4-2017

# Combination Therapy of Prostate Cancer Utilizing Functionalized Iron Oxide Nanoparticles Carrying TNF-a and Lactonic Sophorolipids

James Beach
*Pittsburg State University*

Tuhina Banerjee
*Pittsburg State University*

Jyothi Kallu
*Pittsburg State University*

Ryan Higginbotham
*Pittsburg State University*

Richard Gross
*Pittsburg State University*

# Combination Therapy of Prostate Cancer Utilizing Functionalized Iron Oxide Nanoparticles carrying TNF-α and Lactonic Sophorolipids

James Beach, Tuhina Banerjee*, Jyothi Kallu, Ryan Higginbotham, Richard Gross† and Santimukul Santra*

*DEPARTMENT OF CHEMISTRY, PITTSBURG STATE UNIVERSITY, PITTSBURG, KS 66762

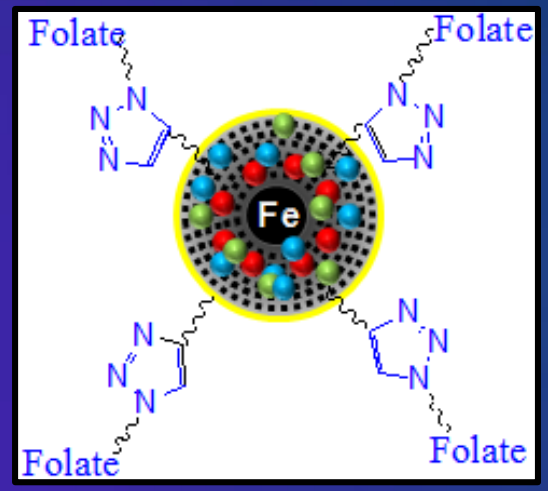
†DEPARTMENT OF CHEMISTRY AND CHEMICAL BIOLOGY, RENSSELAER POLYTECHNIC INSTITUTE, TROY, NY 12180

# Outline

- Introduction
  - What are nanoparticles?
  - Tumor Necrosis Factor-alpha (TNF-α) and Lactonic sophorolipids (LSLs)
- Experimental
  - Synthesis of IONPs & Surface Ligand Modification
- Results
  - Characterizations
  - Microscopy Images
  - Biological Assays
- Conclusion

# Introduction: What are Nanoparticles?

- Nanoparticles are tiny (1-100 nm) particles that exhibit unique properties and characteristics at nano-scale.
- Many uses in the field of biomedicine and therapeutics
  - Targeted drug delivery
    - Encapsulation of small molecules (drugs, optical dyes)
      - Dosage control and imaging
    - Surface ligand modification (folic acid) for receptor specificity
      - Only treat cells of interest
  - MRI Contrast Imaging (Iron Oxide nanoparticles)

