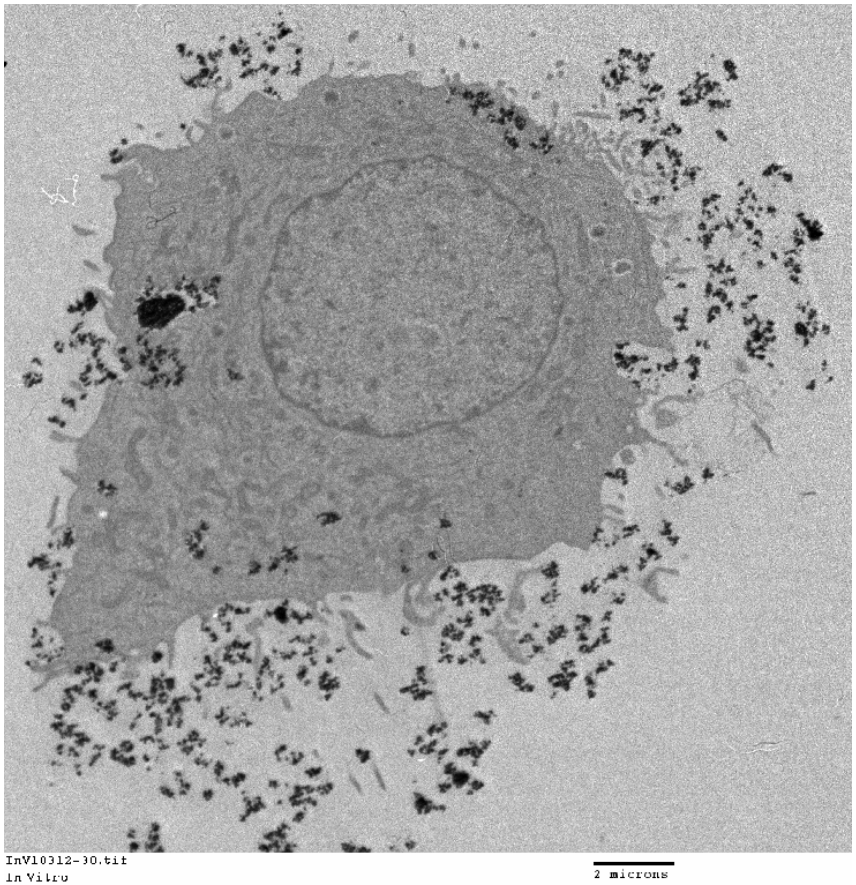


SPION Uptake - 37 C for 30 Minutes

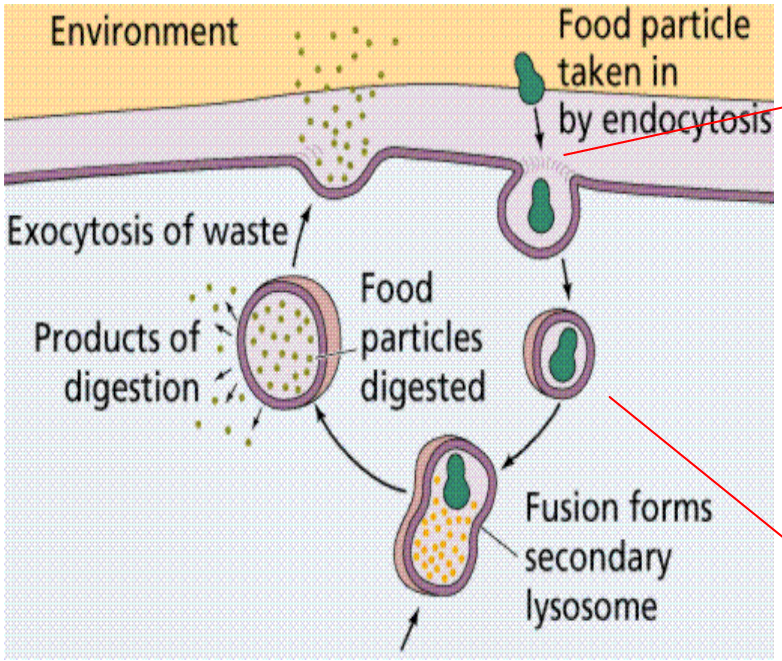


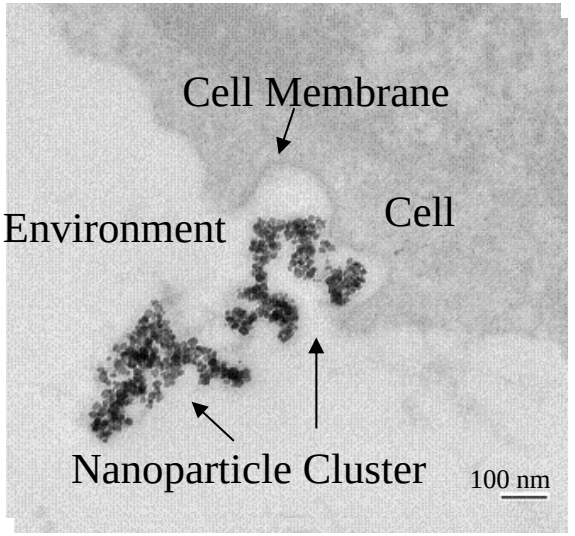
LHRH-SPION Uptake - 37 C for 3 Hours

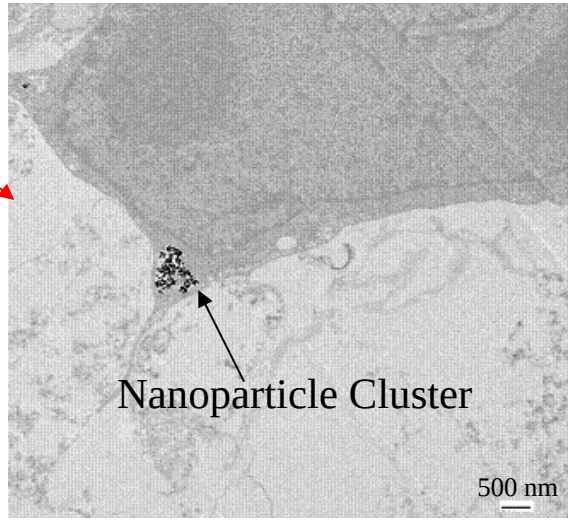
- MNPs-LHRH, 37 C, 3 Hr
- Note encryption process by which cells attach
- Engulfed cells carried within the cell
- Excreted or egested within 30 days



Mechanism of Nanoparticles Uptake







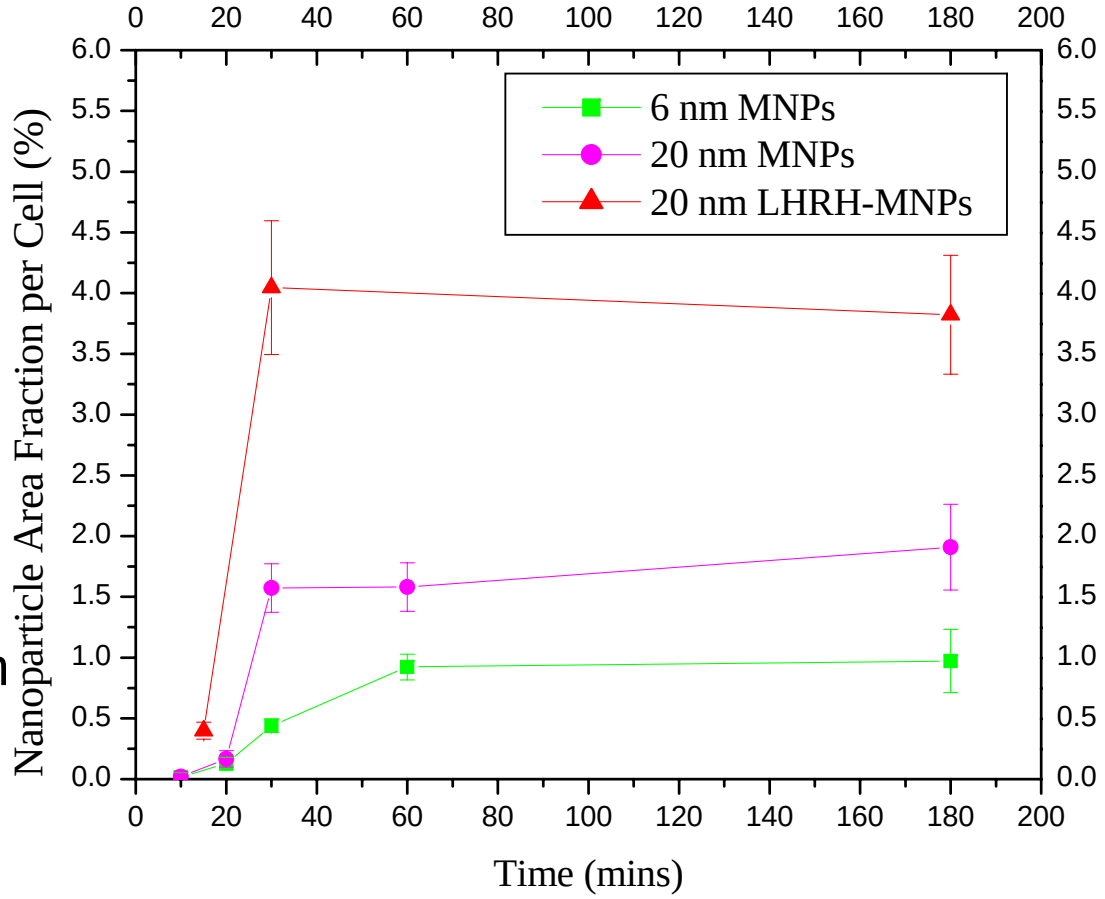
- Active : ATP provides the energy for cell endocytosing medium or particles
- Passive (Huajian Gao, 2005): bonding/adhesion energy overcomes the elastic energy of membrane curvature

