MGMR® Versatile Nano-carrier for Non-viral Cellular Transfection

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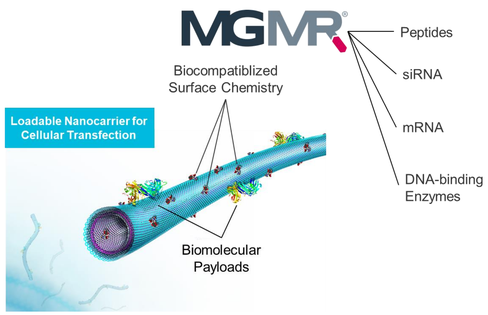
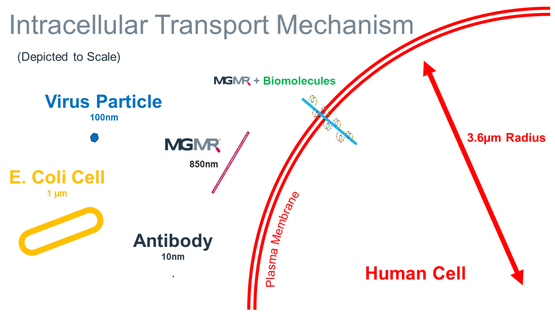


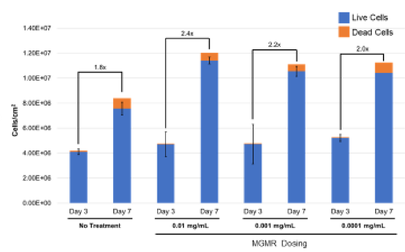
Intracellular Transport Mechanism



MGMR® Supports T-cell Proliferation

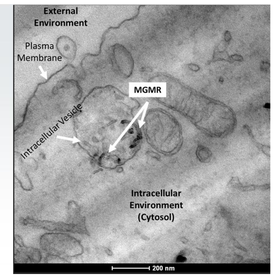
Dose-response Testing for Proliferation of Human T Lymphocyte-like cells (Jurkat)

- MGMR does not reduce cell proliferation relative to control (Growth Media, No Treatment)
- All groups indicated high viability
- Certain dosing-ranges of MGMR may improve cell proliferation and increase yield
- Protocol
  - Groups incubated with various concentrations of MGMR for 3 and 7 Days
  - Control = Standard Growth Media (RPMI 1640 + 15% FBS)
  - Cellular viability and quantification assessed by trypan exclusion assay
  - Stats: 95% CI



Endocytotic Trafficking

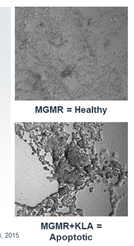
- MGMR is transported into the cell by active transport
  - *Unlike other transport systems, MGMR does not damage the cell's external casing (plasma membrane) in order to enter inside*
- MGMR is initially encapsulated into larger compartments called vesicles - that traffic into the interior of the cell
  - *MGMR is cargo agnostic - it is not prevented from getting inside cells by the size of its molecular cargo*
- Once internalized, MGMR is released from vesicles into the intracellular environment
  - *MGMR can deliver cargo to many different locations inside the cell*

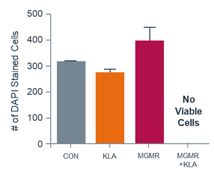


MGMR® Peptide Delivery

MGMR demonstrated to enhance cell penetration of KLA—a cell "impermeable" 1.5kD protein[1,2]

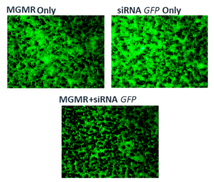
- Proteins are powerful cell-regulators but very challenging for intracellular delivery
  - Do not efficiently cross the cell's membrane
  - Protein activity is very sensitive and easily diminished
- In this case study: MGMR binds to KLA with high affinity, transports it across the plasma membrane, and releases it—triggering apoptosis
  - MGMR+KLA peptide produced absolute apoptosis
  - MGMR alone has no effect (high tolerability)
  - KLA alone has no effect (benign externally)



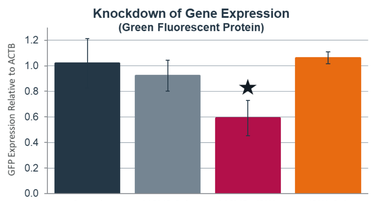


[1]Zurita et al. Cancer Research 64, 435-439, 2004 [2]Yang et al. Methods Mol. Biol. 1266, 29-53, 2015