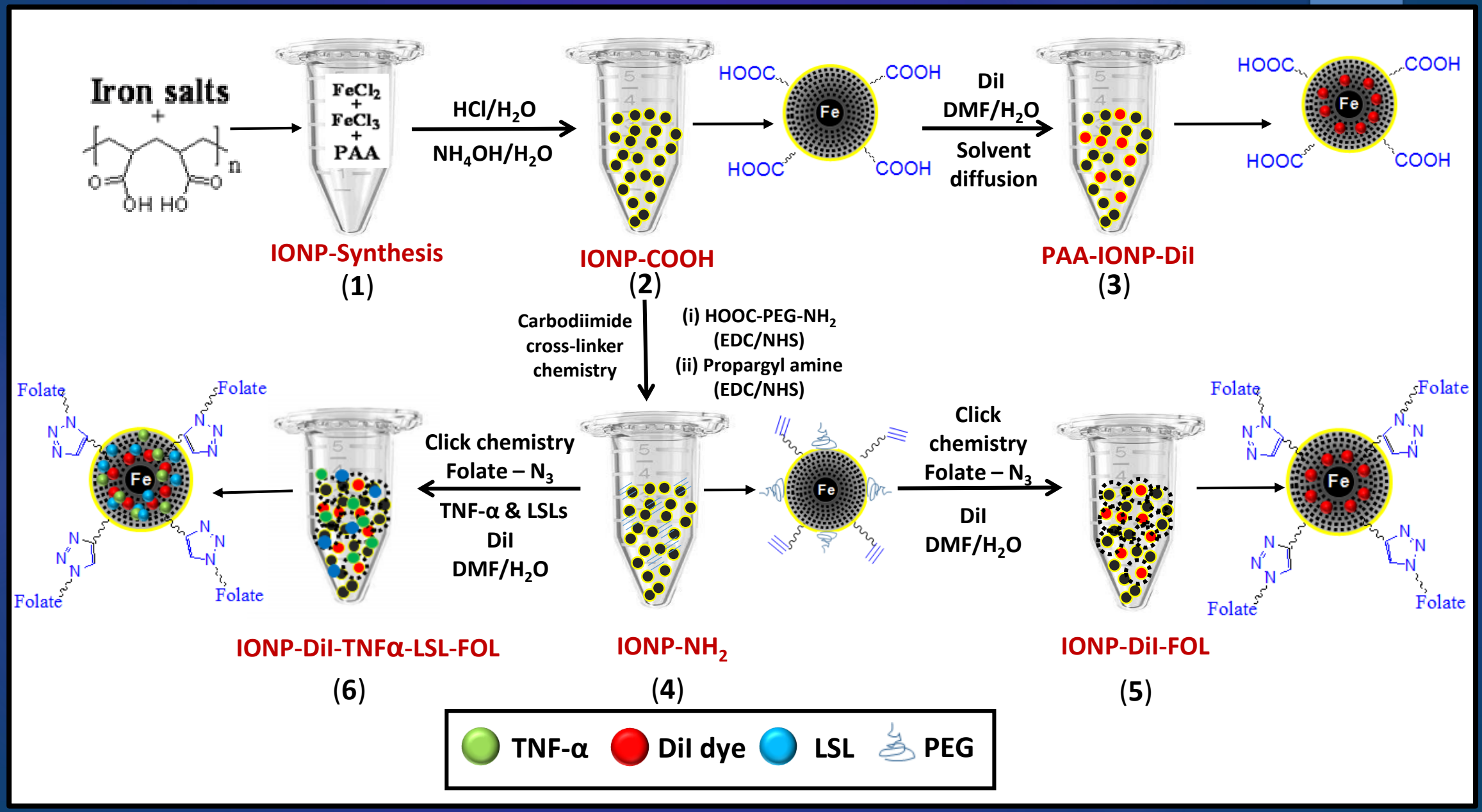


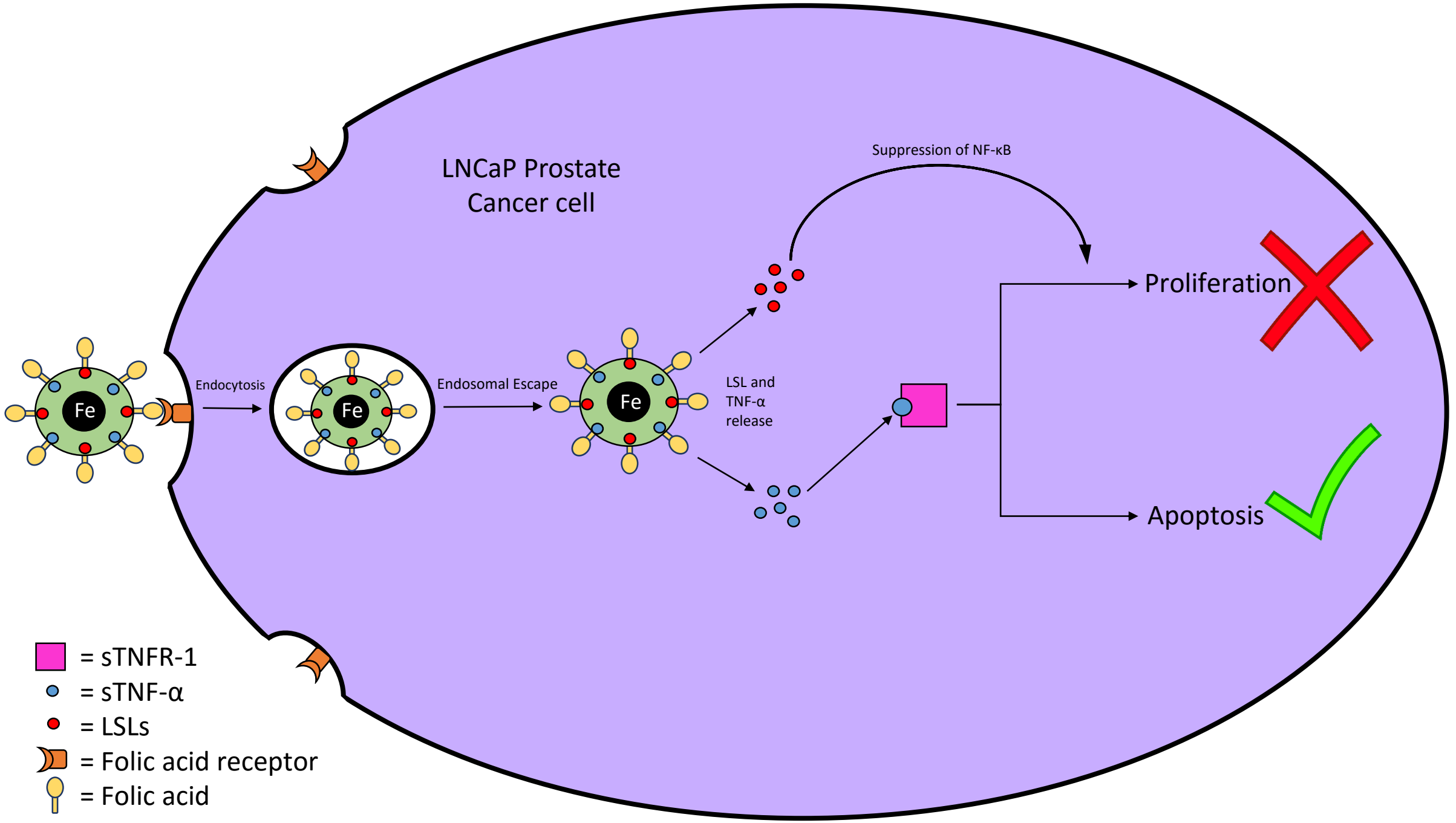

- Our Aim: Treat LNCaP strain prostate cancer with a combination therapy of soluble TNF-α and LSLs with folate-functionalized iron oxide nanoparticles (IONPs)

# Introduction: Why use TNF-α and LSLs?

- TNF-α
  - Cytokine important in many cellular pathways
    - Apoptosis and proliferation pathways
  - In cancer cells, TNF-α and associated proteins behave aberrantly
    - Nuclear factor kappa B (NF-κB) initiates proliferation unchecked
    - Binding to its receptor, TNFR-1, does not occur in tumor cells
  - Solution: Introduction of exogenous soluble TNF-α may help initiate cell death in tumors
    - Inspired by Aurimune* (gold nanoparticle)
- LSLs
  - Glycolipids extracted from non-pathogenic yeast
  - Enhance immune response and reduce inflammation
    - Associated with large decreases in cytokine mRNA
    - Suspected inhibition of NF-κB
  - Implementation inspired by Dr. Richard Gross' research
  - Hypothesis: Synergy between these two compounds?