(www.emc.maricopa.edu/faculty/farabee/BIOBK/endocytosis

Nanoparticles Uptake Curve

Nanoparticle uptake versus Time:

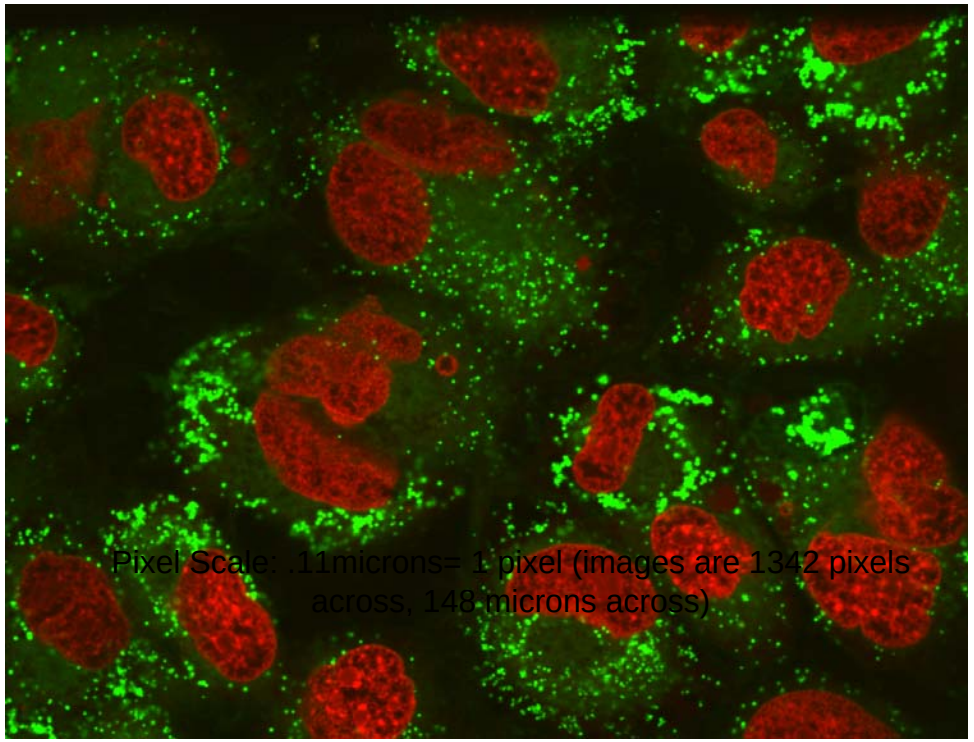
- Cell intake of nanoparticles reaches a peak value after a while and fluctuates around this value (longer duration experiment needed)
- Cell take more LHRH coated MNPs than uncoated MNPs



Confocal Image of Nanoparticals Uptake

- Confocal Image
  - ❖Red – Nucleus
  - ❖Green Spots
    - – Nanoparticles

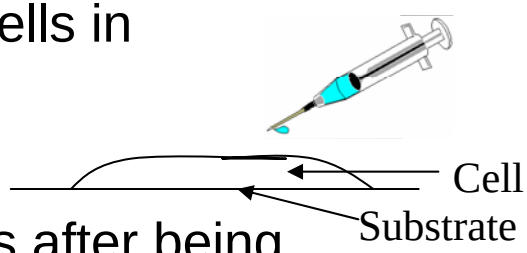
TEM	Confocal
Look inside of cells	Look inside of cells
Long time preparation (2 w)	Short-time preparation (2 h)
High resolution	Low resolution



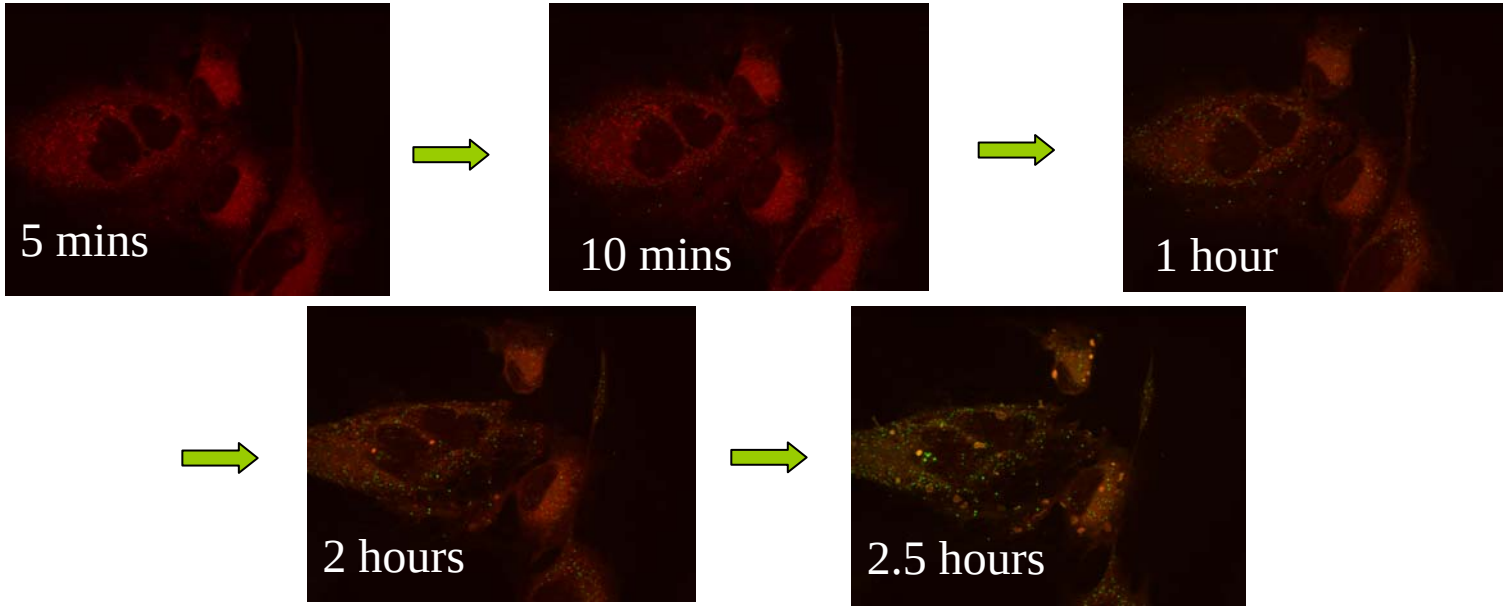
Confocal Movie of Cell Intake

- Experimental process

  ① Cells grow in round flask overnight ⇒ ② Stain cells with Cell Tracker Orange ⇒ (Color Cytoplasm) ③ Settle cells in confocal microscope and add nanoparticles ⇒ ④ Observation

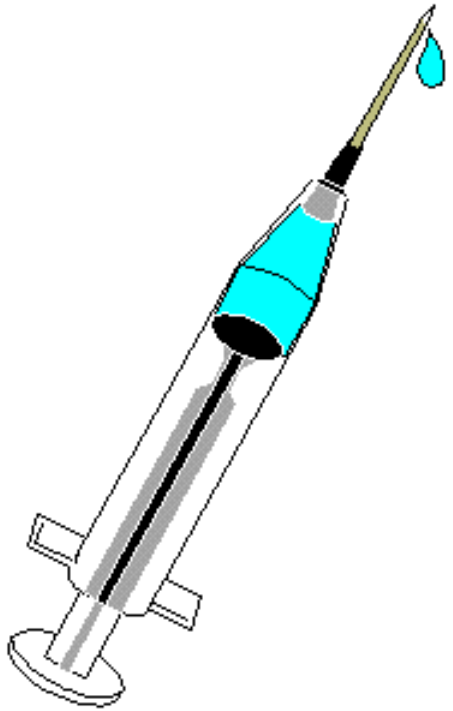


- Nanoparticles start to enter cell at about 10 mins after being added into the flask



In-Vivo Experiments

- Mice injected in 4 different ways:
  1. LHRH nanoparticles
  2. saline solution
  3. nanoparticles
  4. LHRH nanoparticles but with mice that do not contain breast tumor



Materials Characterization of Organs (TEM and Histology)

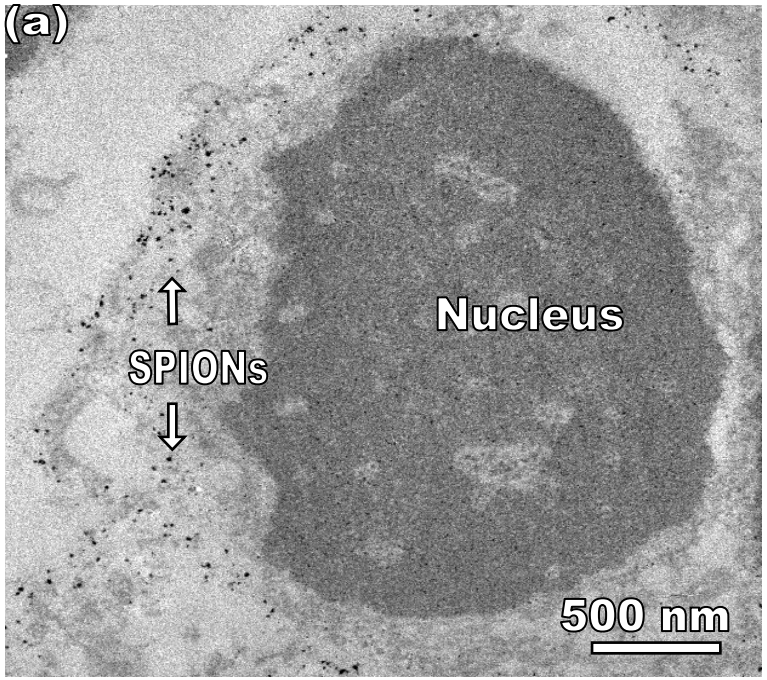
Organs obtained:

- breast or prostate tumor
- Kidney
- Lung
- Liver

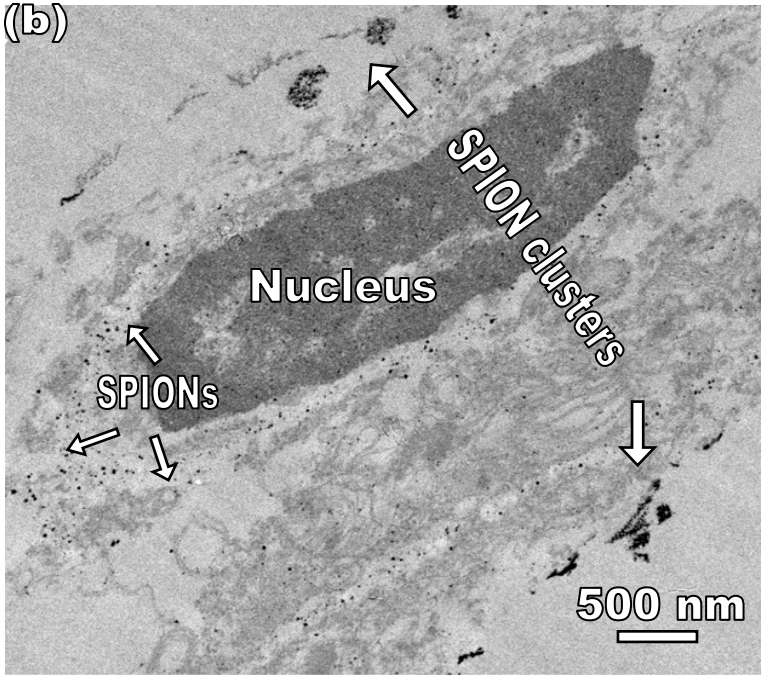
Ensure that the nanoparticles do not accumulate in other major organs.

