Genetic programming of macrophages to perform anti-tumor functions using targeted mRNA nanocarriers

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Disclosure

I, Fan Zhang have no financial relationships to disclose in relation

to the content of this activity

# Macrophage Heterogeneity

M2

M2-like

M1-like

M1

Farbstudie Quadrate, 1913

Macrophage Regulate Tumor Progression

Kaplan-Meier survival analyses of 110 stage III-IV epithelial ovarian cancer patients
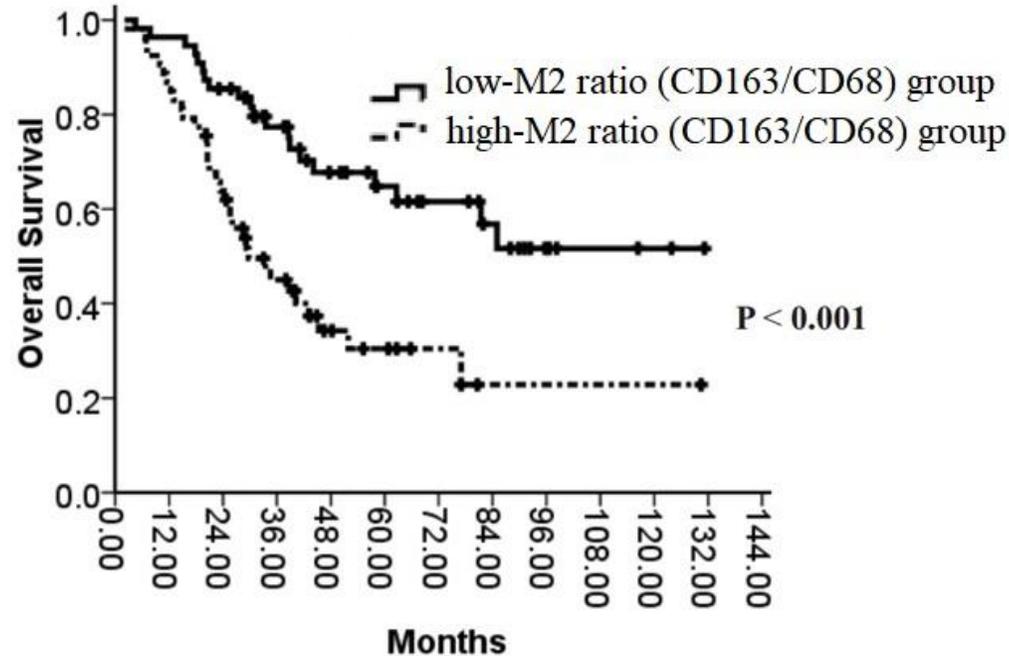
Increased M2/M1 macrophages ratio is associated with tumor progression

Lan C. Technol Cancer Res Treat. 2013 Jun;12(3):259-67.

Status quo strategies to target macrophages

Small molecule drugs	Cytokine blockade: macrophages recruitment (CSF1-CSF1R, CCL2-CCR2)
Antibodies	Cytokine blockade Antibodies Anti MARCO antibodies Anti-CD47 antibodies

Lack of response rate;   Disrupt the homeostasis;

HYPOTHESIS

We can program anti-tumor macrophages by using **mRNAs** encodes for
the **transcription factor** that is the master regulator of **macrophage polarization**
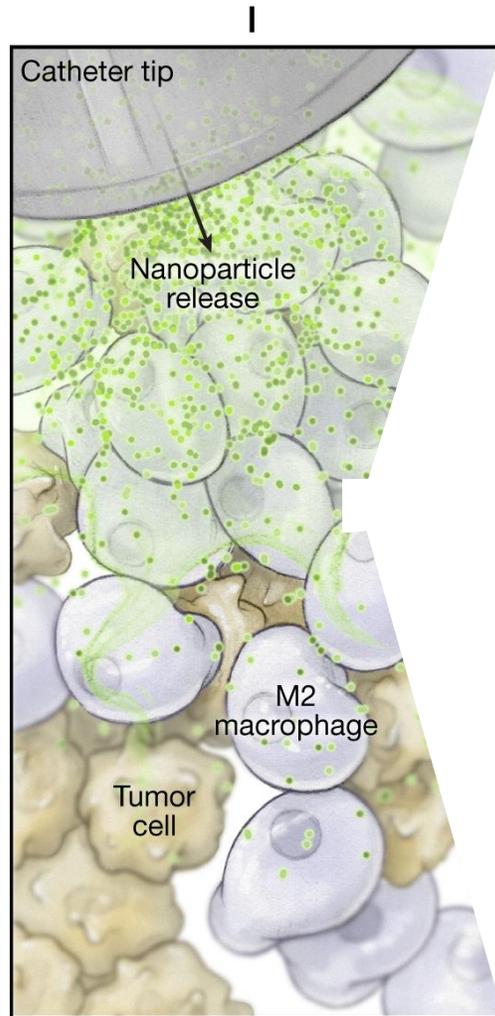
Interferon Regulatory Factor 5 (IRF5) Promotes Inflammatory Macrophage Polarization