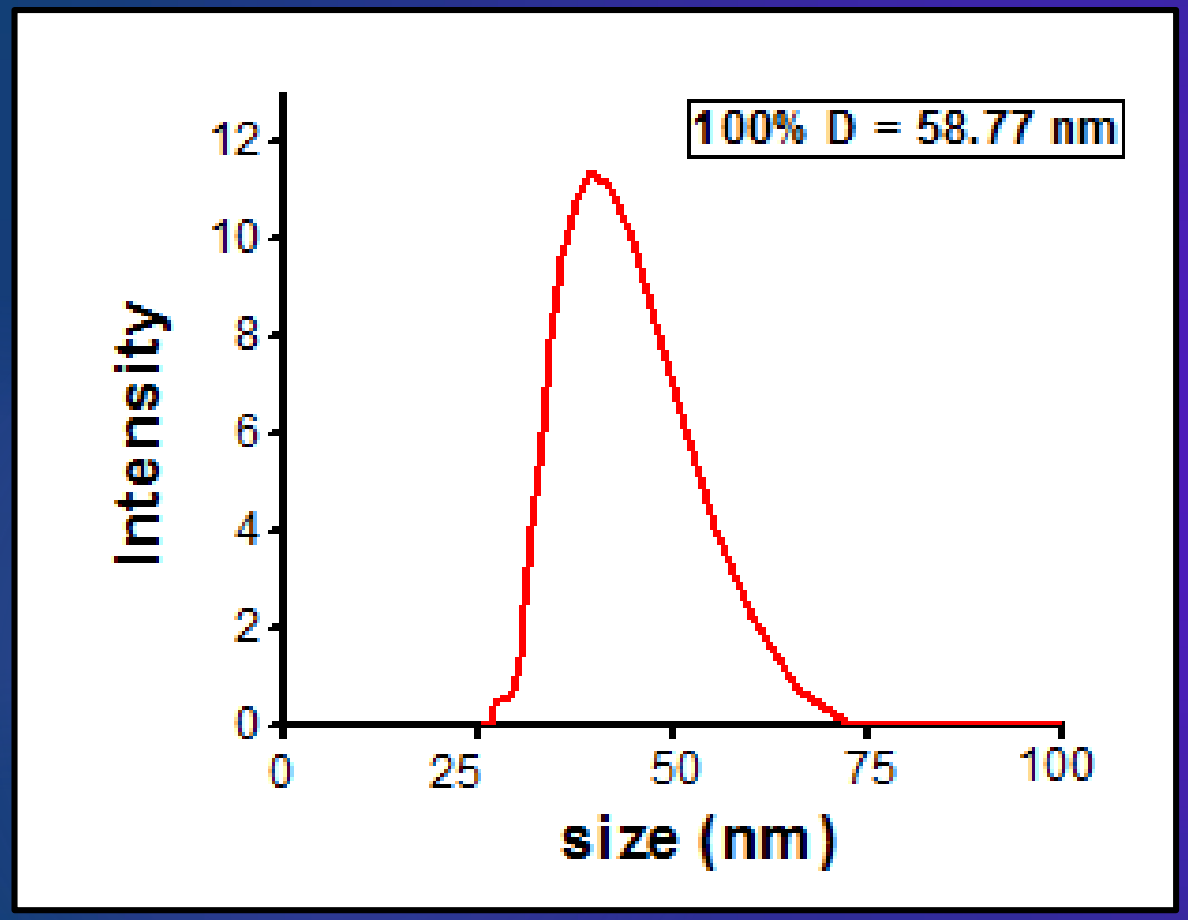
# Experimental: Nanoparticle Synthesis



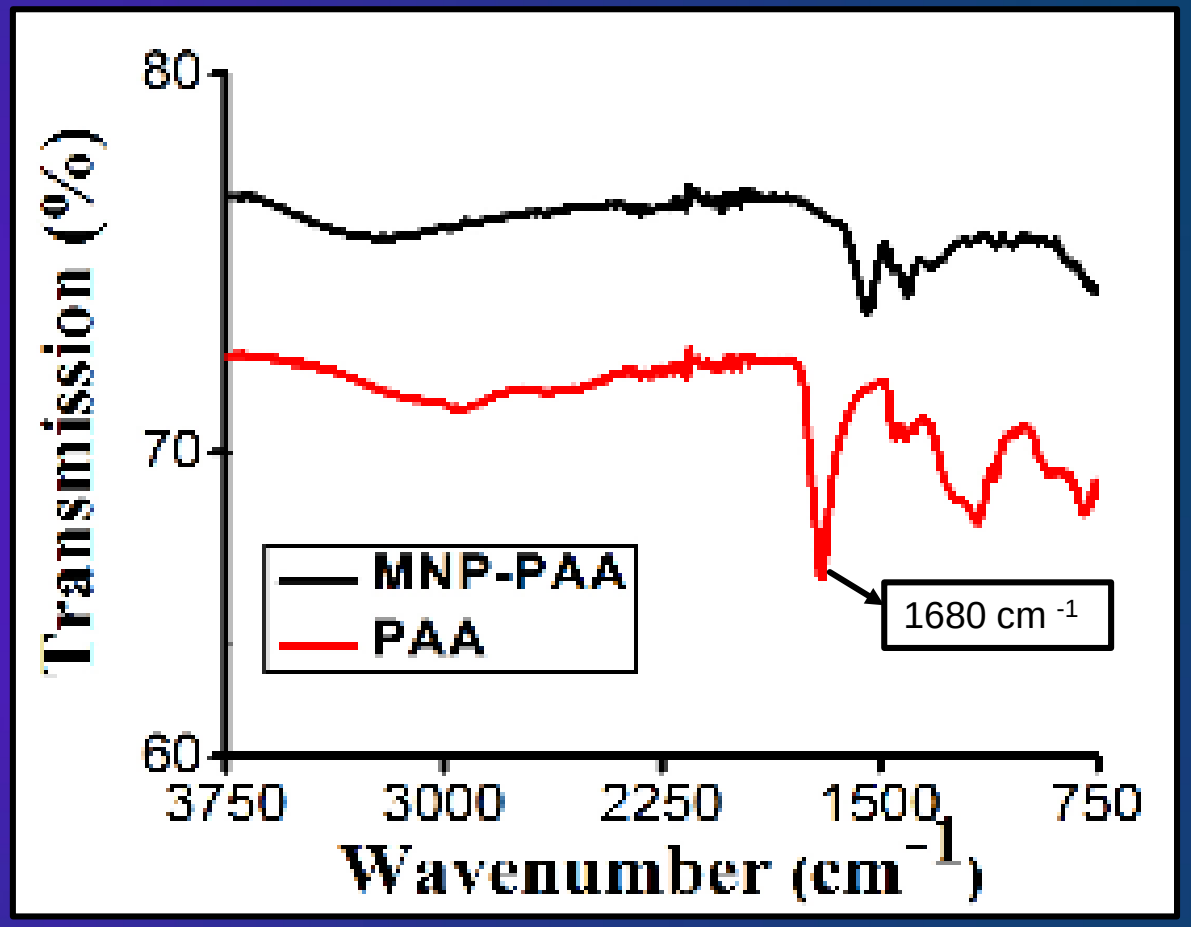

LNCaP Prostate
Cancer cell
Suppression of NF-κB
Proliferation
Apoptosis
Endocytosis
Endosomal Escape
LSL and
TNF-α
release
Fe
Fe
Fe
= sTNFR-1
= sTNF-α
= LSLs
= Folic acid receptor
= Folic acid

