Targeting Neuroblastoma's Weak Spots with GD2-Directed siRNA-Lipid Nanoparticles

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CRS 2025 ANNUAL MEETING & EXPOSITION
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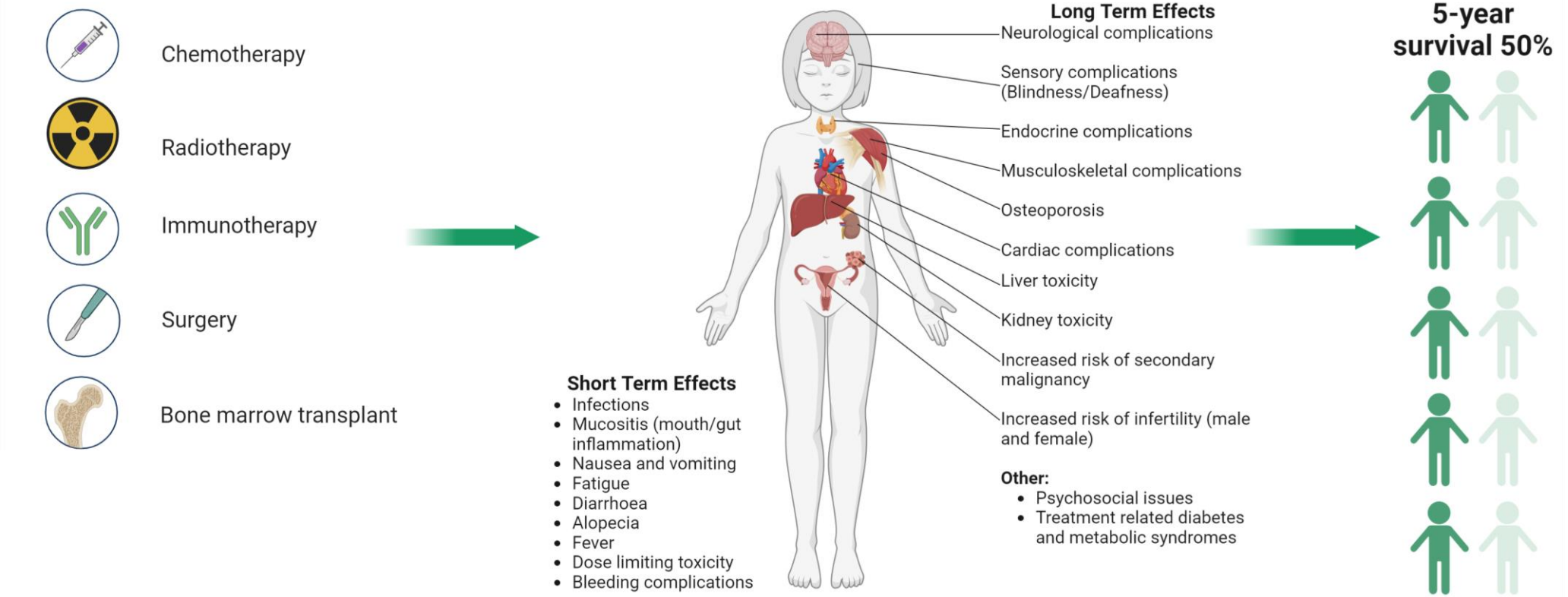
JULY 13-18, 2025