Diseases with significant IRF5 upregulation
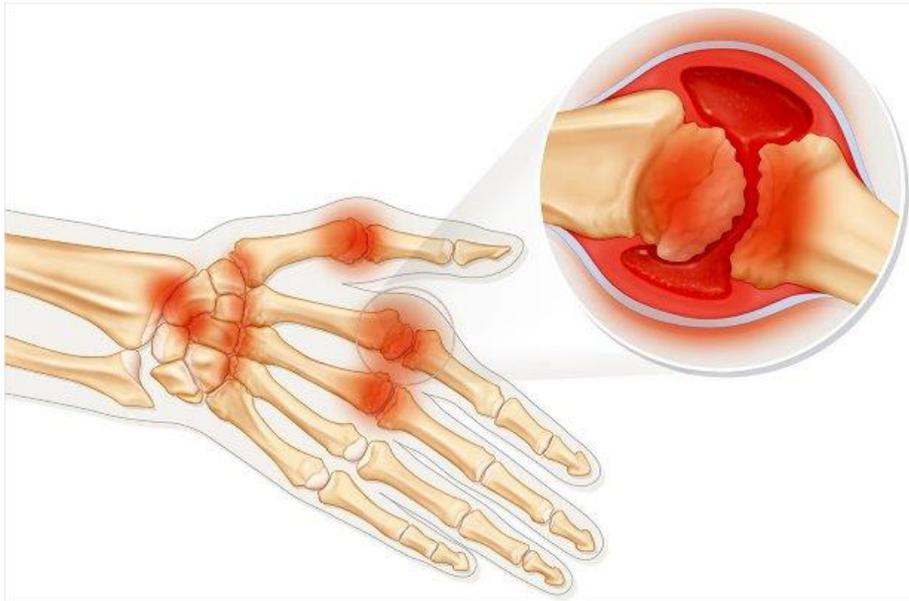

Rheumatoid arthritis
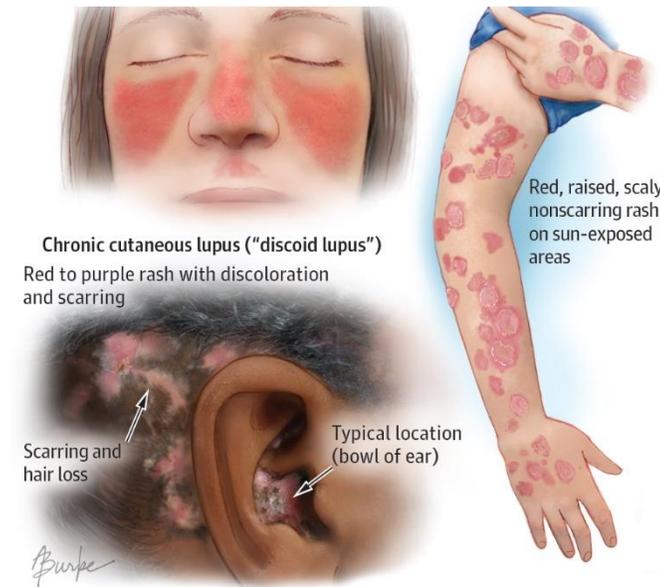
Lupus

# Apply nanotechnology to target-deliver mRNA-based therapeutics

Polymer

PGA

Targeting ligand (Dimannose)