# Results: IONP Characterization

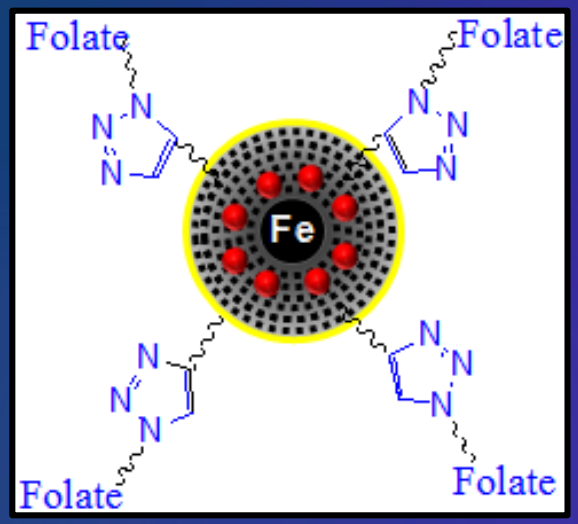
Dynamic Light Scattering

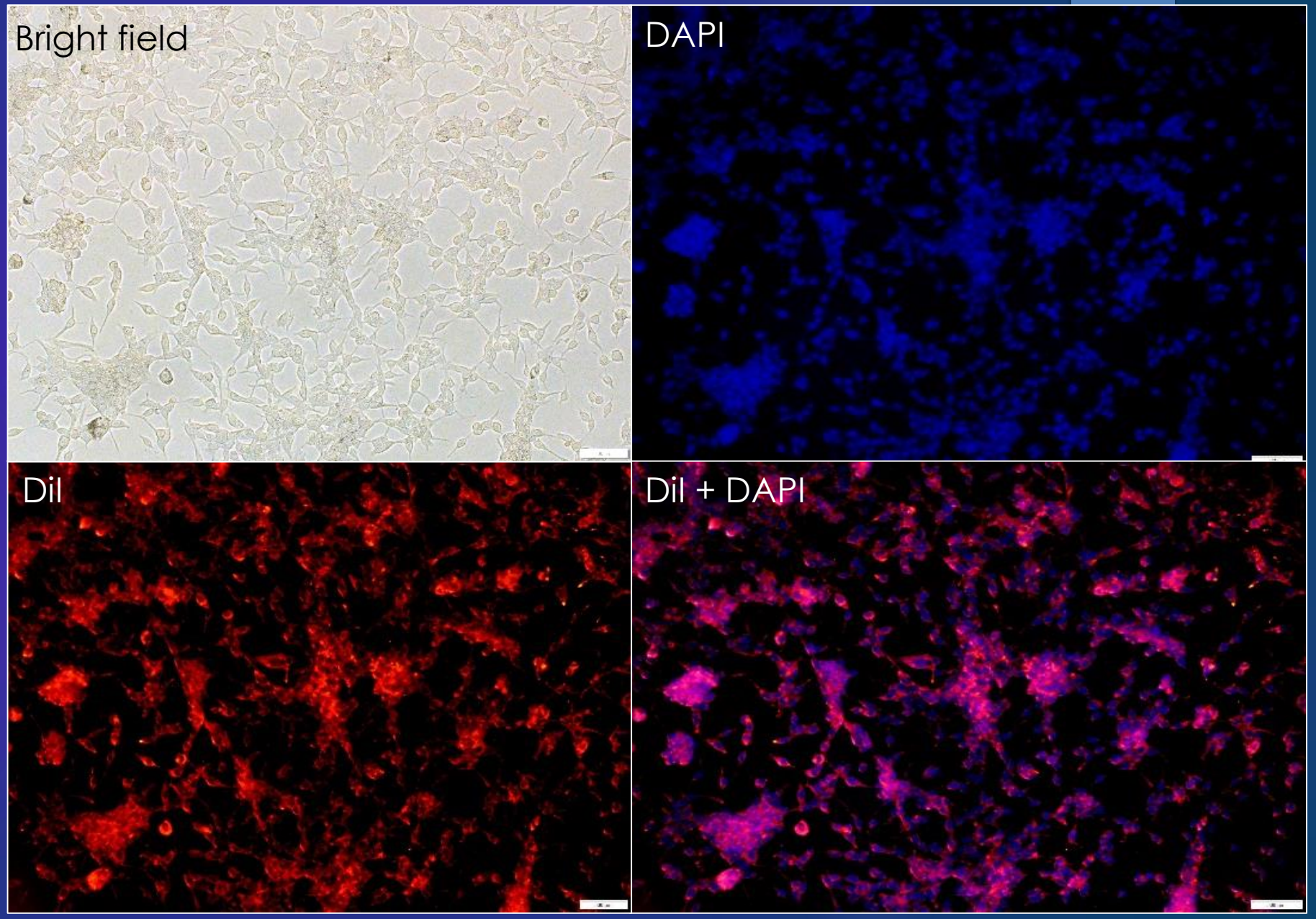



FT-IR

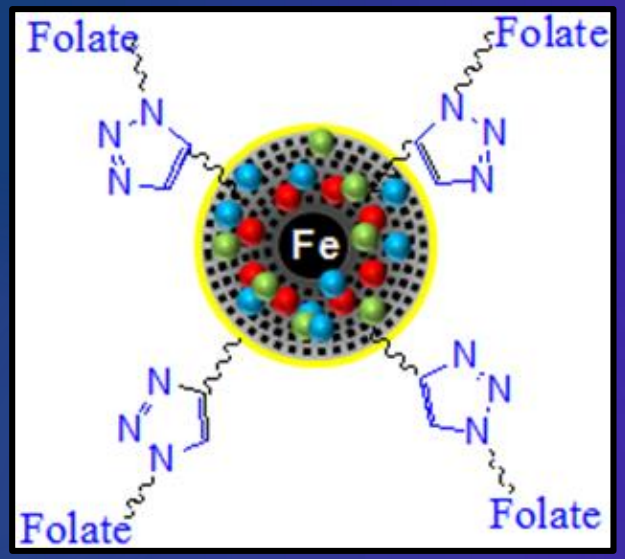


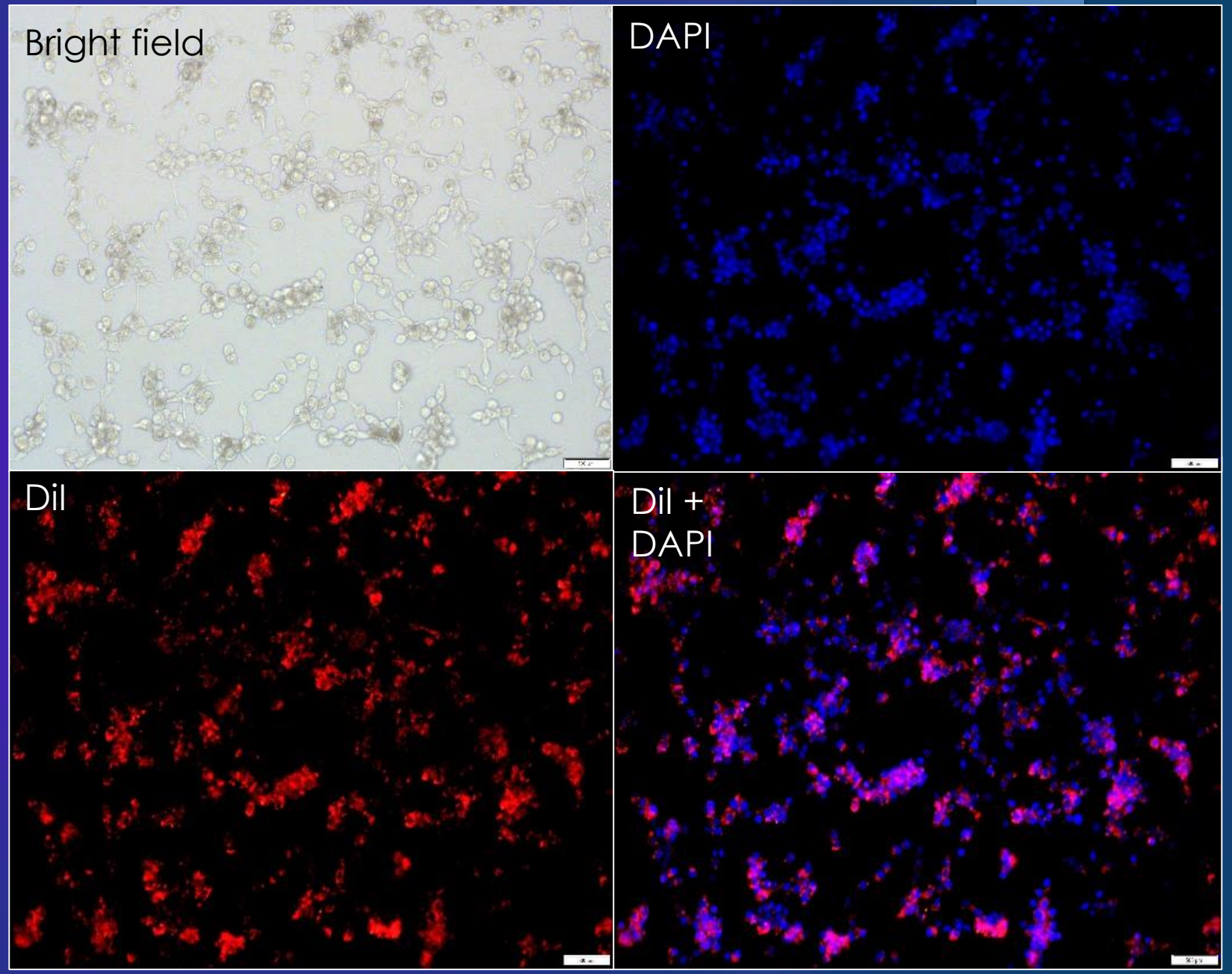
# Results: Fluorescence Microscopy – Dye Internalization