NEXT-GENERATION DELIVERY INNOVATIONS

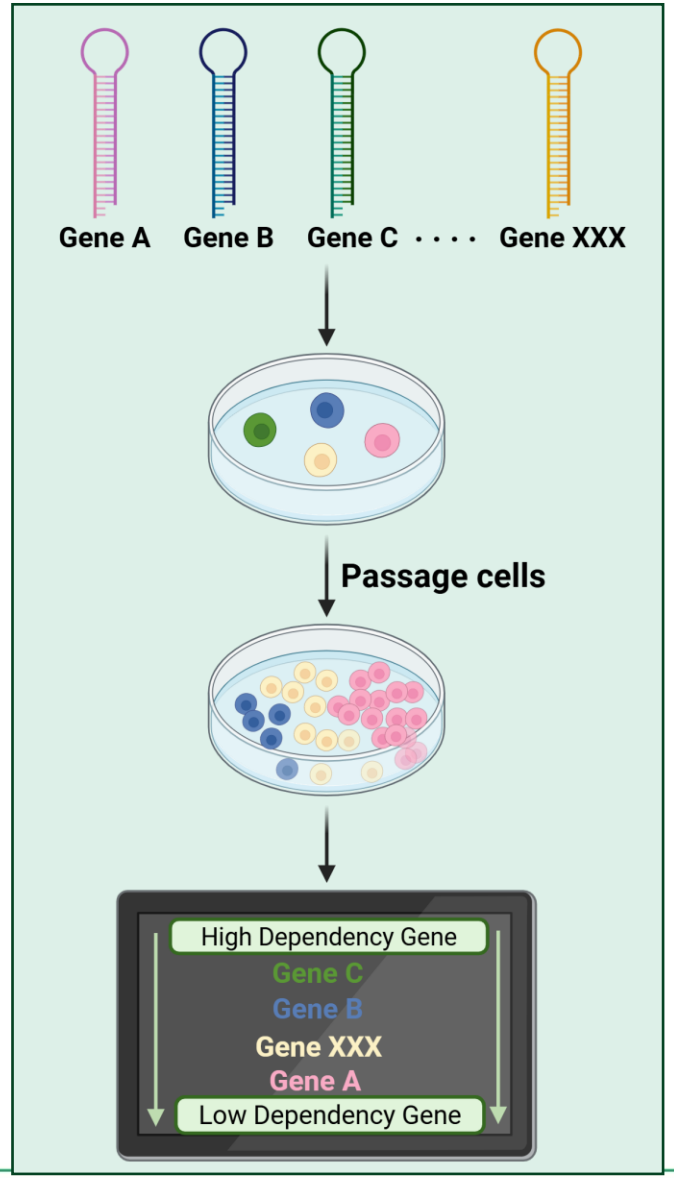
High-risk neuroblastoma

These factors have led to increased interest in developing treatments that can directly target genes associated with tumour growth and survival



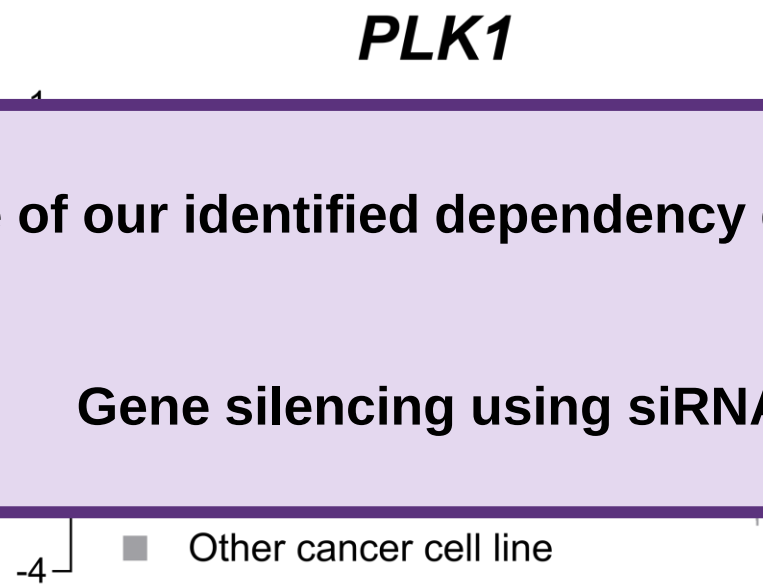
Created with Biorender.com

Identifying Genes Essential for Tumour Growth Through Dependency Mapping



Depmap.org/portal , McDonald, de Weck *et al., Cell* (2017).

Genome wide CRISPR Screening Identifies Potential Neuroblastoma Dependency Genes

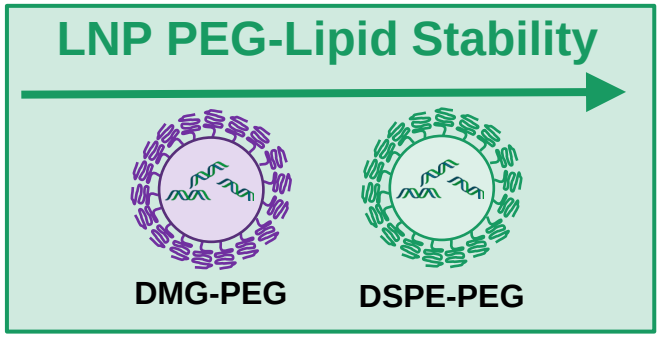


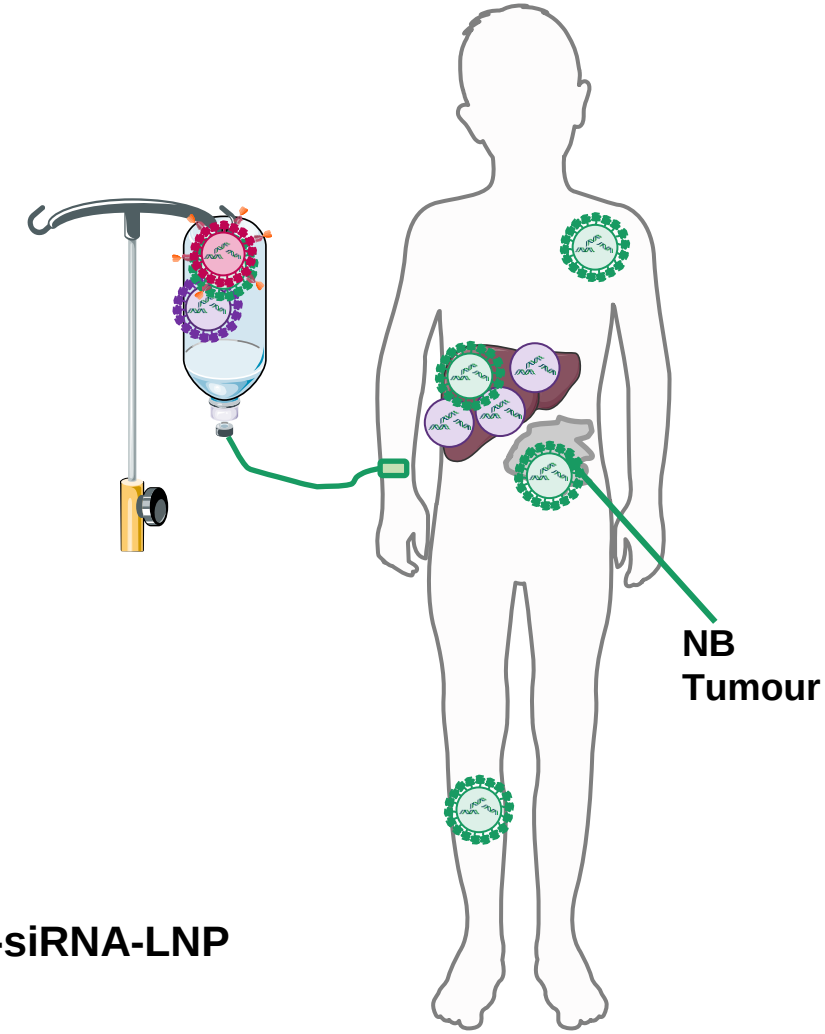
None of our identified dependency genes have drugs or inhibitors available for use, and most are considered undruggable.