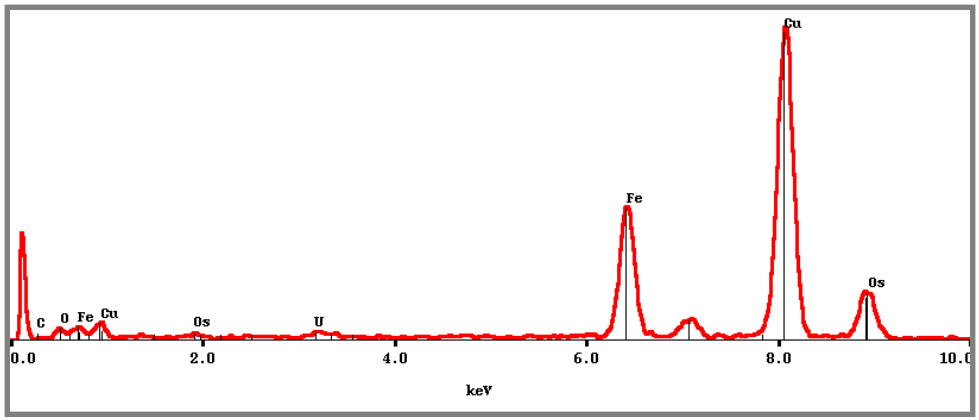
SPION/SPION-LHRH in Breast Tumor

SPION in Tumor



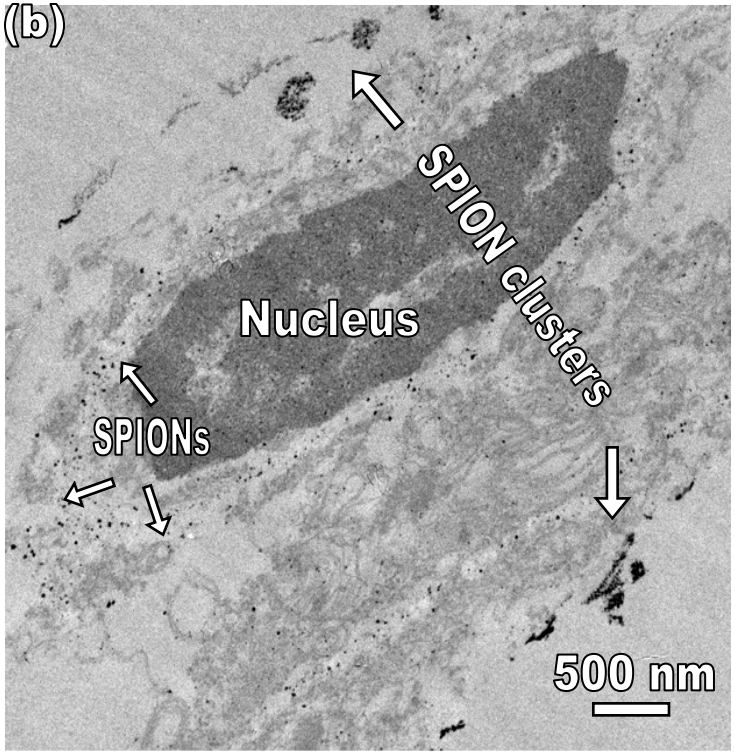
LHRH-SPION in Tumor



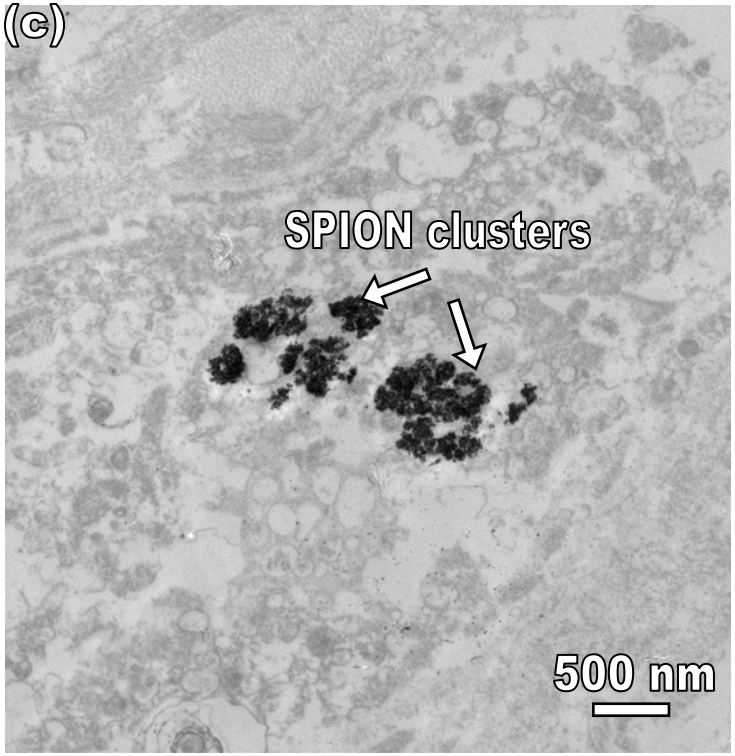


SPION/SPION-LHRH in Breast Tumor

LHRH-SPION in Tumor

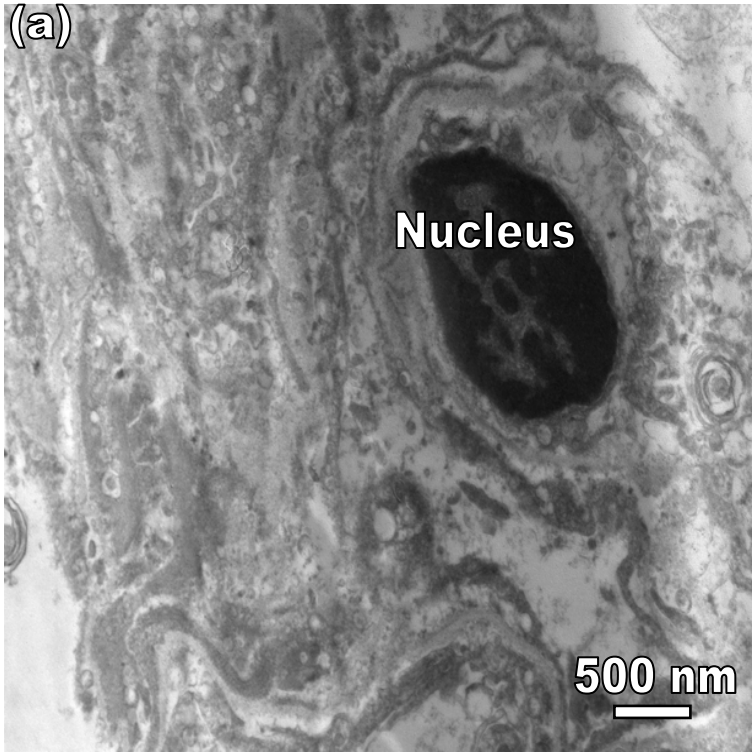


LHRH-SPION in Tumor

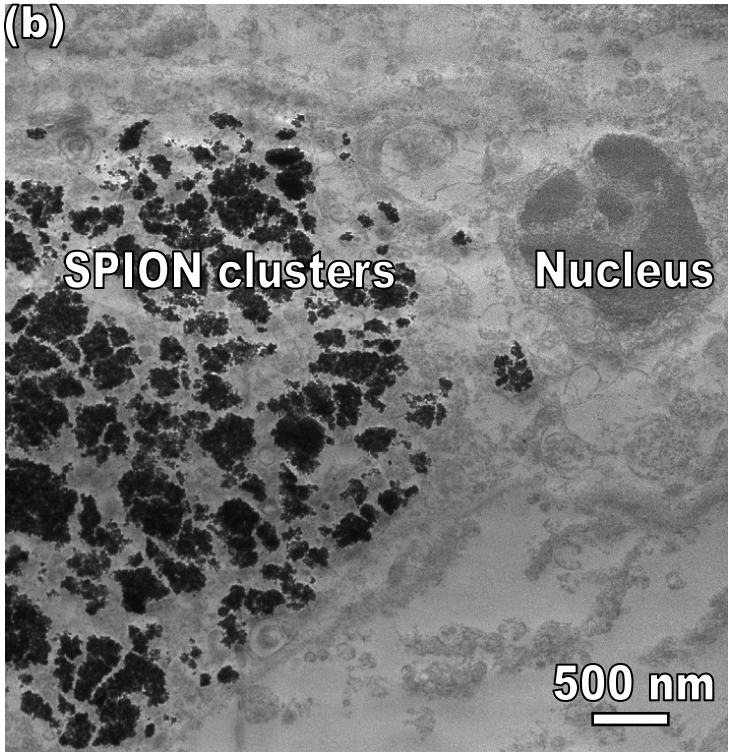


SPION in Lung

SPION in Lung

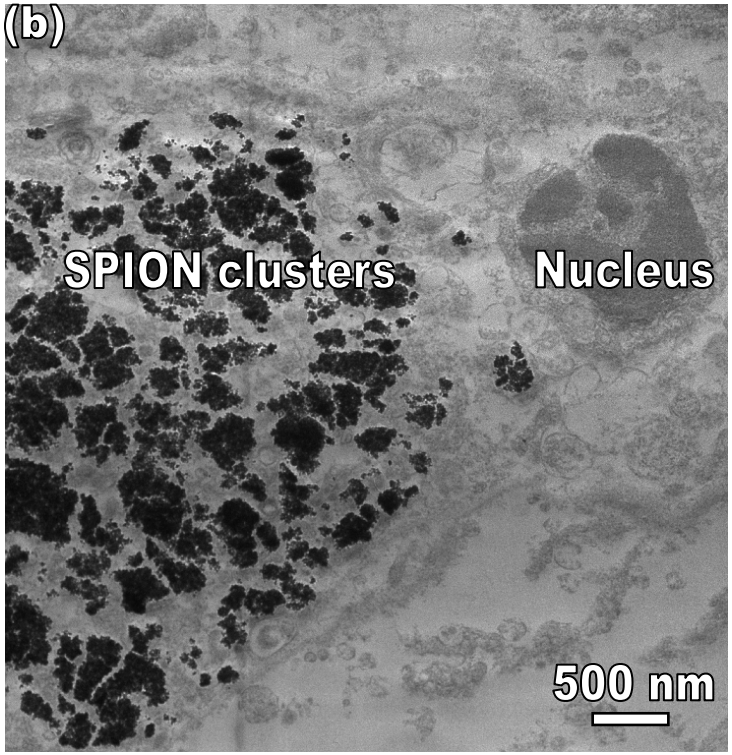


LHRH-SPION in Lung

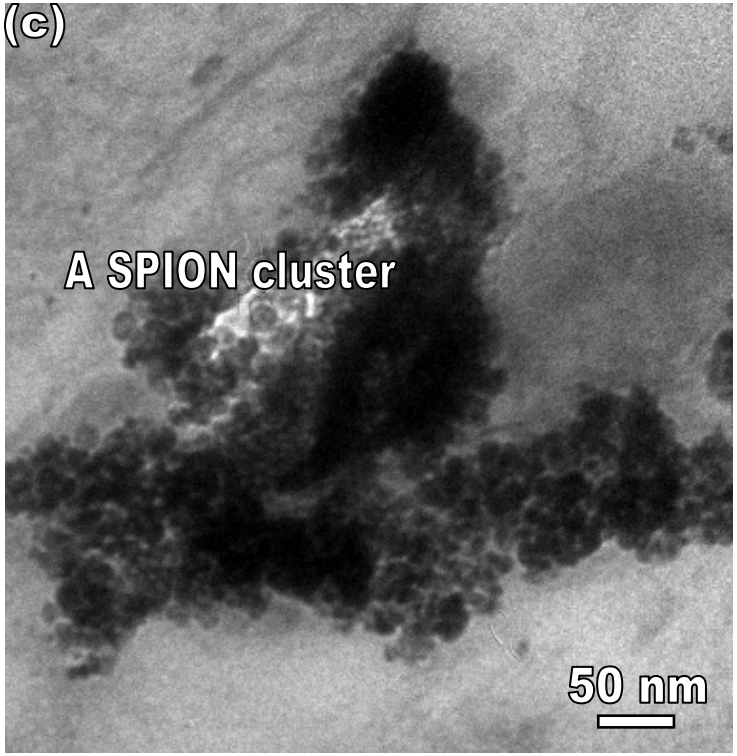


SPION in Lung

LHRH-SPION in Lung

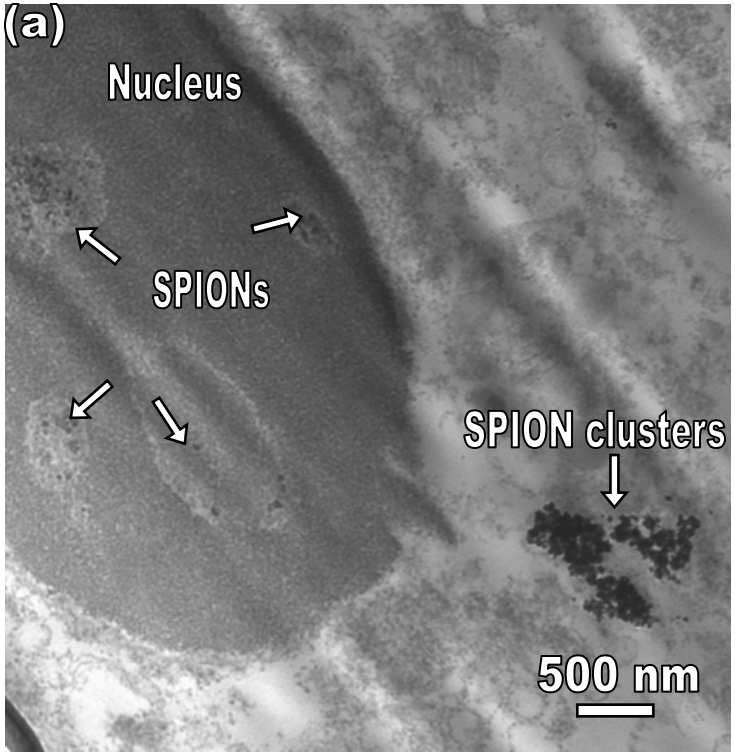


LHRH-SPION in Lung

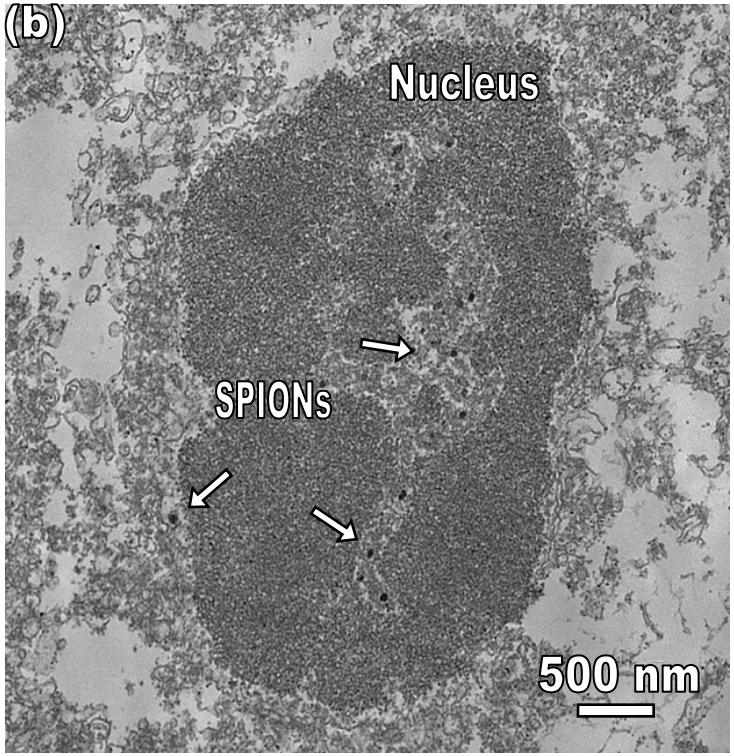


SPION/SPION-LHRH in Liver

SPION in Liver

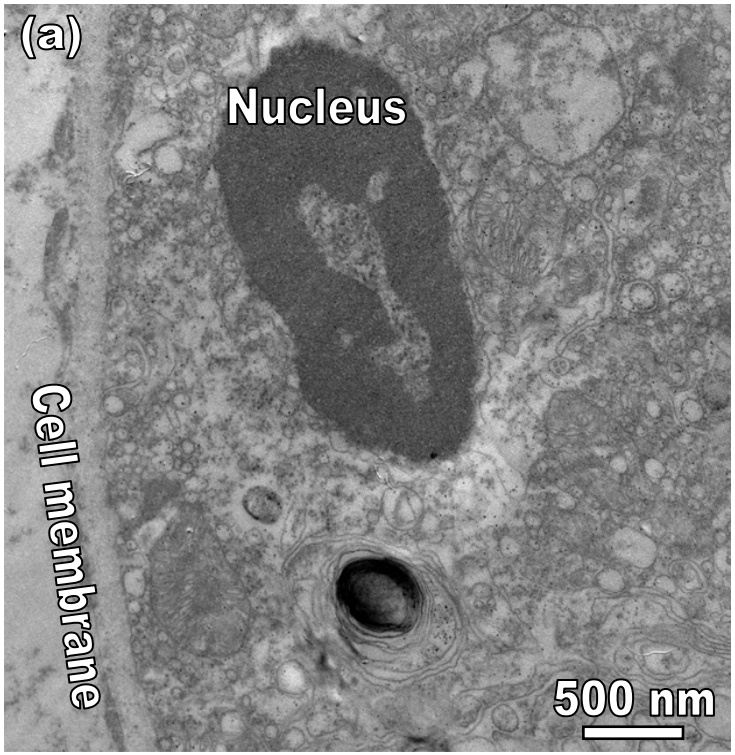


LHRH-SPION in Liver

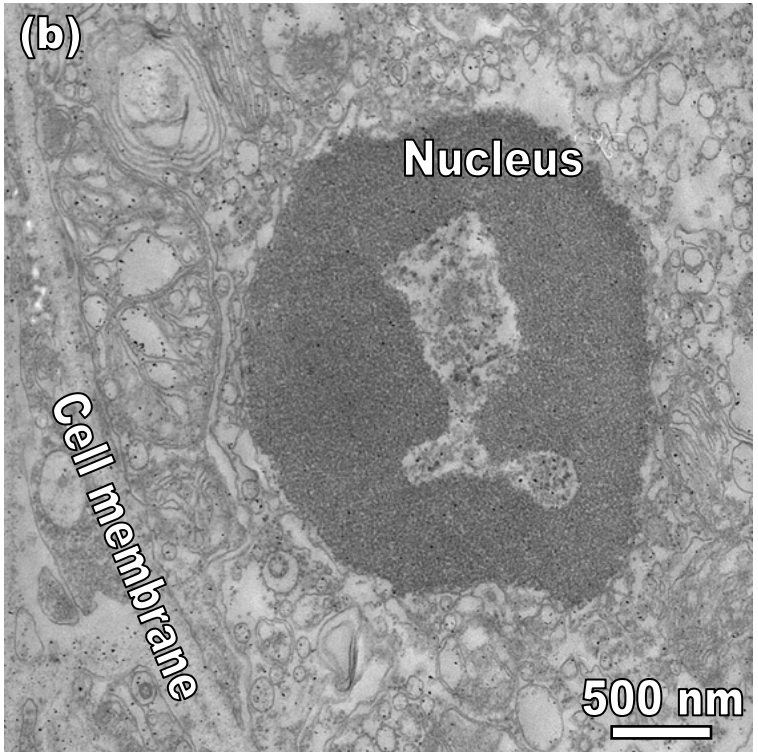


LHRH-SPION in Kidney

SPION in Kidney

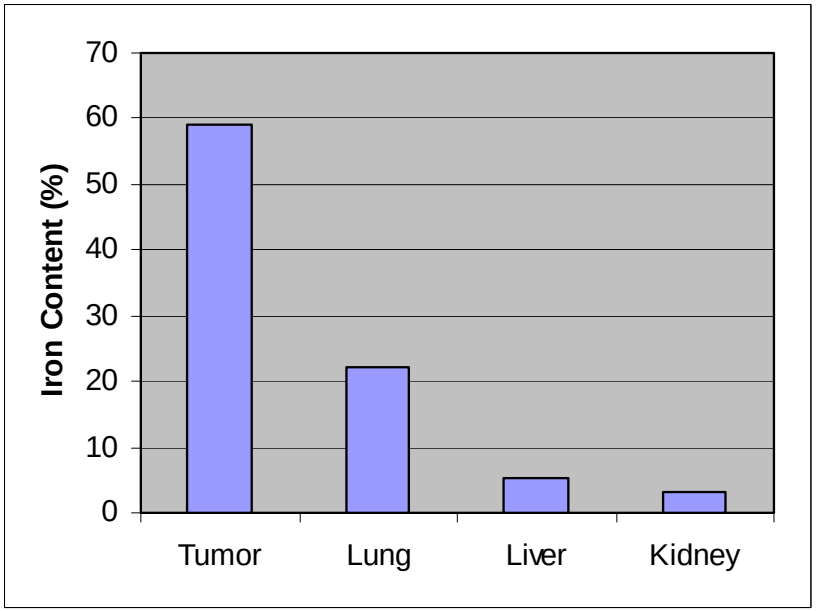


LHRH-SPION in Kidney

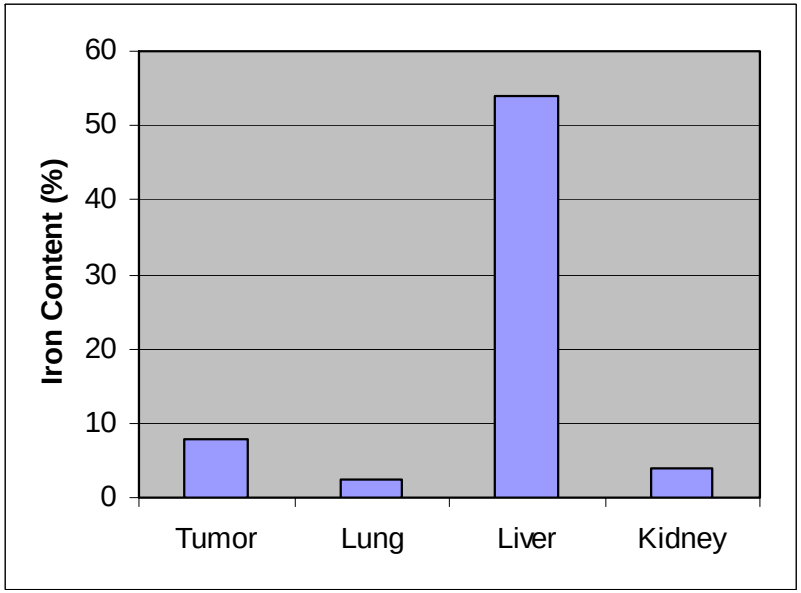