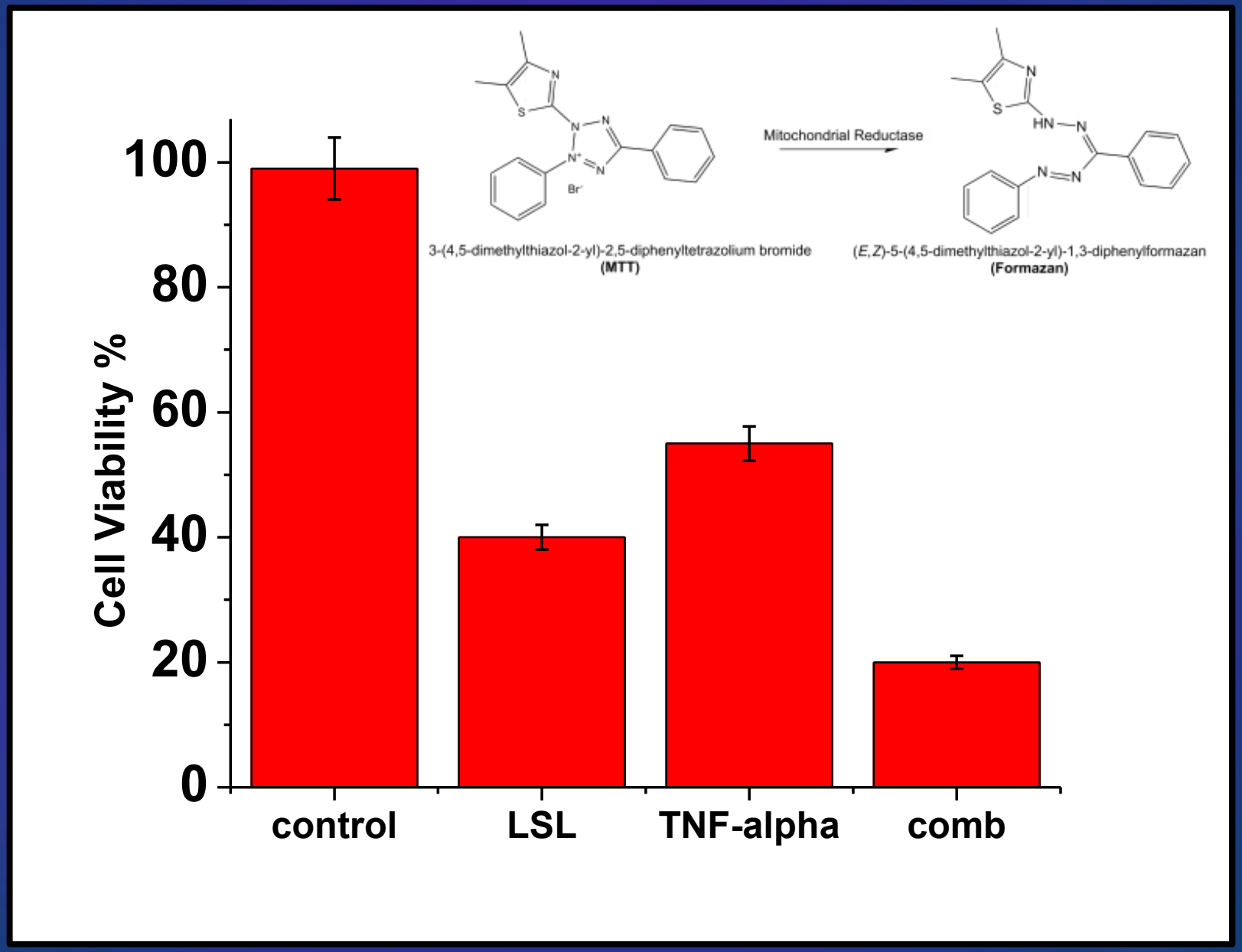



# Results: Fluorescence Microscopy – Dye and Combination Therapy



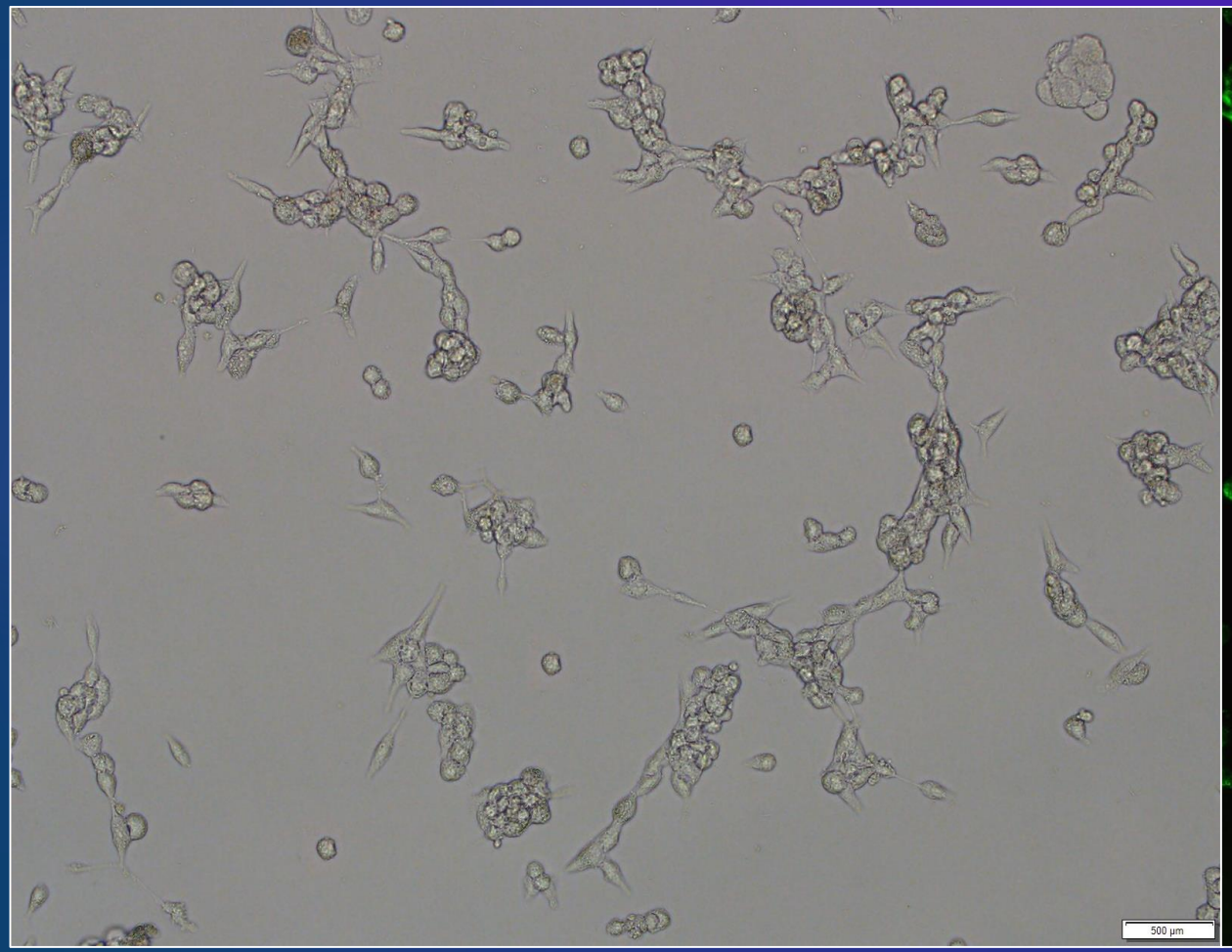



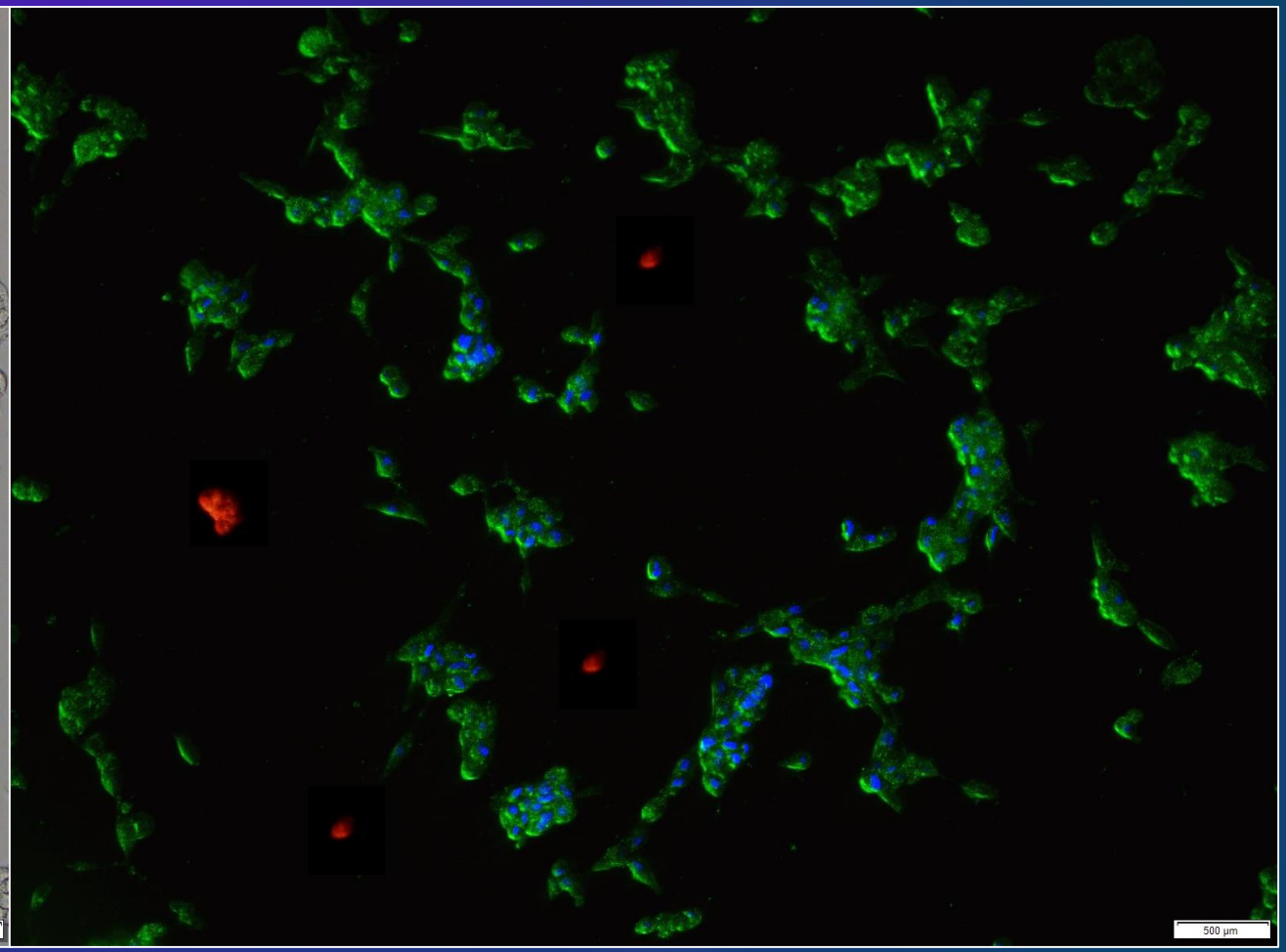