IVT mRNA

ARCA Kozak Protein coding sequence α globin 3'UTR polyA

Pseudo-UTP 5-Methyl-CTP

G G A U C A

Transcription factor controlling macrophage phenotype

100 nm

500 nm
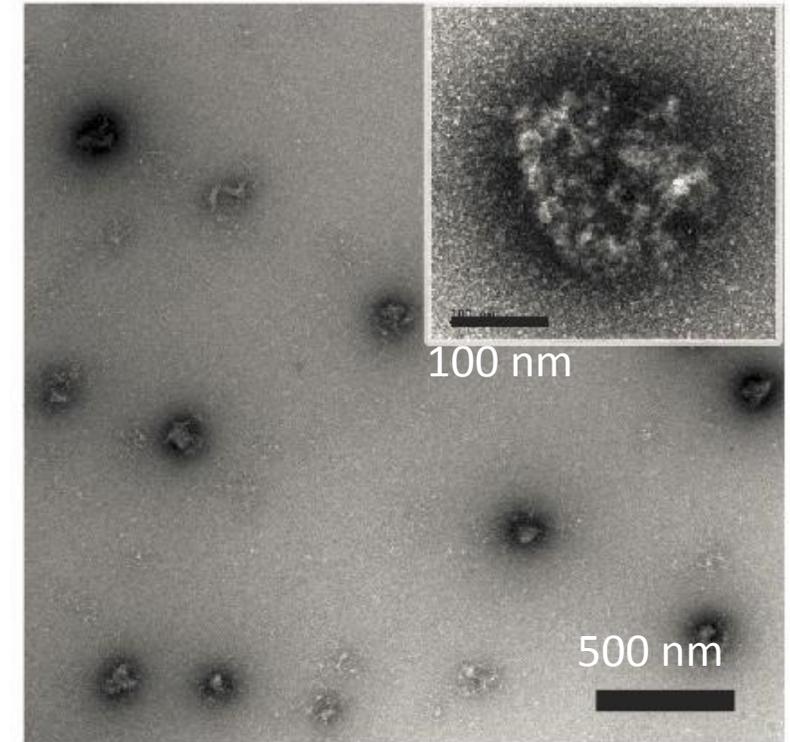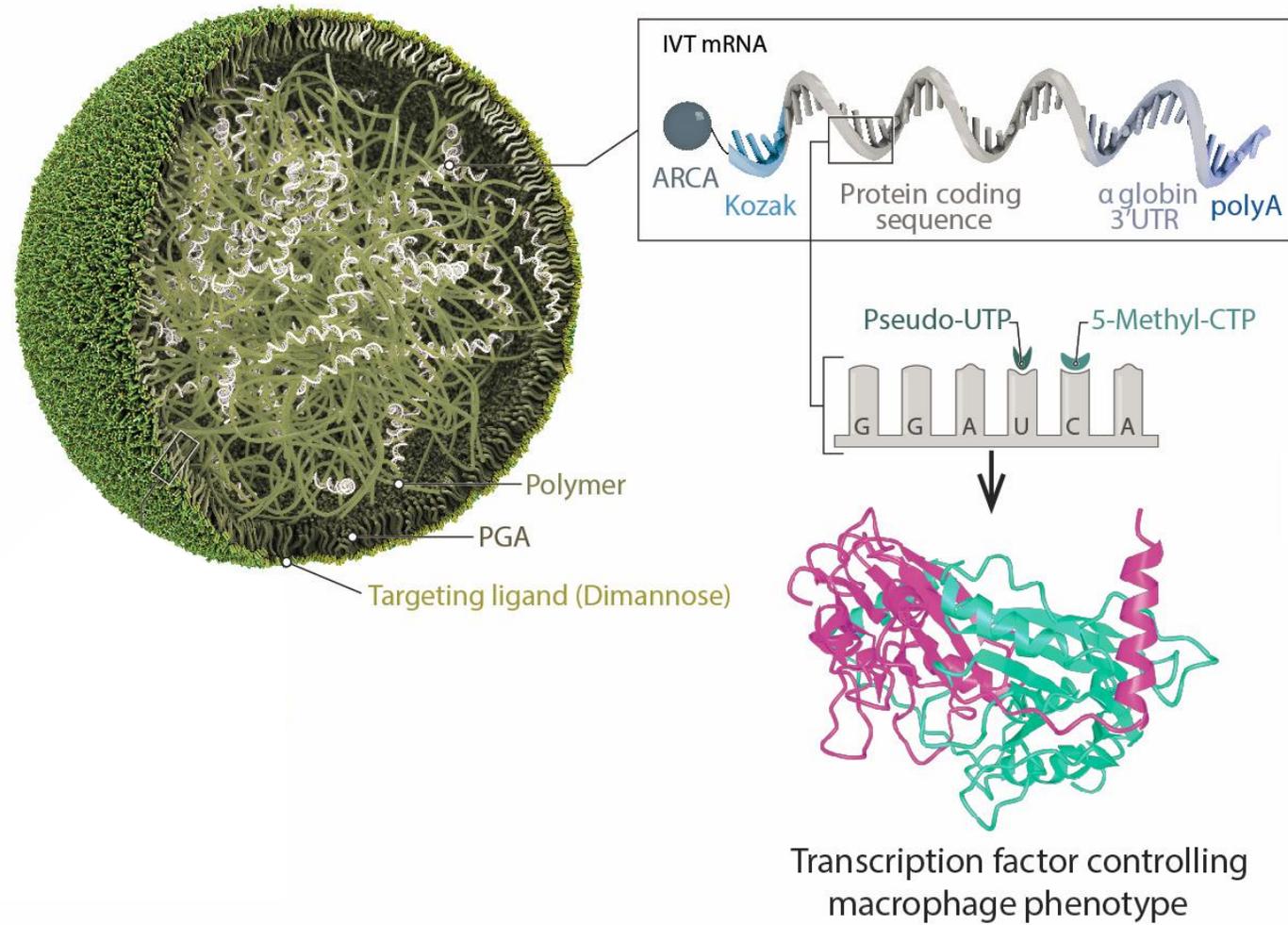
Nanoparticles program M1-like macrophage in vitro