Gene silencing using siRNA offers the opportunity to directly target these genes.

Tumour targeted LNPs to deliver siRNA to neuroblastoma

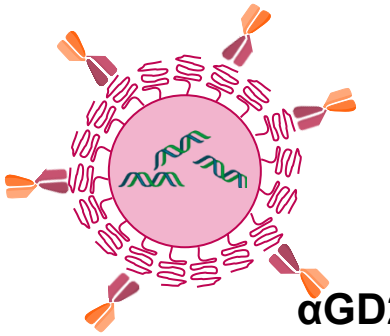
1. Changing PEG-lipids to increase LNP circulation times





2. Antibody targeting using GD2 Bispecific Antibody (αGD2)

Disialoganglioside GD2 binding domain
PEG binding domain

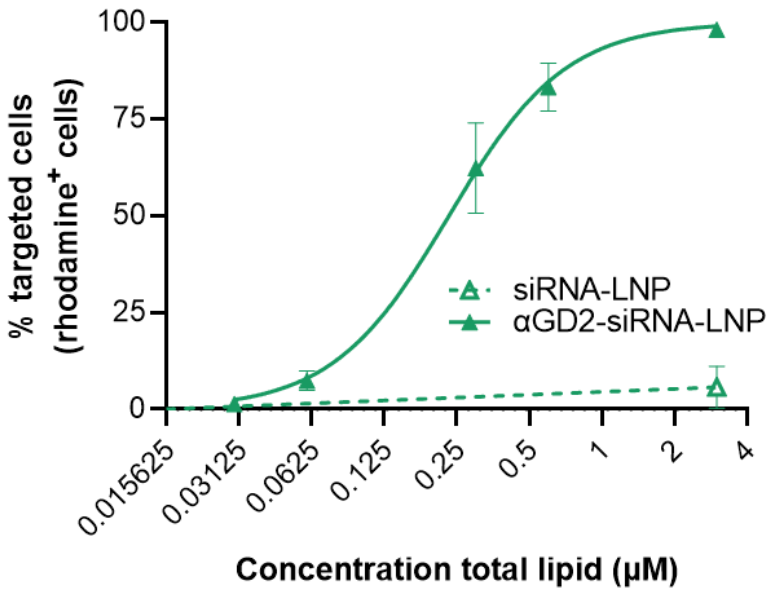


Disialoganglioside GD2 is an antigen expressed on the surface of neuroblastoma cells with **limited expression in normal tissues**

- Anti-GD2 immunotherapy part of standard care for high-risk neuroblastoma
- GD2 directed CAR T also in clinical trials
- Conventional GD2 therapeutics have side effects

αGD2 BsAb improves siRNA-LNP cell association and anti-cancer efficacy

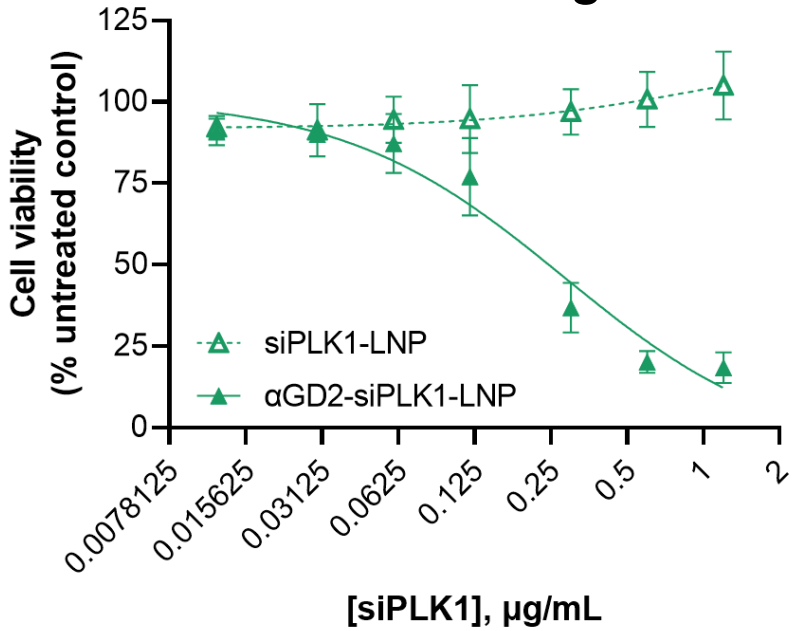
Cell Targeting



DSPE-PEG	EC_{50} (μM)
Untargeted siRNA-LNP	>3
αGD2-siRNA-LNP	0.23

LNP cell targeting in CHP-134 cells following 4-hour incubation.