# Results: MTT Assay



# Results: Apoptosis/Necrosis Assay (TNF-α)

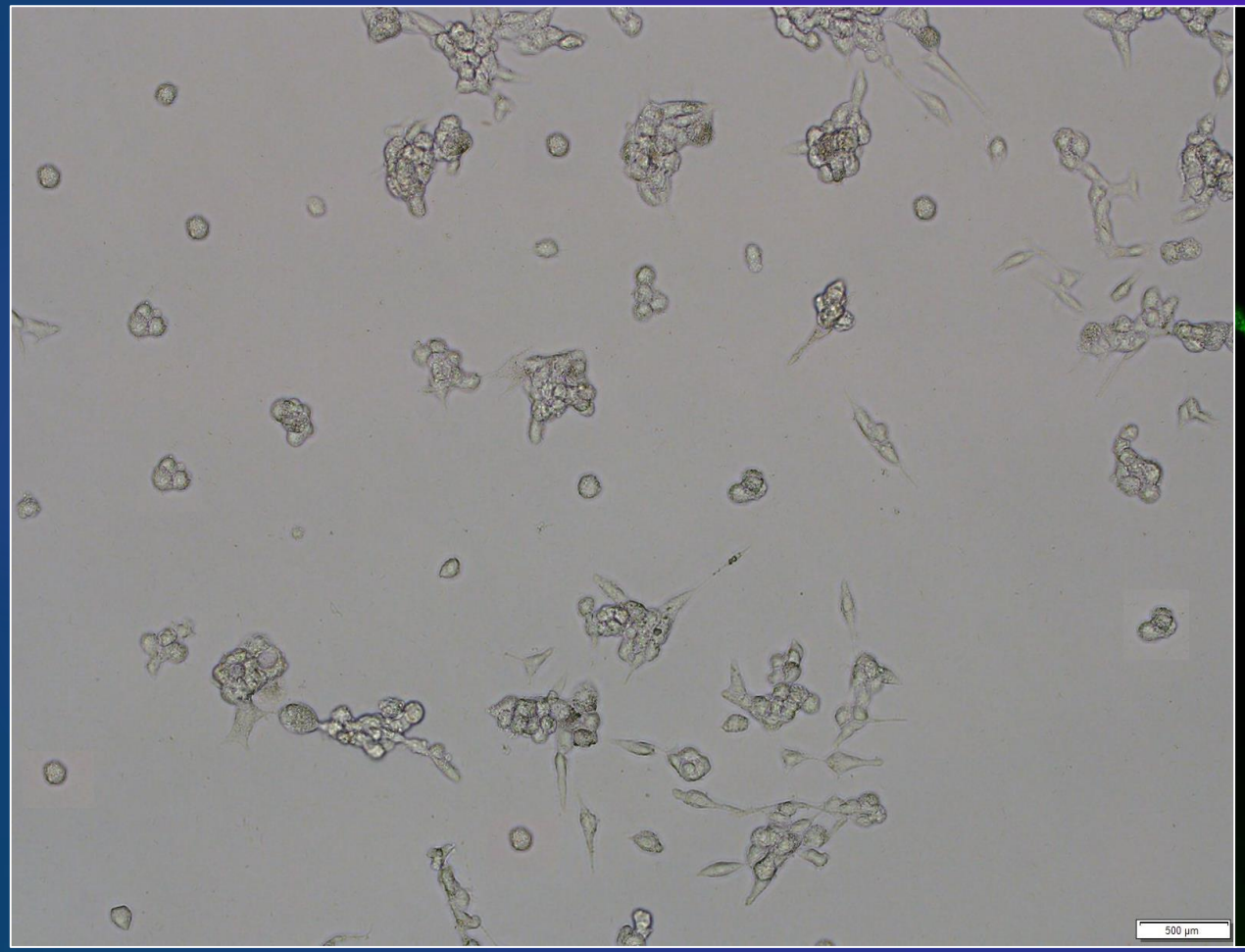
Annexin-V/Fluorescein
Hoescht
Ethidium homodimer