- Immediate
- Transient
- High expression


M1-like macrophage signature genes
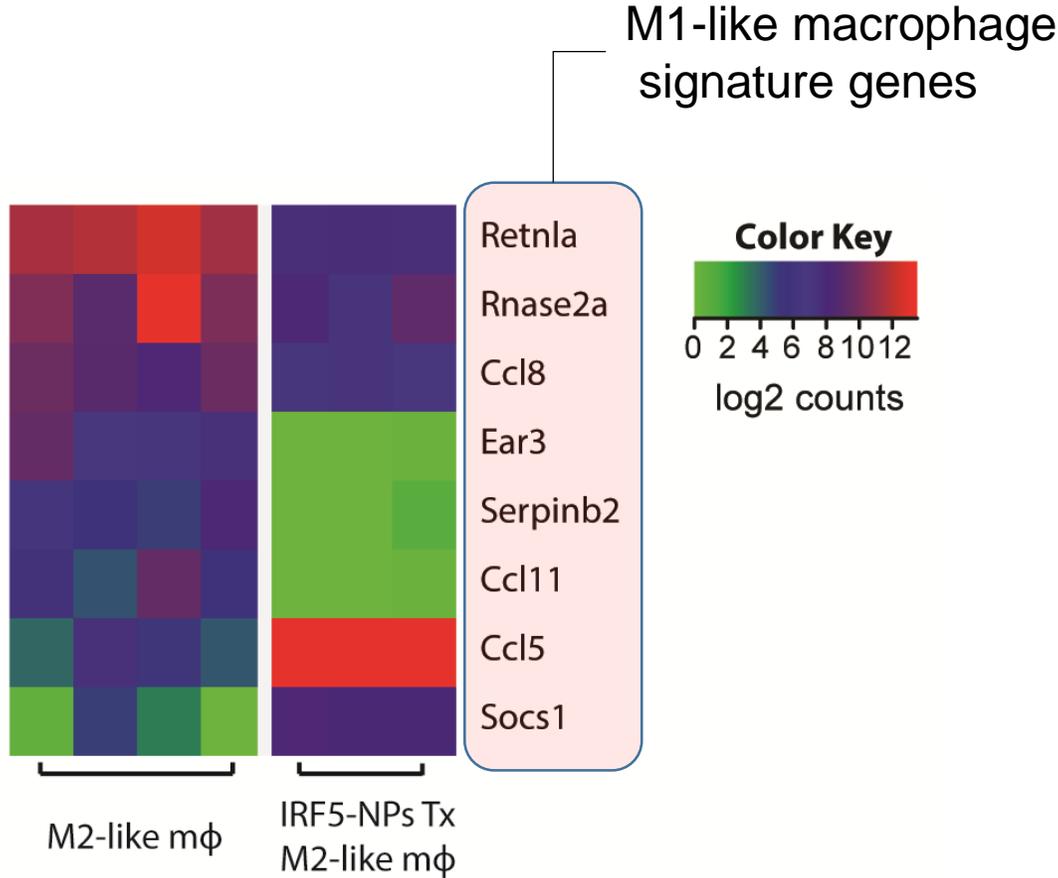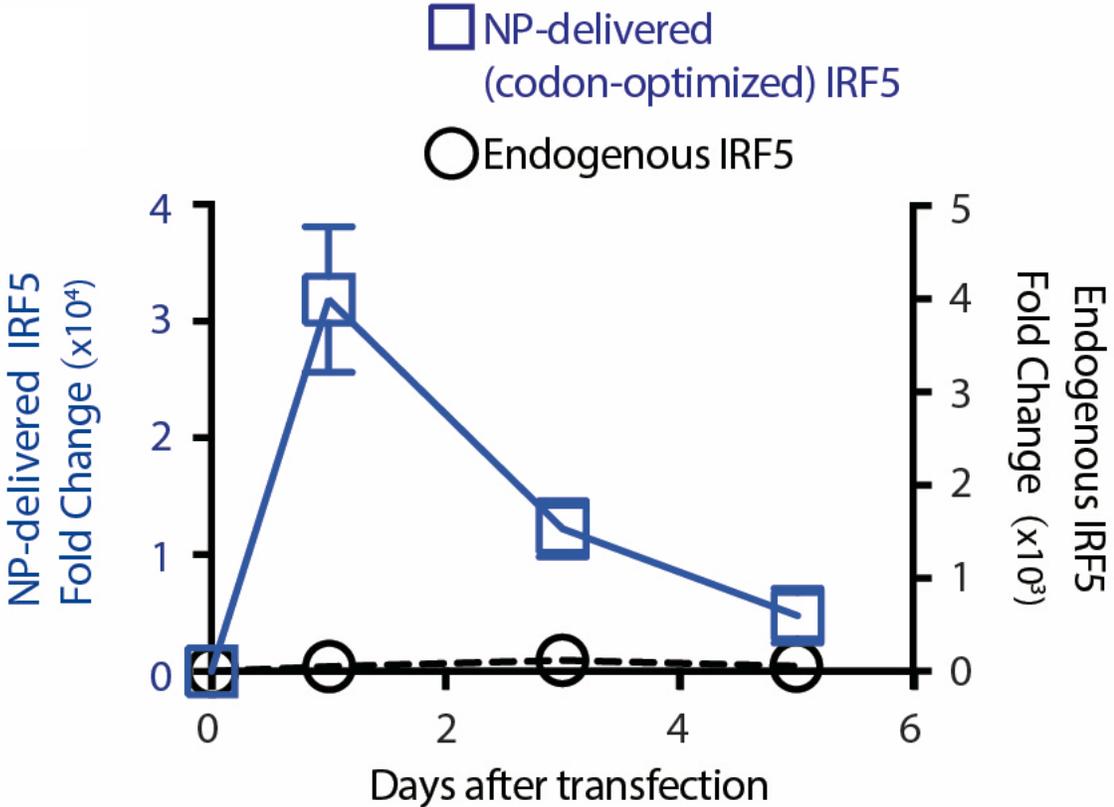
# Nanoparticles Target Macrophages in Ovarian Tumor through Mannose Receptor
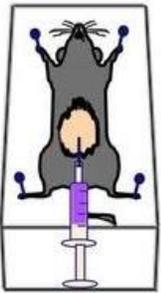
➤ A single i.p. injection of nanoparticles;