Biological Distribution of SPIONs

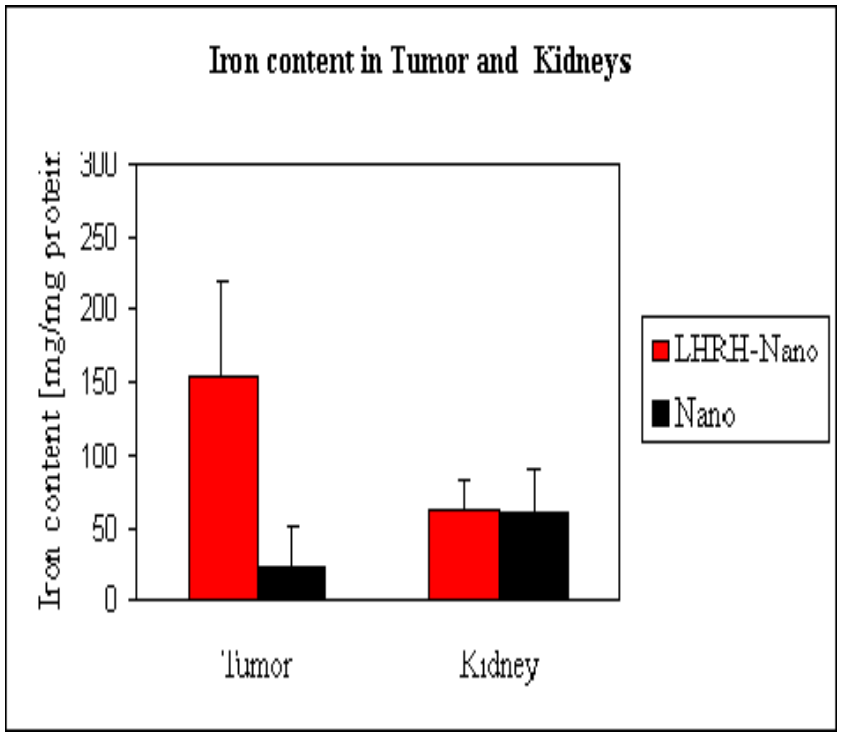
LHRH-SPION in Mouse



SPION in Mouse



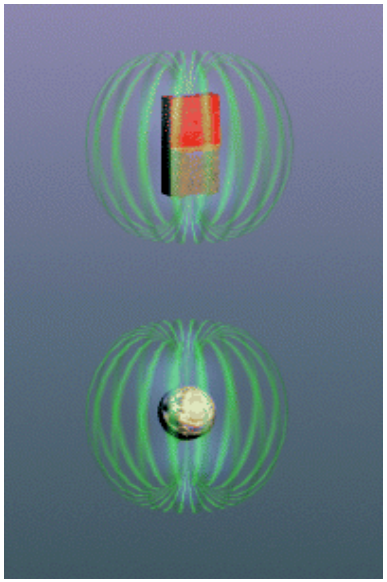
Targeted Destruction of Prostate Cancer in Balb/c athymic nude mice

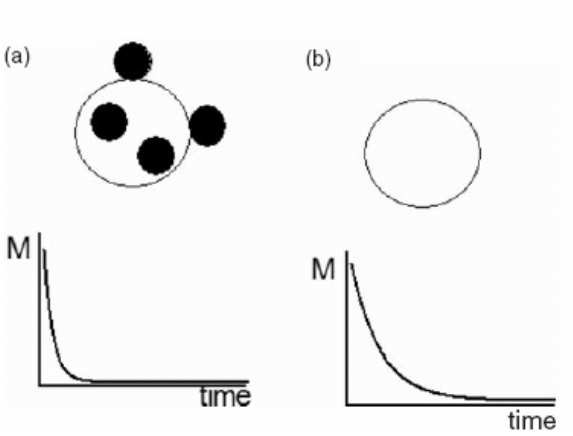


- PC-3.luc Xenograft bearing male nude mice were used
- LHRH bound nanoparticles effectively bind to tumor
- Use of Nano-LHRH results in accumulation 68% of nanoparticles in tumor
- Distribution of iron in other tissues is being mapped

Introduction to MRI

- How does MRI work?
  - Interaction between external magnetic field and spins of protons in hydrogen
  - Spins align due to the external field (z axis)
  - RF pulse tips spins to x-y plane
  - After this pulse, spins relax back
- How does MRI get contrast?
  - Different tissue – hydrogen density / different relaxation property
- How do Contrast Agents work?
  - Change the relaxation property of tissue