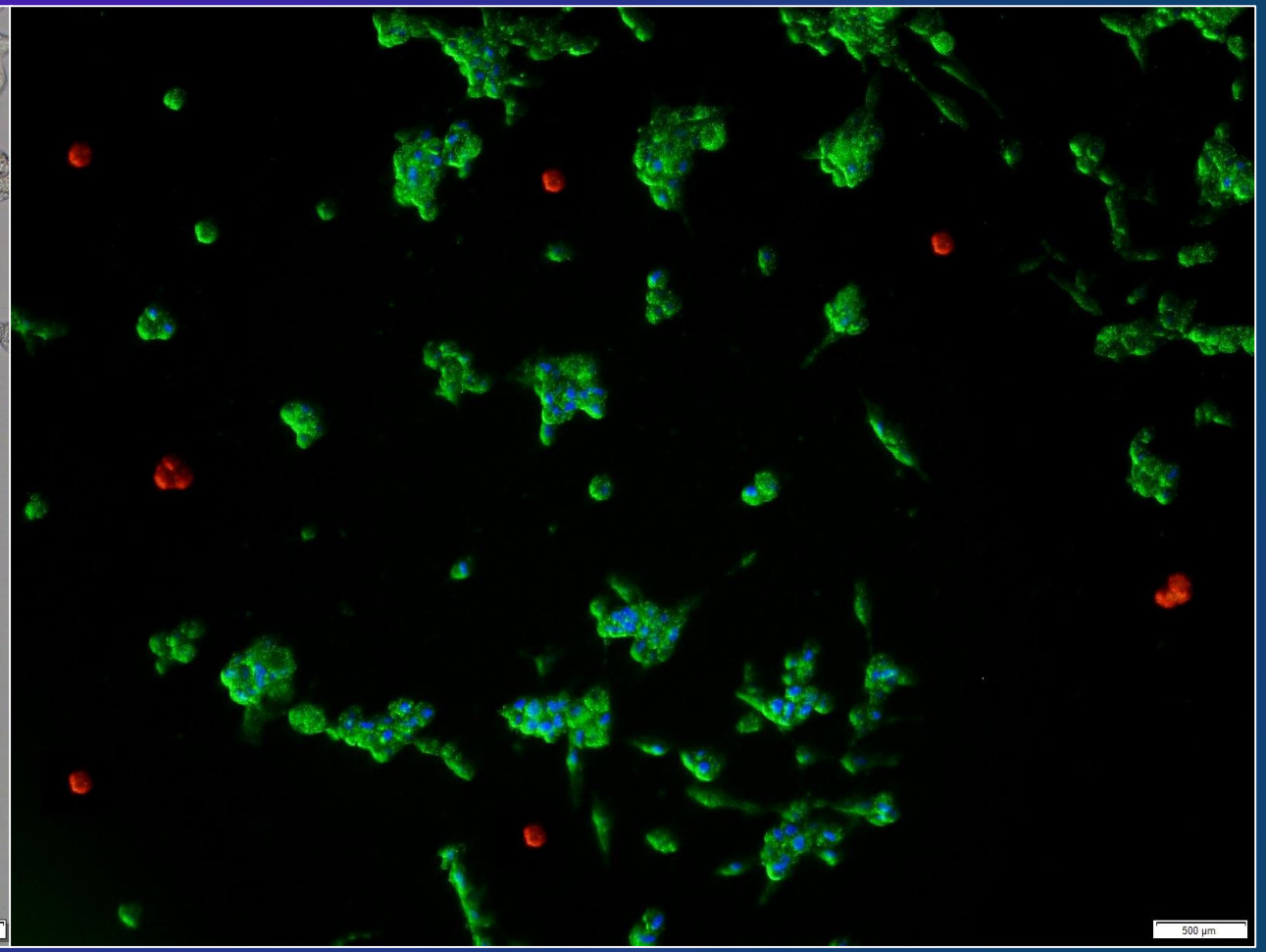


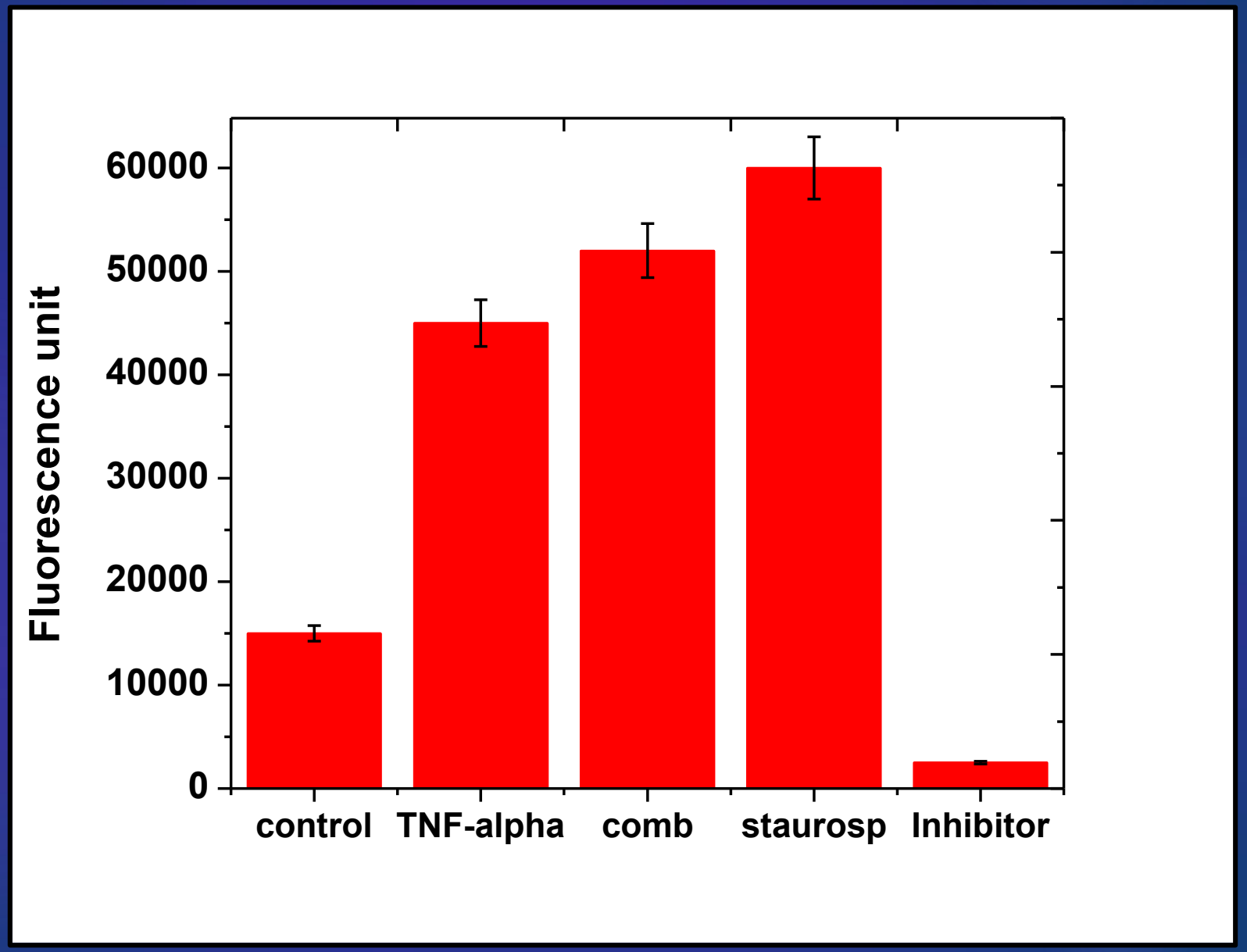
# Results: Apoptosis/Necrosis Assay (Combination)