➤ Analyze the cells in the TME;

Mouse with advanced ovarian tumor
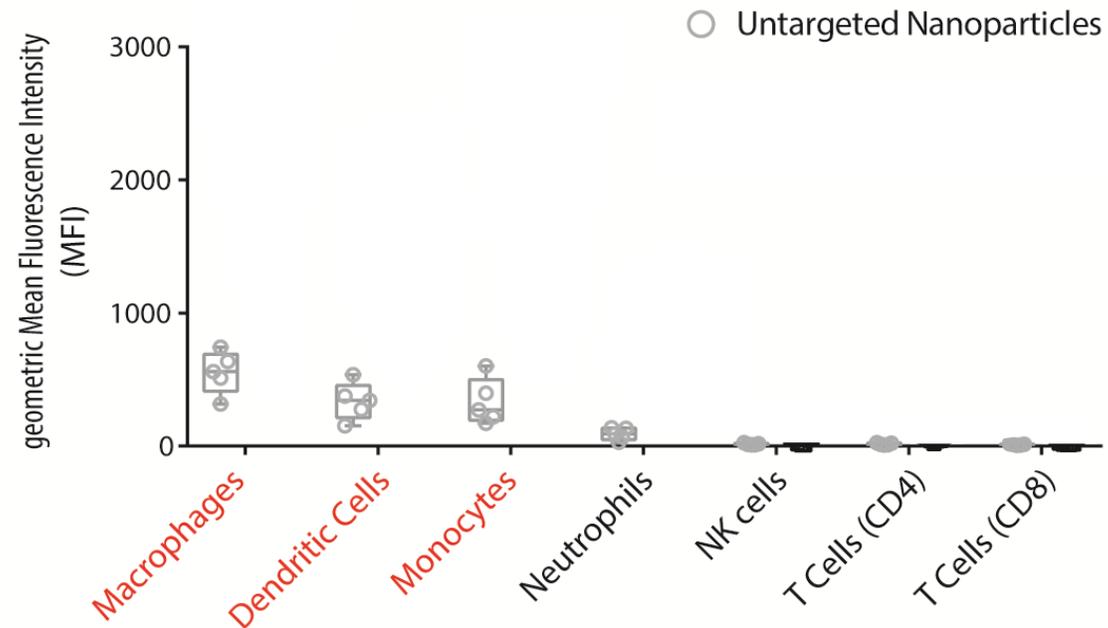
Nanoparticles regressed tumor growth in mouse model of ovarian cancer

PBS   eGFP-NPs   IRF5-NPs

D 14
D 19
D 26
D 40
D 84
D 131
D 152

Max
Min

I.P. injection of NPs for 9 **doses**,
2 doses/week, 50 µg/dose (mRNA-base)