Cell Killing

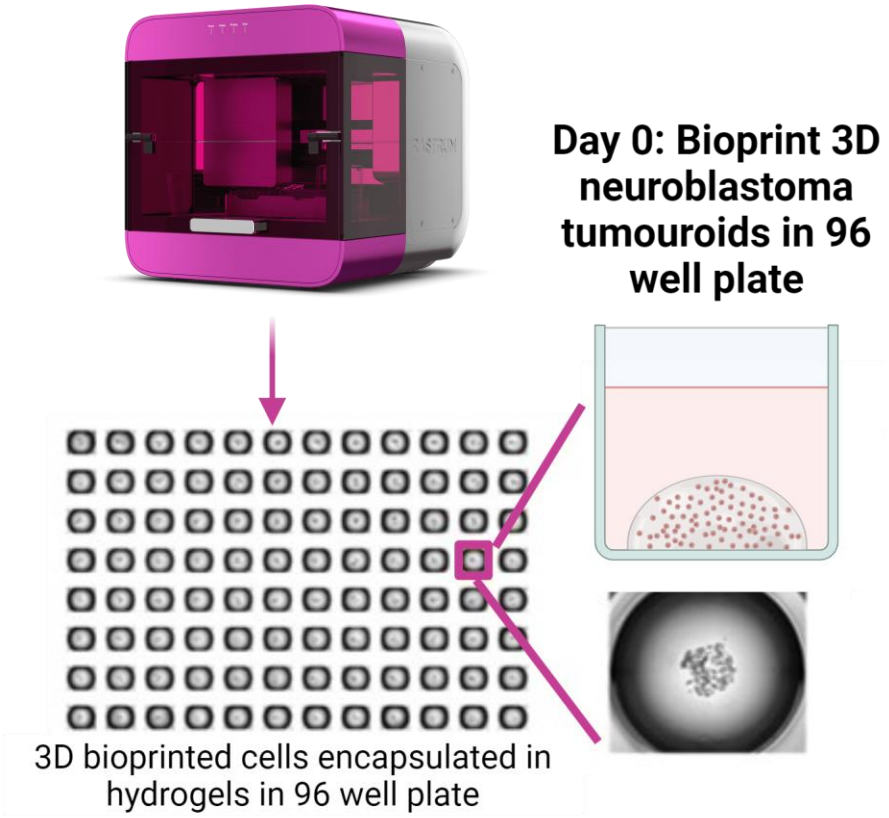


DSPE-PEG	IC_{50} (μg/mL)
Untargeted siRNA-LNP	>1.2
αGD2-siRNA-LNP	0.28

LNP anti-cancer efficacy (cell killing) in CHP-134 cells following 72-hour incubation.