Annexin-V/Fluorescein
Hoescht
Ethidium homodimer

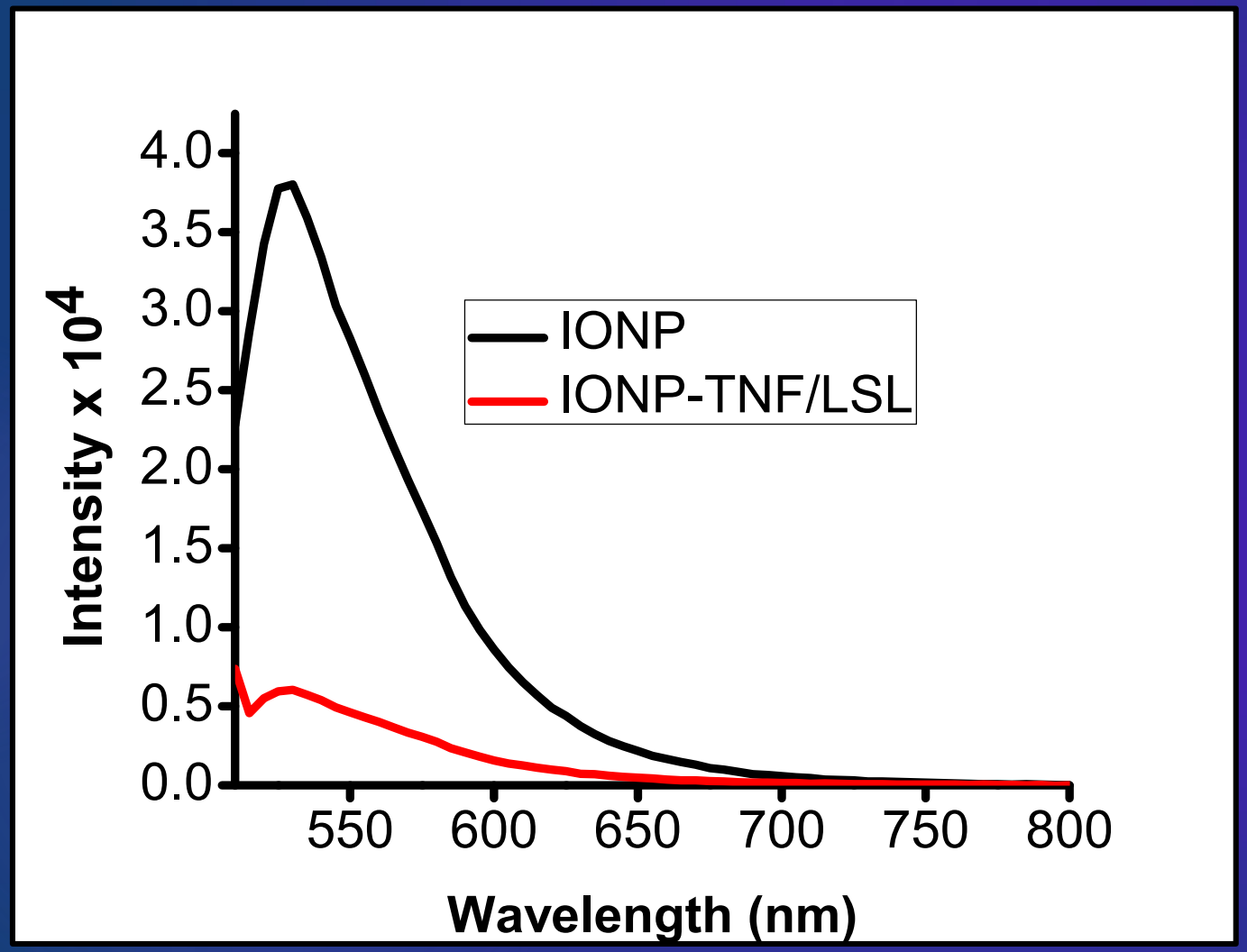


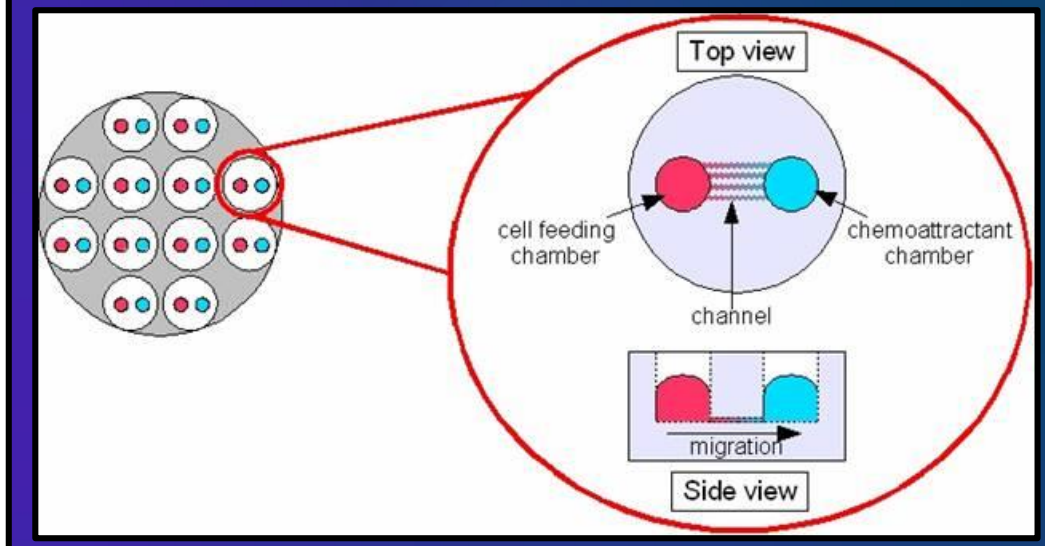



# Results: Apoptosis/Necrosis Assay Results



# Results: Migration Assay



# Conclusions

- Successful synthesis of folate-conjugated IONPs and encapsulation of TNF-α and LSLs
- Results of cytotoxicity assays show up to 80% cell death with combined treatment after 24 hrs
- Significant increase in apoptotic initiation following 24 hr. incubation with TNF-α and combination treatment
- Our results support our hypothesis the synergistic combined therapy
- Next step: Look to in-vivo mouse models for treatment

# Thank You!

# References


- Van Horssen R., Ten Hagen T.L.M., Eggermont A.M.M. TNF-α in Cancer Treatment: Molecular Insights, Antitumor Effects, and Clinical Utility. *The Oncologist*. **2006**, 11, 397-408.
- Hardin R., Pierre J., Schulze R., Mueller C.M., Fu S.L., Wallner S.R., Stanek A., Shah V., Gross R.A., Weedon J., Nowakowski M., Zenilman M.E. Bluth M.H. Sophorolipids Improve Sepsis Survival: Effects of Dosing and Derivatives. *J. Surg. Res*. **2007**, 142, 314–319.
- Sethi G., Aggarwal B. TNF: A master switch for inflammation to cancer. *Frontiers in Biosci*. **2008**, 13. 5945-5107.
- Shenoi M., Iltis I., Choi J., Koonce N., Metzger G., Griffin R., Bischof J. Nanoparticle Delivered Vascular Disrupting Agents (VDAs): Use of TNF-alpha conjugated Gold Nanoparticles for Multimodal Cancer Therapy. *Mol Pharm*. **2013**, 10 (5), 1683-1694.
- Wang L., Du F., Wang X. TNF-α Induces Two Distinct Caspase-8 Activation Pathways. *Cell*. **2008**, 133, 693-703.