Tumor inoculation

D 0   D 14 (Week 2)   D 77 (Week 11)
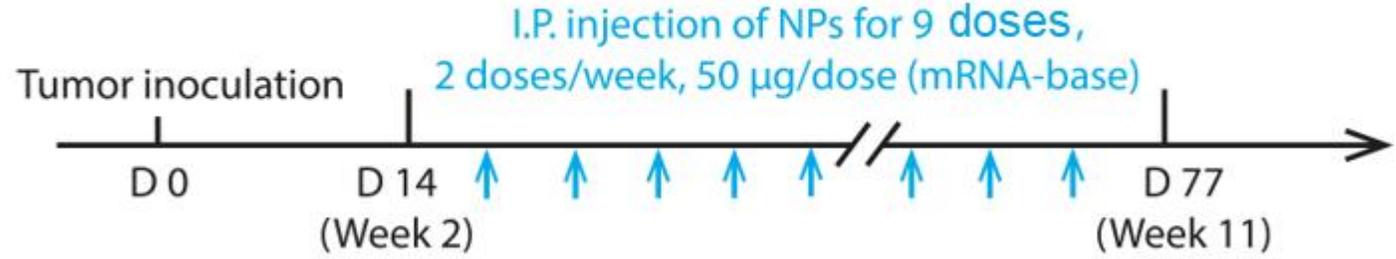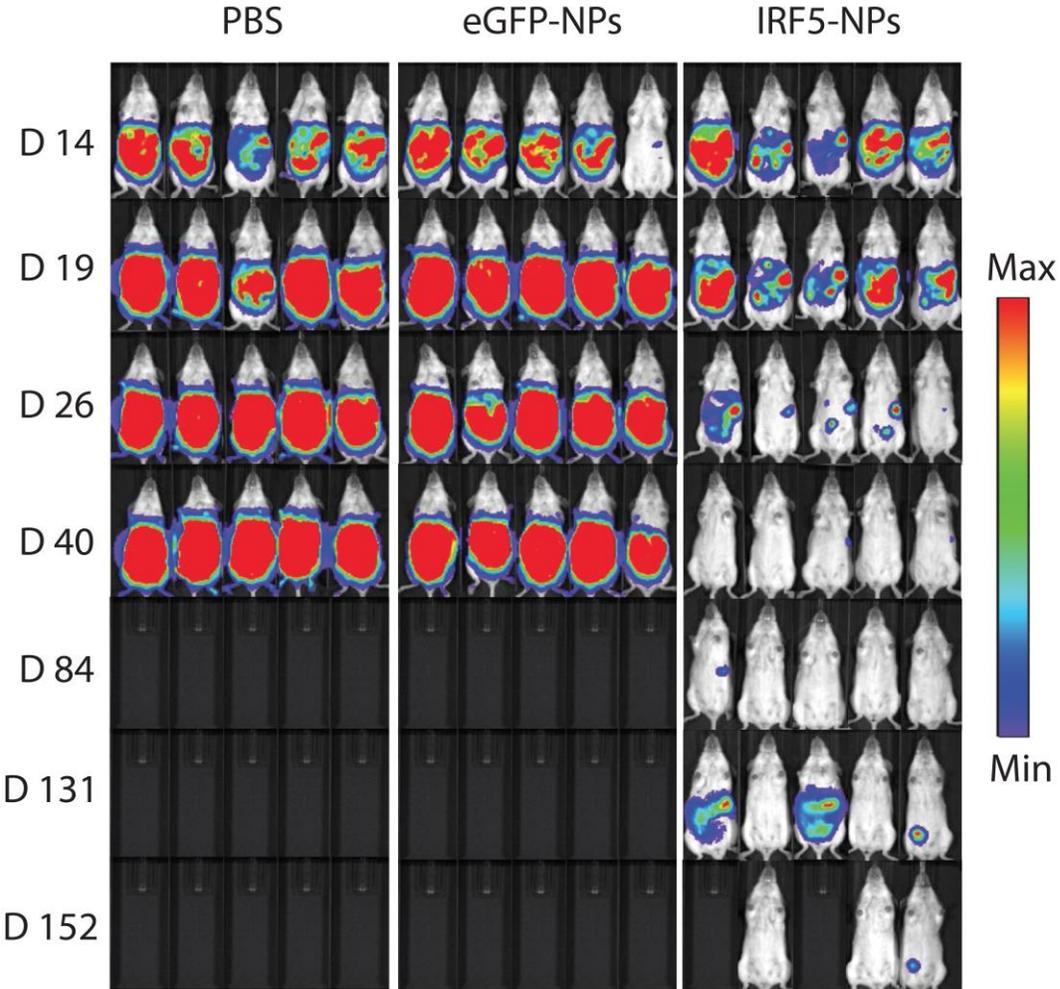
○ PBS, n=9 (ms=60)
△ eGFP-NPs, n=8 (ms=57.5)
✕ IRF5-NPs, n=10 (ms=141.5)