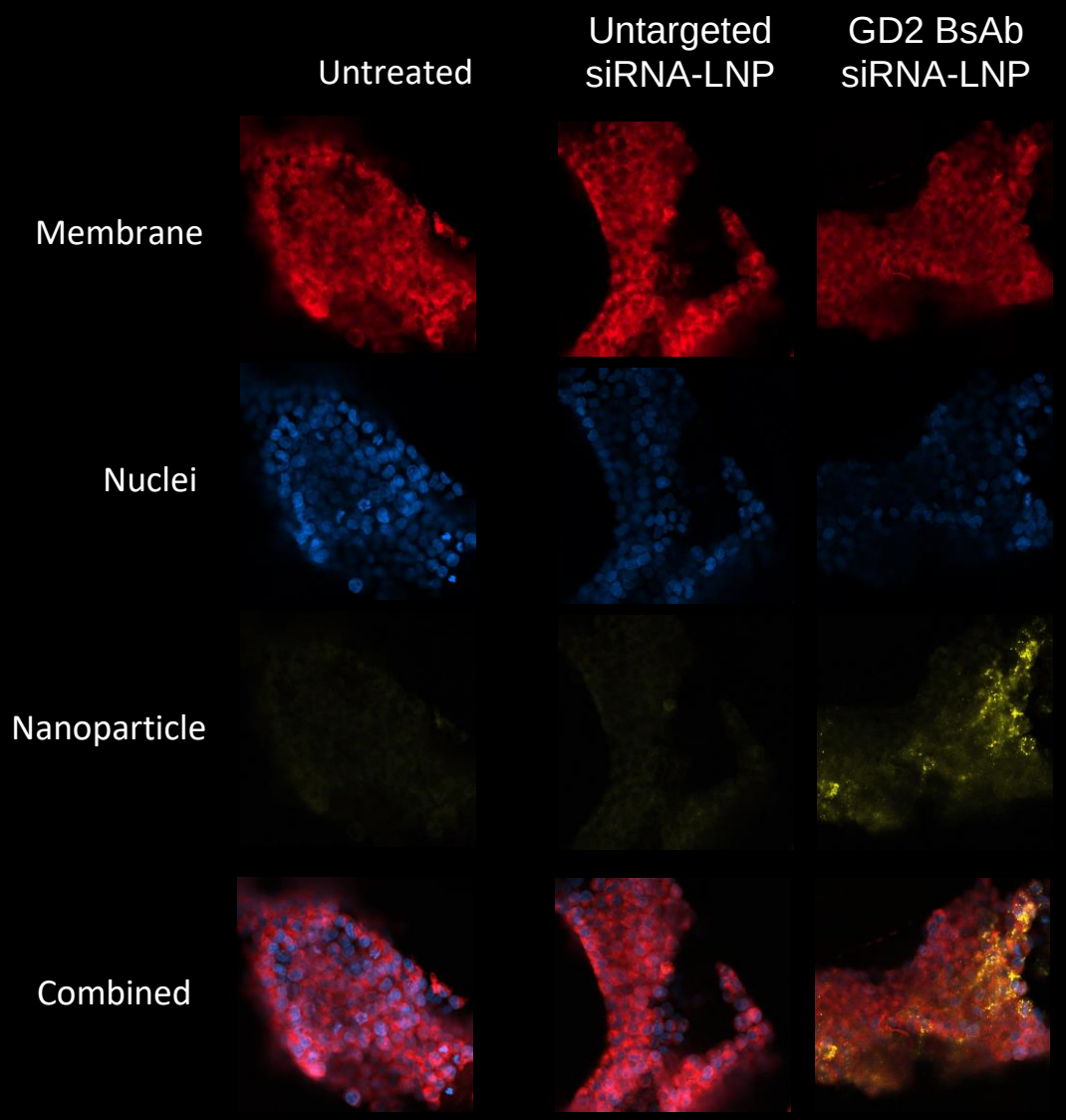
Unpublished data, please do not post.

Examining uptake of αGD2-siPLK1-LNPs in a 3D tumouroid model



Utama RH…..Kavallaris M, Gooding JJ. *Macromolecular Bioscience* 2021
Utama RH#, Lakmali A#.....Kavallaris M*, Gooding JJ*. *iScience* 2020

Nanoparticle Analysis in Bioprinted Samples – Uptake (8hr)



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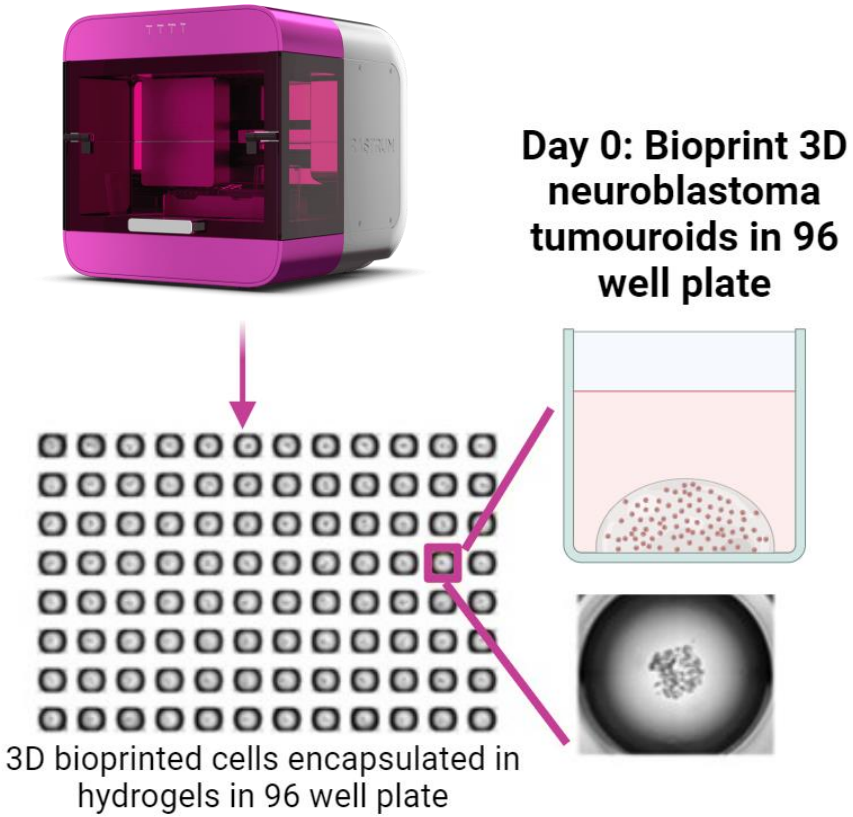
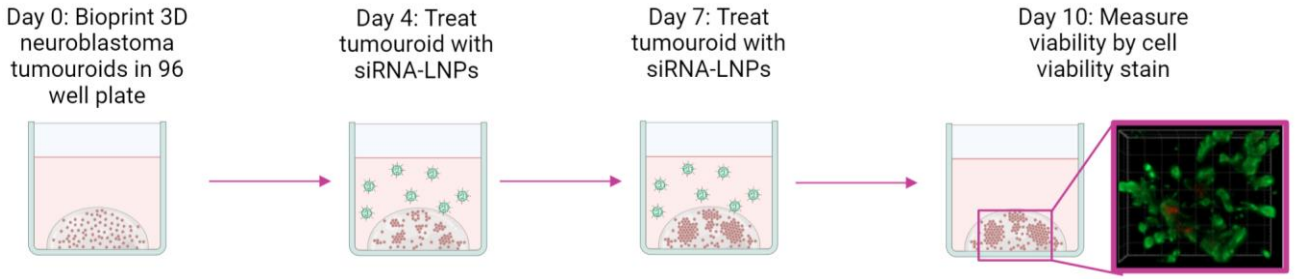
CHP-134 cells printed and cultured up to Day 7, then treated with LNPs for 8hrs. Samples were fixed and stained for confocal imaging.