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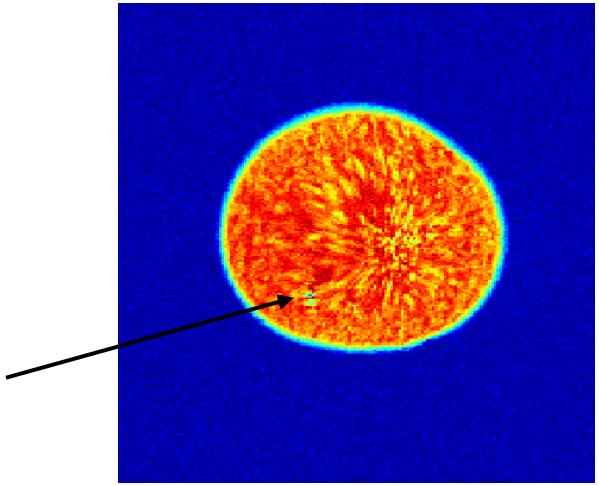
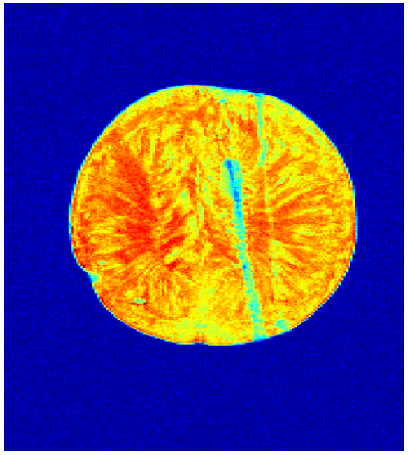
Initial MRI Experiments: Cherry Tomato and Grape

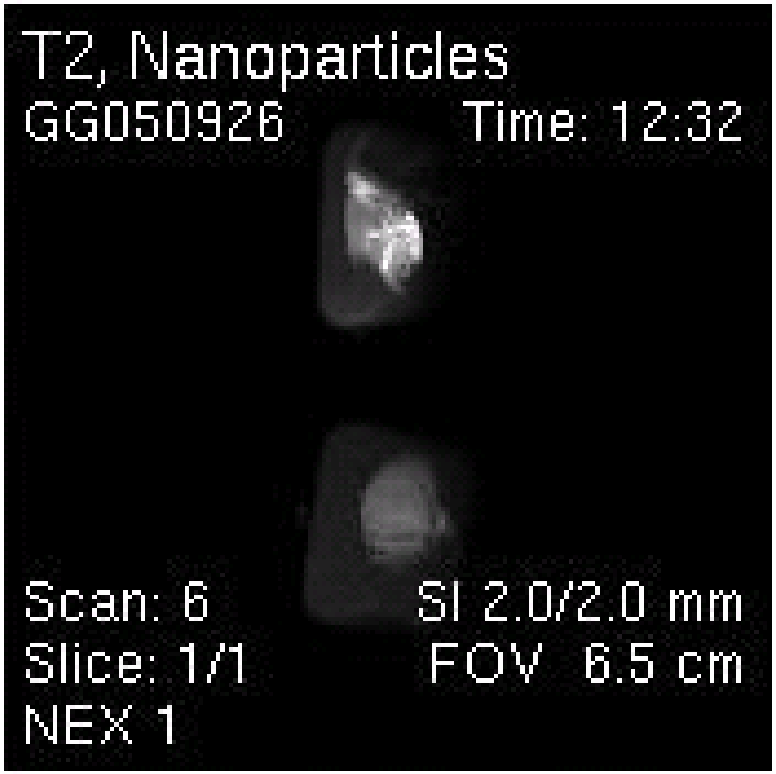
- Injected grapes with saturated saline solution of nanoparticles
- Observed contrast at the location of the injection (nanoparticles)

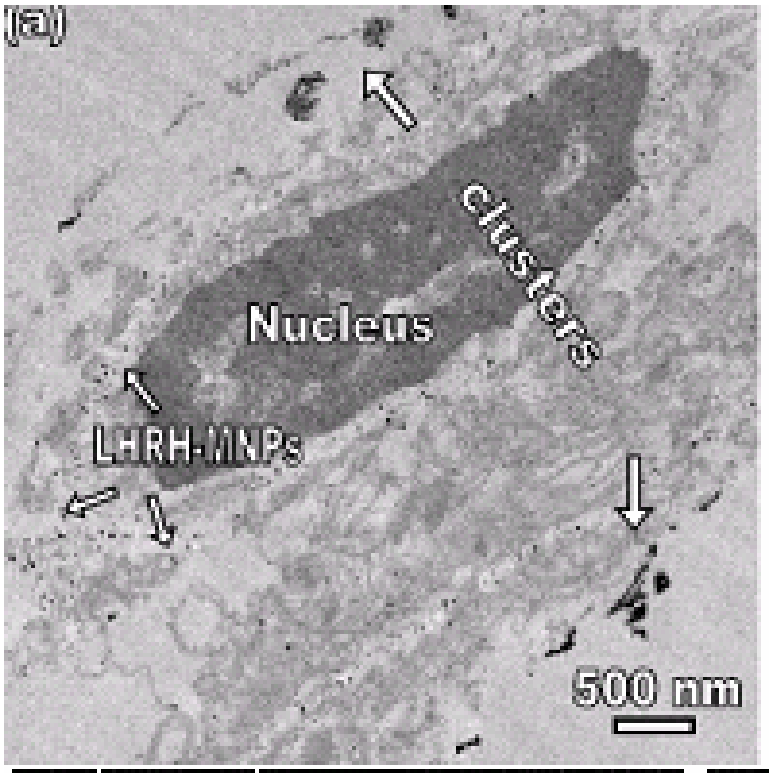
The iron creates a magnetic field in the water, thus creating a blind spot (dark) for the MRI





T2 Images of Tumors – Contrast Enhancement Due to LHRH-MNPs





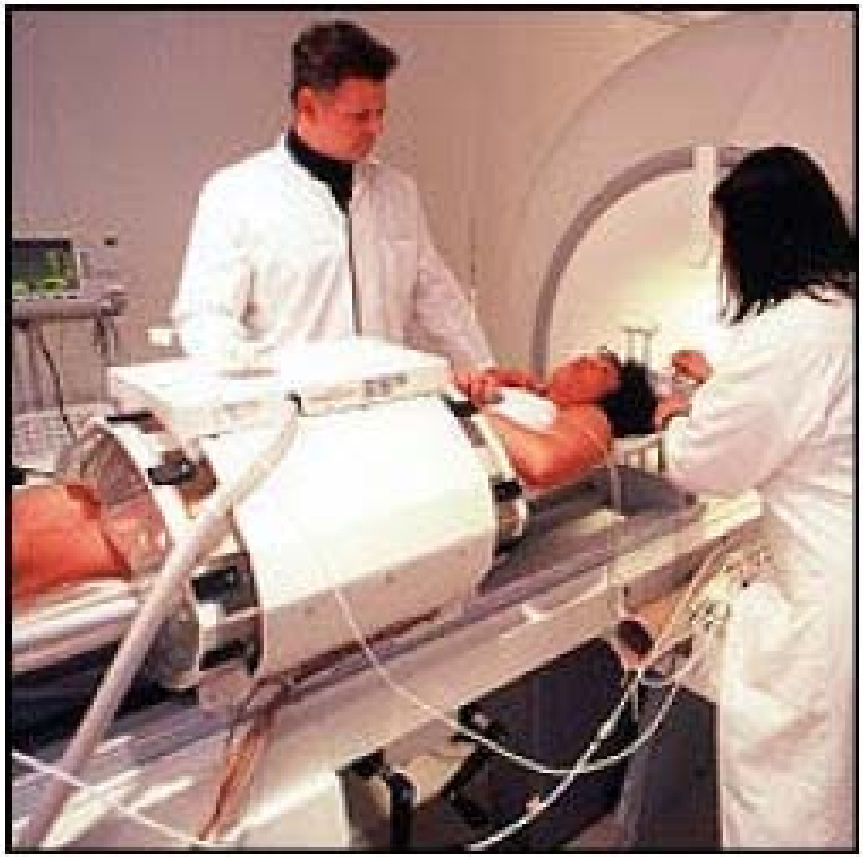
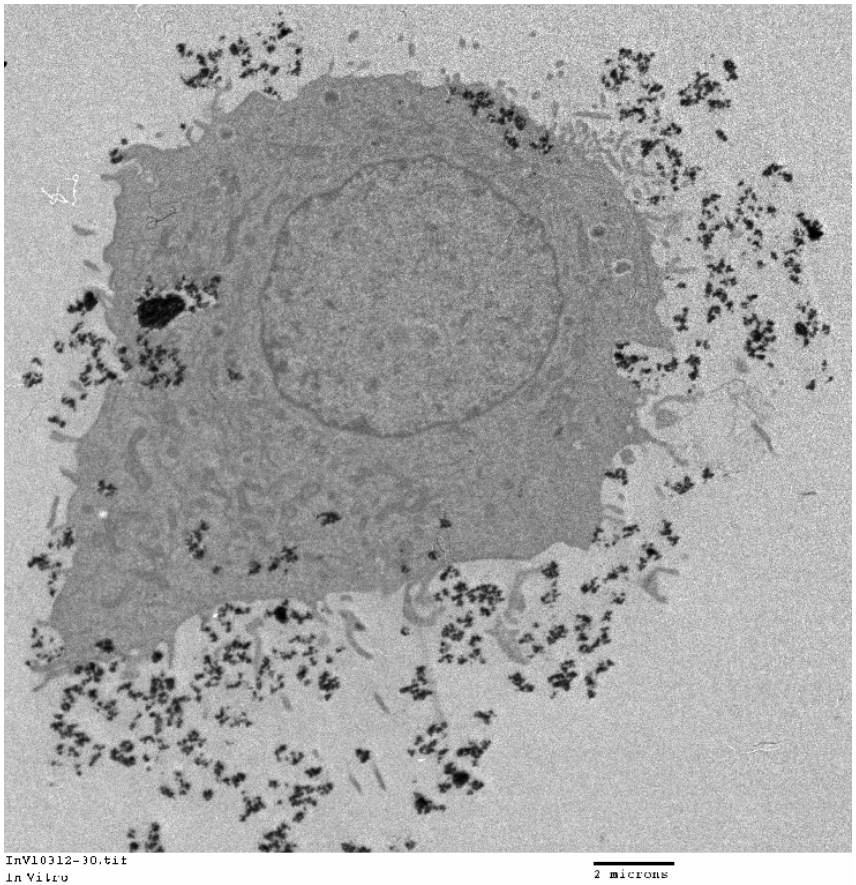