$p<0.0001$
$p<0.0001$

Percentage survival

100

50

0

0   50   100   150   200

Days after tumor inoculation

40%
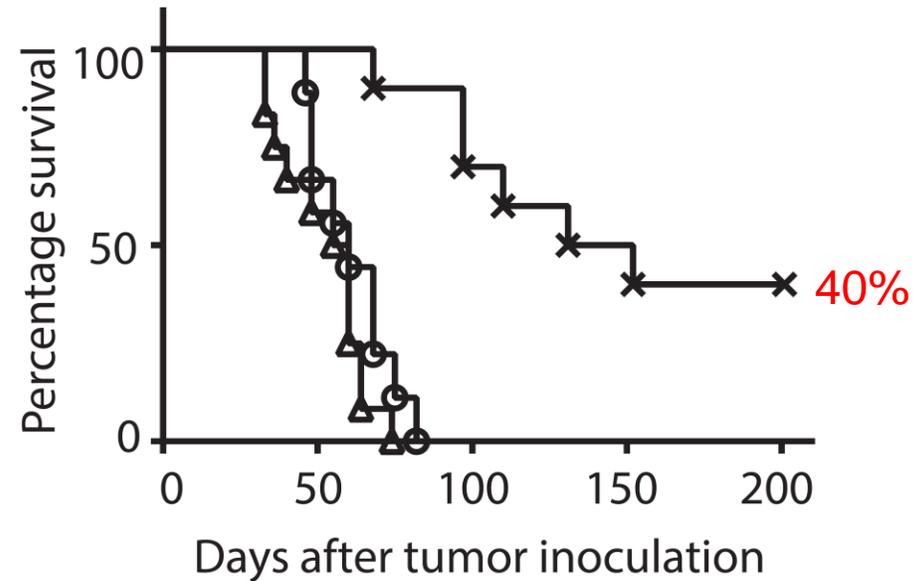
Nanoparticles Program M1-like Macrophages in Ovarian Tumor
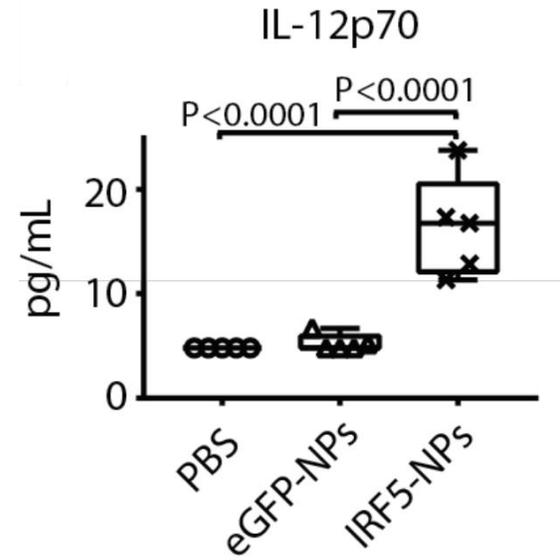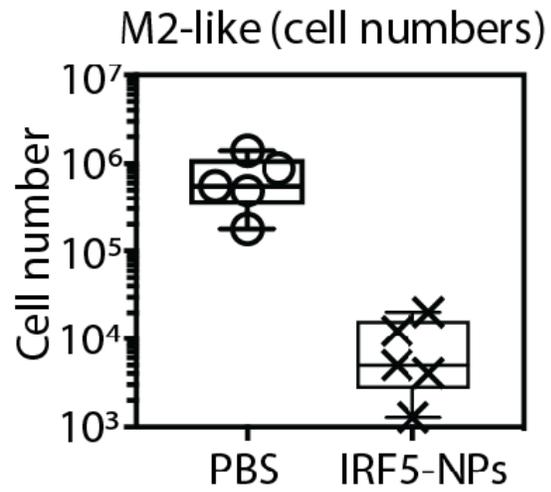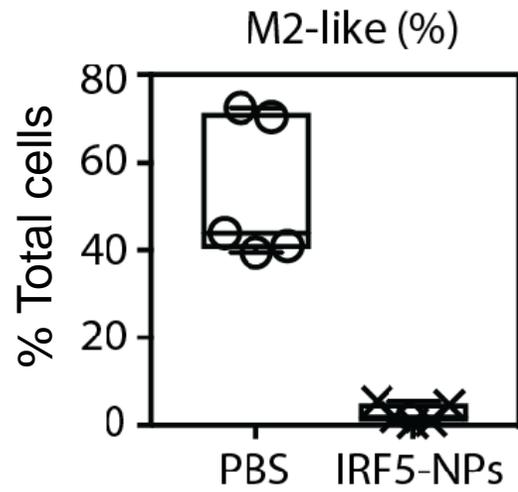
M1 (MHCII+, CD206-)   M2 (MHCII-, CD206+)