Unpublished data, please do not post.

Examining effects of αGD2 in a 3D tumouroid model

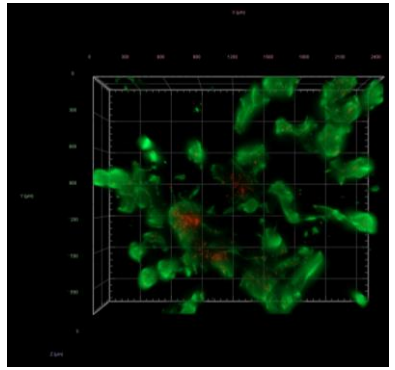


αGD2-siPLK1-LNPs reduce neuroblastoma viability in a 3D tumouroid model



DSPE-PEG

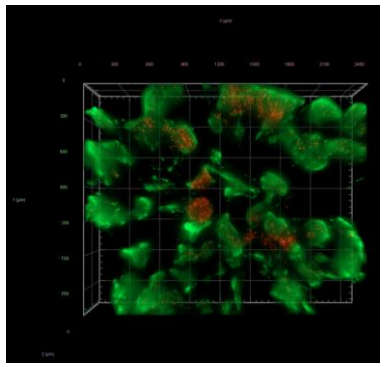
Untreated control

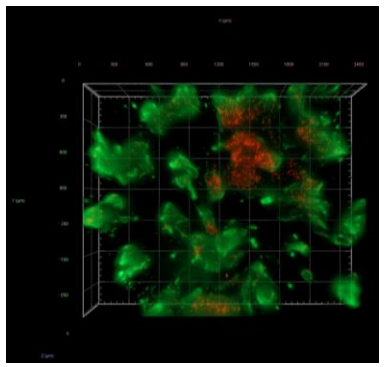


Live cells
Dead cells

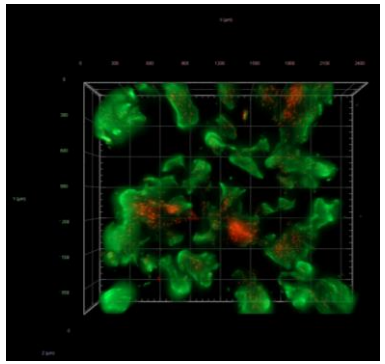
Untargeted LNP | **αGD2-targeted LNP**

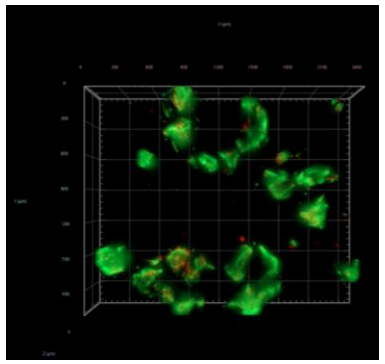
siCTRL

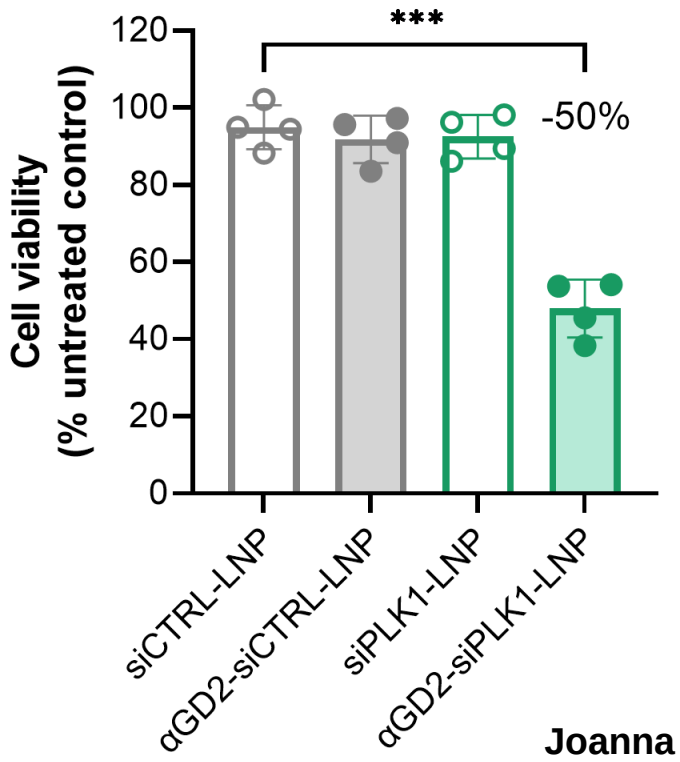




siPLK1







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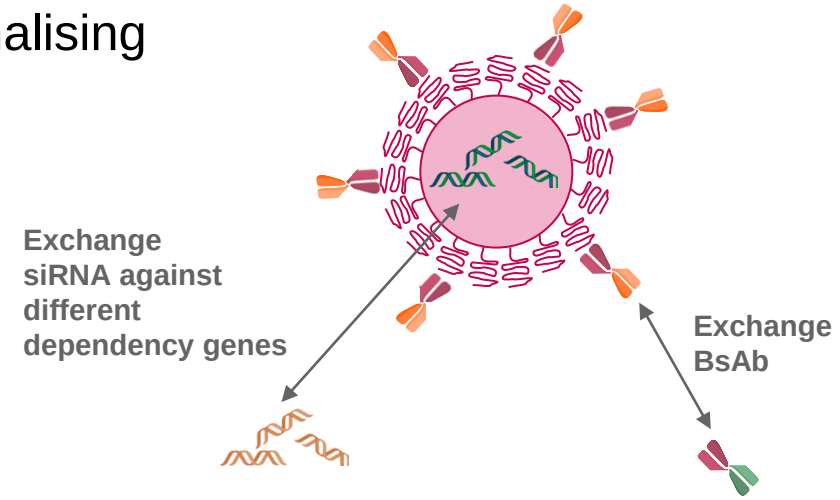
Figures representative of 2 independent experiments. Unpublished data, please do not post.