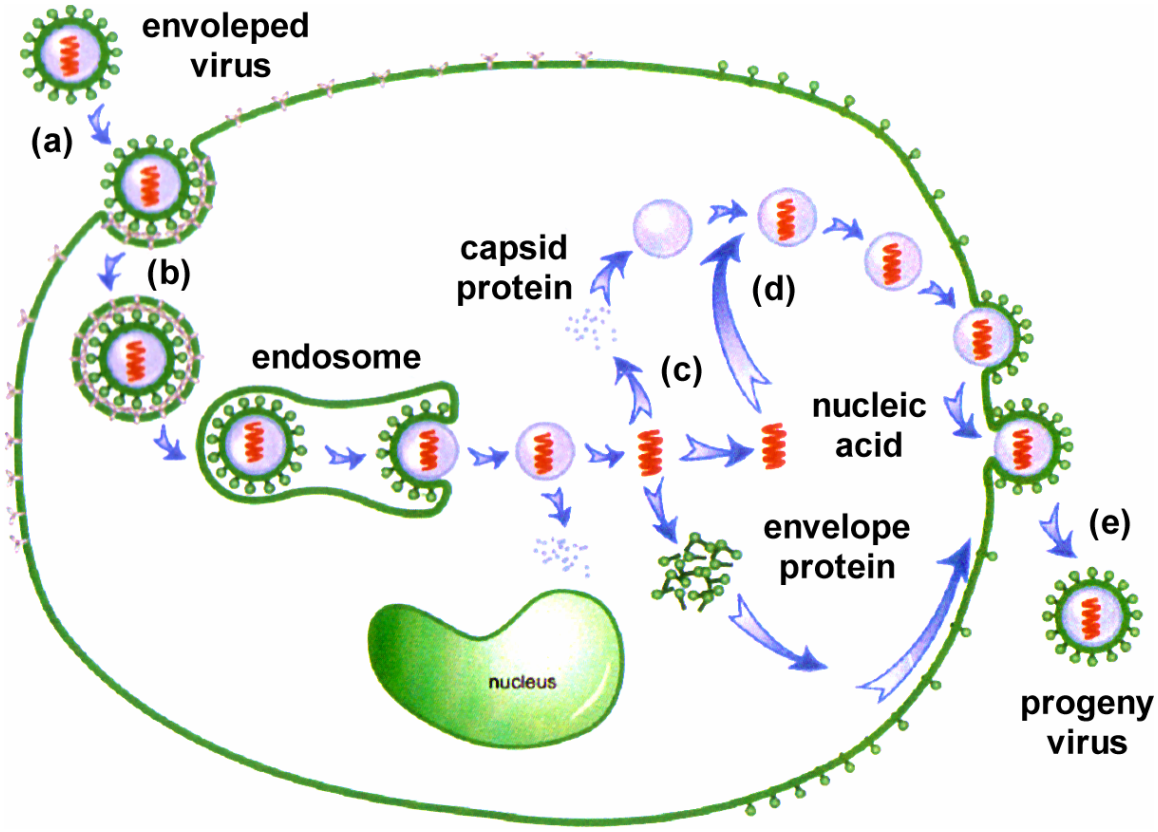
Fig 1 Pictures of hyperthermia treatment equipment (European Society for Hyperthermic Oncology, 2003)

LHRH-SPION Uptake - 37 C for 3 h – Implications Beyond Cancer

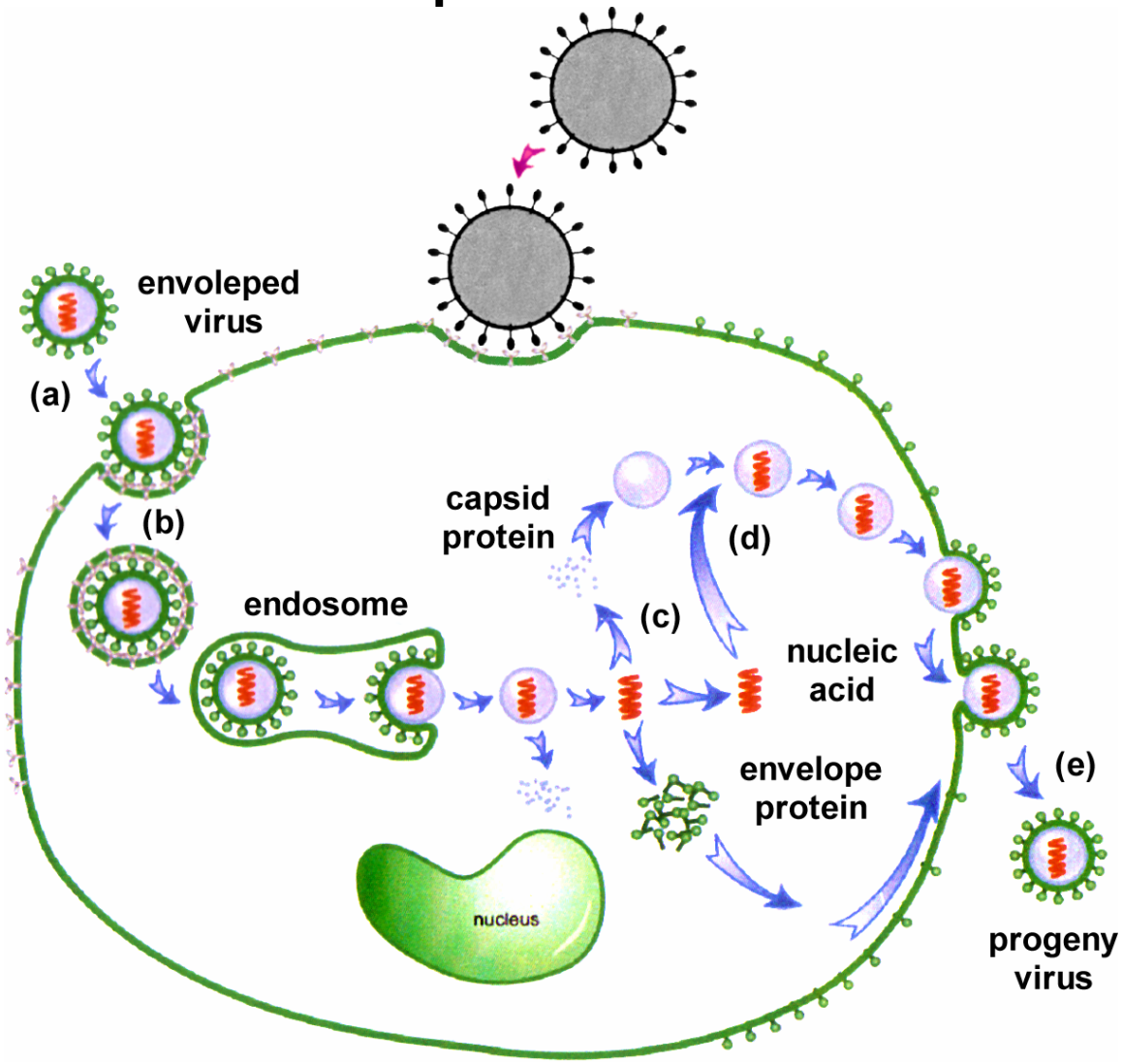
- MNPs-LHRH, 37 C, 3 Hr
- Note endocytosis process by which cells attach
- Engulfed cells carried within the cell
- Excreted or egested within 30 days



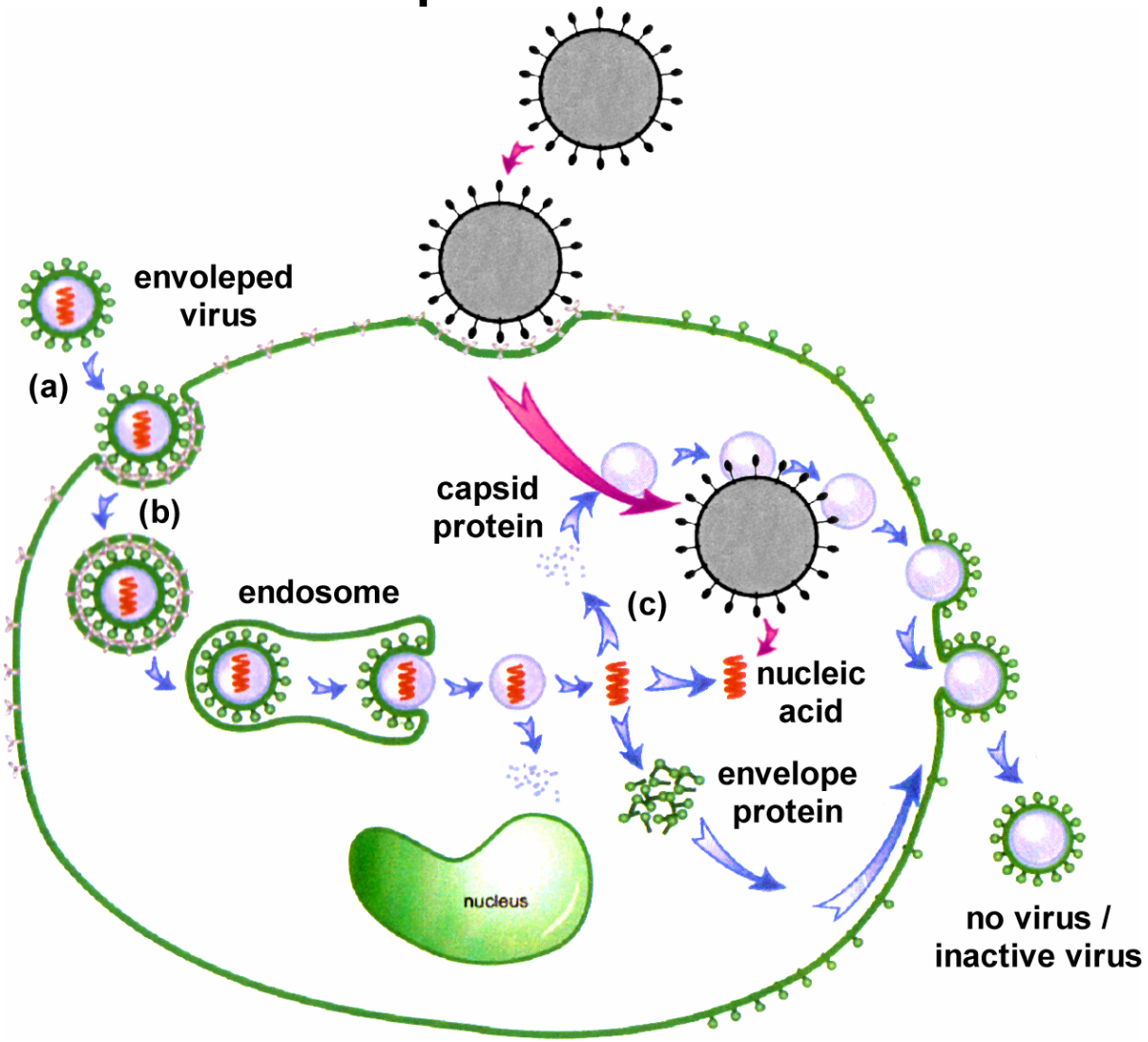
Antagonistic Virus Control with Nanoparticles