# Nanoparticles treatment induces T cell infiltration

PBS   IRF5-NPs

CK8

DAPI
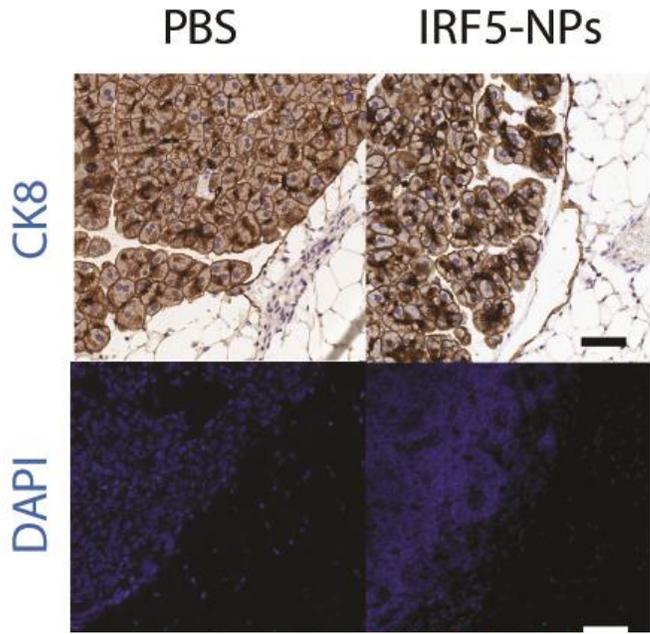
← Metastatic tumor nodules

The tumor-regressing effect of nanoparticles is partially mediated by the CD8+ T cells

Tumor burden

Average Radiance

9.0×10^{07}

6.0×10^{07}

3.0×10^{07}

0

PBS
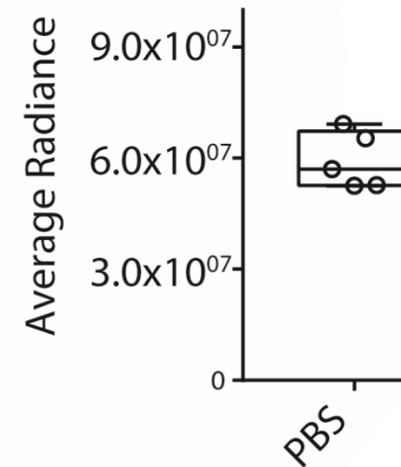
Scale bar: 100 μm

A second-line therapy for Stage IV ovarian cancer

mRNA encodes for transcription factor (TF)

Nanoparticle carrying encoded mRNA

Intra-peritoneal IV delivery

Metastatic tumors

Ovarian tumor mass
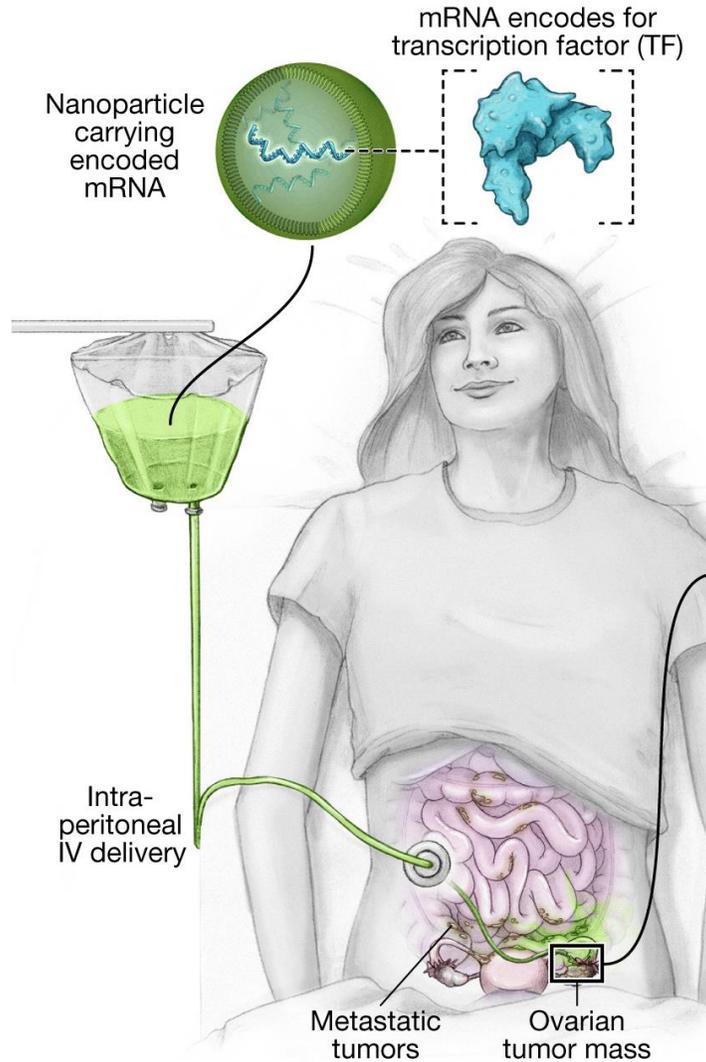
Summary

Infusions of nanoparticles formulated with mRNAs encoding interferon regulatory factor 5 reverse the immunosuppressive, tumor-supporting state of TAMs and reprogram them to a phenotype that induces anti-tumor immunity and promotes tumor regression.

Acknowledgement

Neha Parayath, Ph.D.
Michael Coon, Ph.D.
Sirkka Stephan, Ph.D.
Smitha Pankajavally Somanathan
Pillai, Ph.D.
Chibawanye Ene, M.D., Ph.D.
Eric Holland, M.D., Ph.D.
Histopathology Core Facility
Matthias Stephan, M.D., Ph.D.