Conclusions and potential significance

- Cancer dependency genes are critical for tumour cell growth. siRNA offers a unique opportunity to target these genes that are often considered undruggable
- αGD2 bispecific antibodies improved uptake and anticancer effect of siRNA-LNPs in 2D and 3D neuroblastoma models
- Mix and match targeting approach that can be adapted to **target different tumour antigens and dependency genes** → personalising treatment



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PHILADELPHIA, PA
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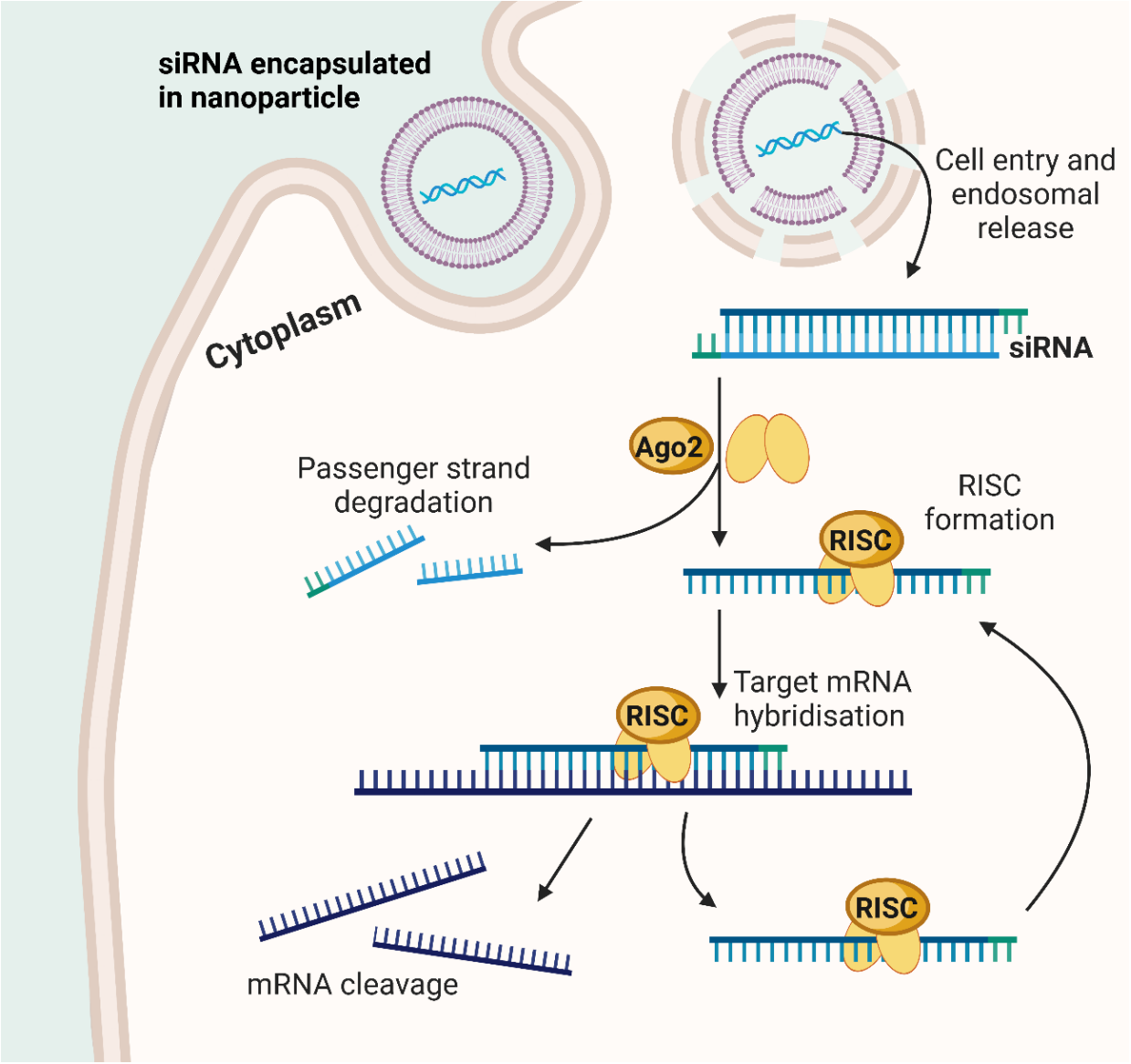
NEXT-GENERATION DELIVERY INNOVATIONS