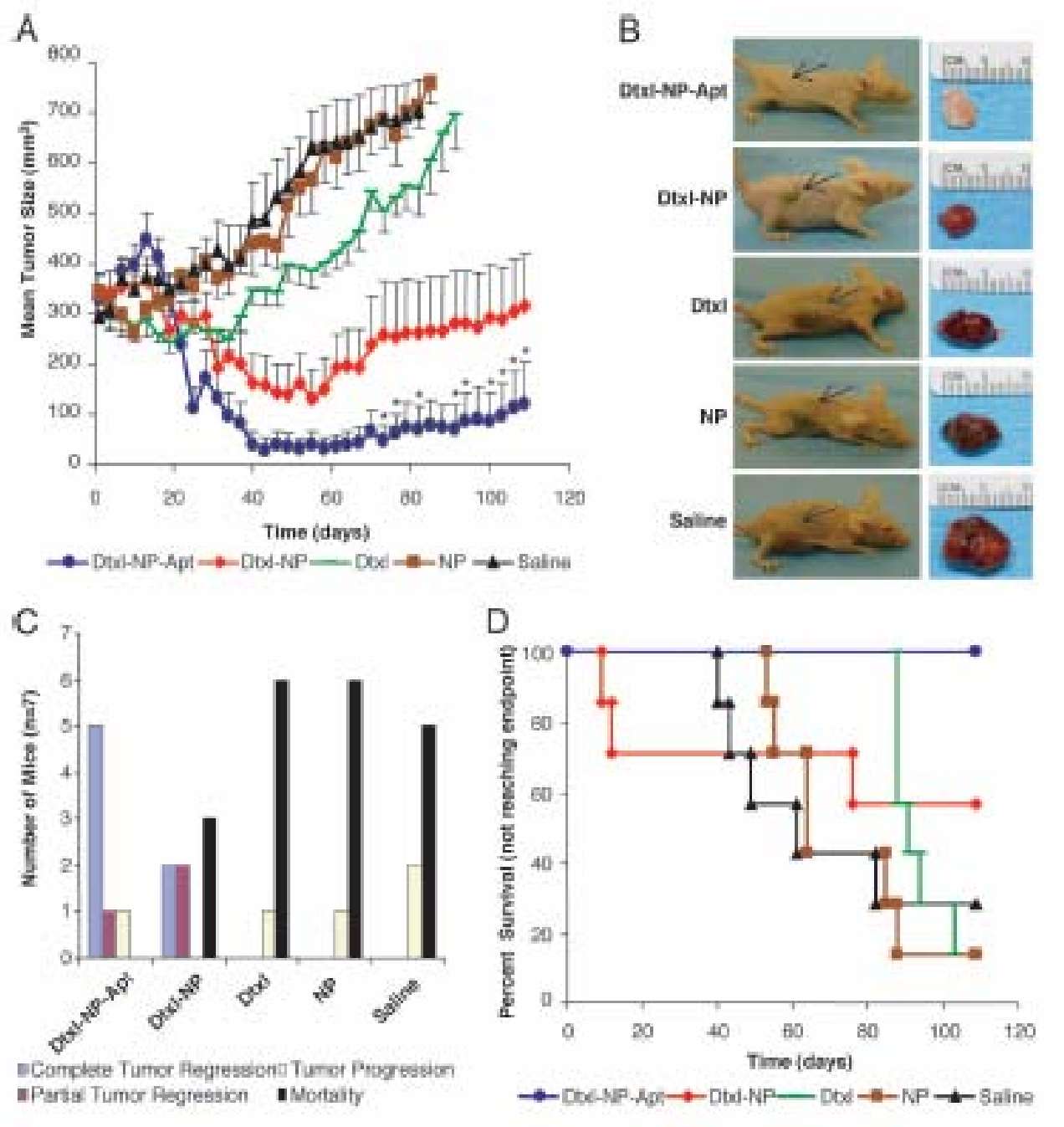


Antagonistic Virus Control with Nanoparticles



Antagonistic Virus Control with Nanoparticles

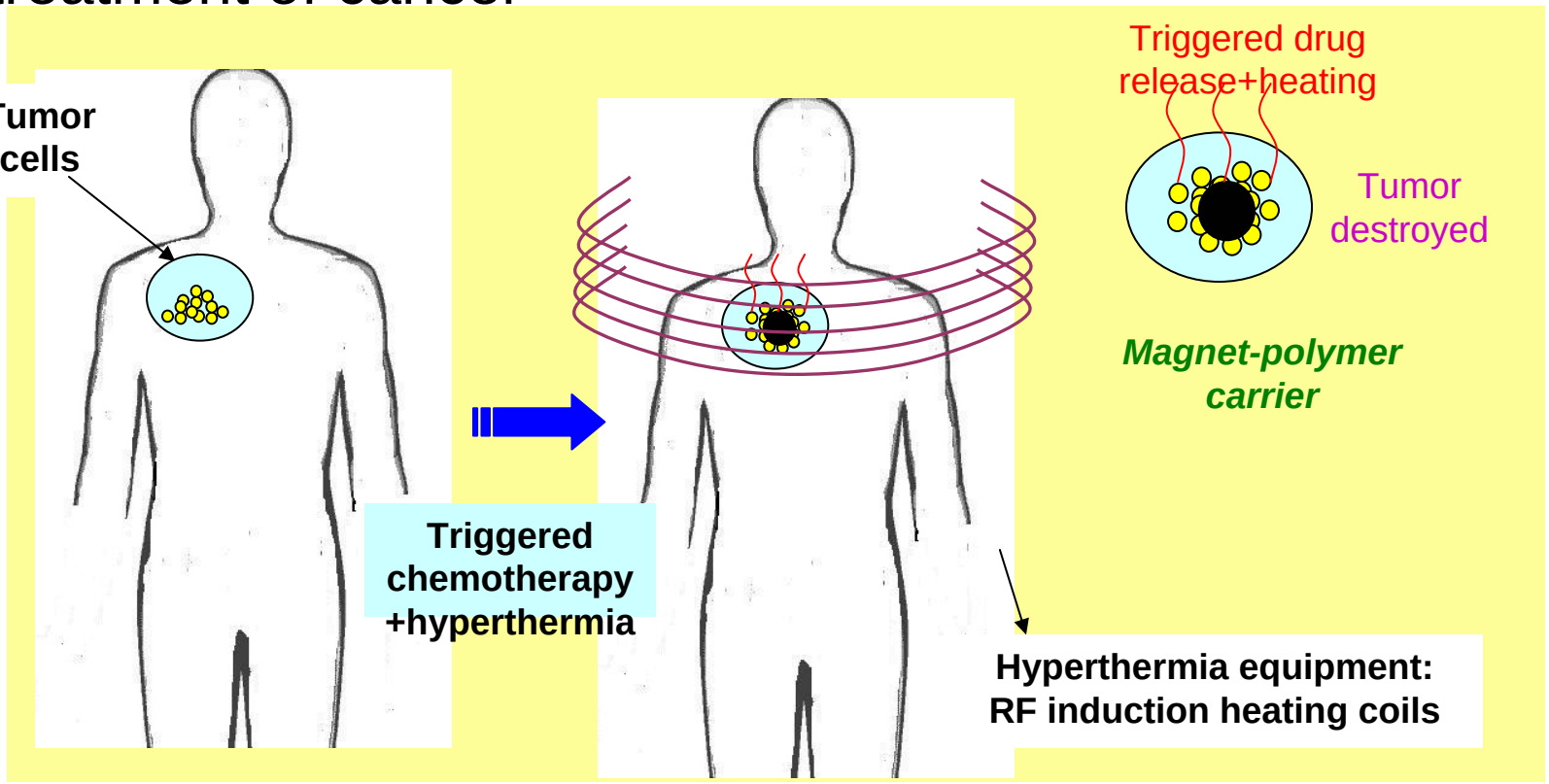




- Tumor Reduction Due to Localized Drug Delivery
- Work of Langer et al., 2006
- USAMI goal is to use materials science approaches to explore ways of shrinking the tumor size to zero
- The other goal is to use localized delivery to reduce the side effects of chemotherapy
- Collaboration with Prud'homme

Triggered Drug Release & Hyperthermia

Combined chemotherapy + Heating of certain organs or tissues to temperatures between 41 and 43^{O}C as a treatment of cancer



Summary and Concluding Remarks

- Overview of some recent work on bio-nanotechnology for disease (mostly cancer) detection and treatment
- In-vitro and in-vivo microscopy reveal stages of specific receptor-mediated nanoparticle endocytosis
- LHRH-coated magnetite particles provide opportunities for early MRI detection and treatment of breast & prostate cancer
- Potential for localized hyperthermia and chemotherapy (to kill cancer cells) being explored
- We welcome your involvement in the ongoing program

THANK YOU!