siRNA: Targeting the Undruggable

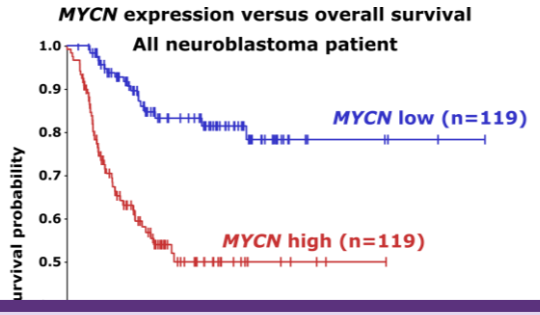
None of our identified dependency genes have drugs or inhibitors available. Many are involved in transcription, and are considered undruggable.

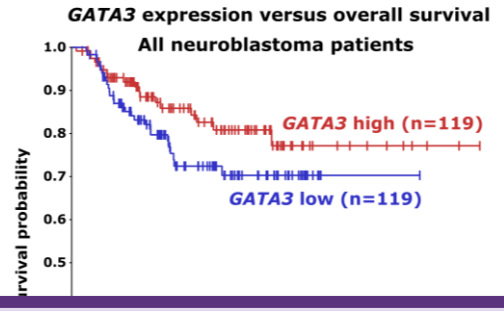
Benefits of siRNA:

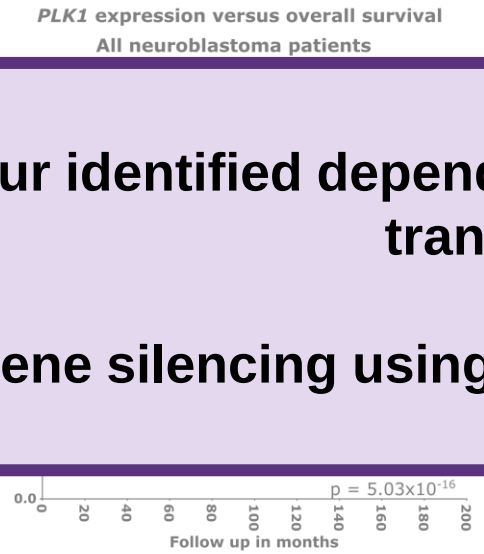
- Higher target specificity, reduced off-target effects.
- Can be used to target proteins that are considered undruggable with inhibitors.



Dependency Genes and Survival in Neuroblastoma







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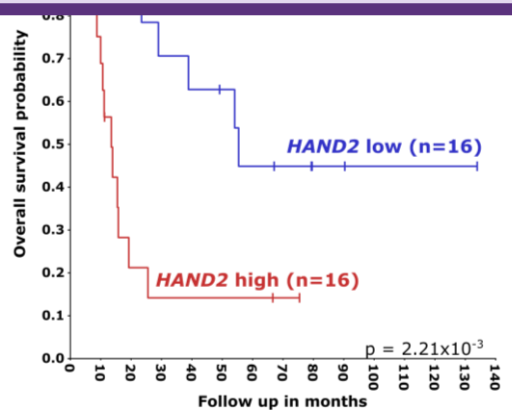

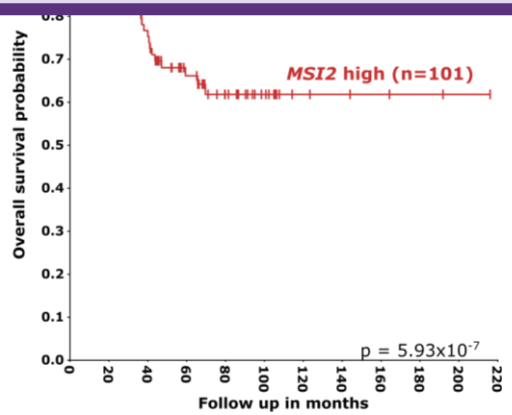

Kaplan-Meier analysis comparing survival in 1st vs last quartile by gene expression. Analysis conducted in R2 using Kocak neuroblastoma dataset.

Destiny Tsai