MGMR® siRNA Delivery



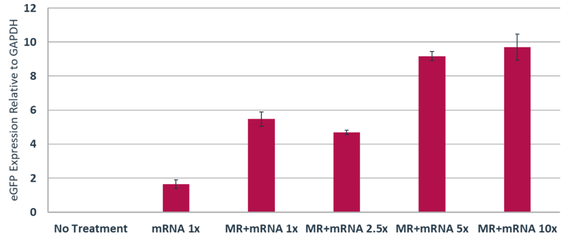
More Dark Space = More Knockdown of Green Fluorescent Protein



- Unlike detergent-based delivery systems, MGMR is non-cytotoxic and compatible across many types of molecular cargo

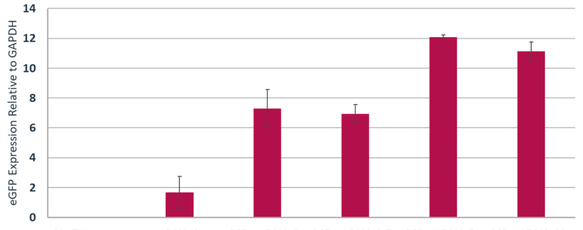
MGMR® mRNA Delivery

mRNA Delivery to T Lymphocytes (Jurkat)



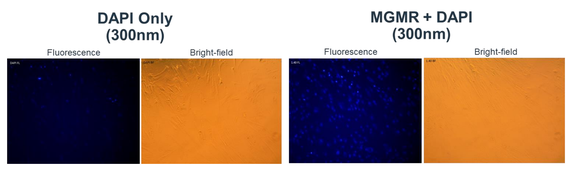
- 72-hour incubation • 1x = 500ng/mL • n = 3

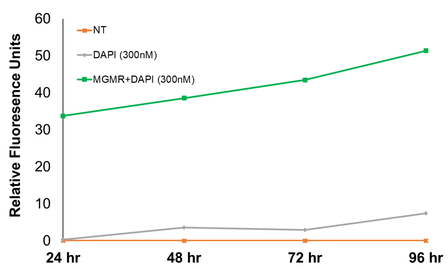
mRNA Delivery to Monocyte-like Cells (U937)



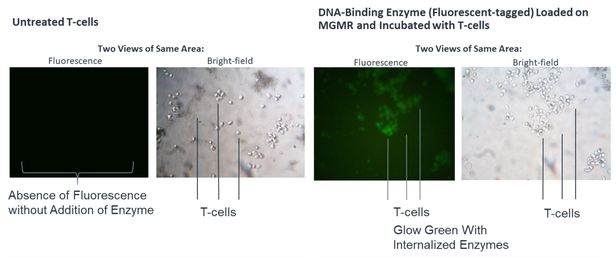
- 72-hour incubation • 1x = 500ng/mL • n = 3

MGMR® Nuclear Staining in Mesenchymal Stem Cells





MGMR® Delivery DNA-binding Enzymes in T-cells



Cells Not Treated with DNA-binding Enzymes Do Not Fluoresce

MGMR Transports DNA-binding Enzymes into Cells

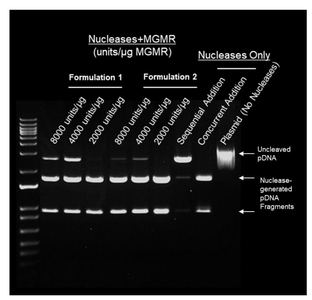
MGMR® Endonuclease-loading for Sequential DNA Cleavage

Protocol

- ScaI+MGMR incubated with 1ug plasmid for 15 min at 37°C, followed by HincII+MGMR for additional 15 min at 37°C
- Electrophoresis (70 min) - 1% agarose

Results

- Individual nucleases loaded on MGMR can cleave pDNA in series
- MGMR preserves nuclease activity and demonstrates loading density-dependent pDNA cleavage
- Sequential addition of "free" nucleases without MGMR produces very low-efficiency pDNA cleavage
- Formulation 2 demonstrated greatest efficiency for serial plasmid cleavage relative to Formulation 1 and sequentially added free nucleases



MGMR® Next-generation Transfection Technology

Cargo	MGMR™	Viral Vectors	Polymers	Liposomes
Small Molecules	+	-	+	0
Peptides	++	+	+	0
Nucleic Acids	++	++	+	+
Proteins	++	+	+	0
Lipopolysaccharides	++	-	+	0
Log(P) Independent	+	+	-	-
Properties				
Non-immunogenic	+	-	+	+
Non-GMO	+	-	+	+
Simple Optmization	++	-	0	-
Scalable	++	+	+	-
Eukaryotes	+	-	0	0
Prokaryotes	+	-	0	-
Plants	+	-	0	-

Key	
++	Advantage
+	Compatible
0	Limited Compatibility
-	Non-Compatible

How can MGMR® improve cell engineering?

- **Eliminates** viral-mediated transfection
  - No risk of insertional mutagenesis
  - No viral toxicity and immunogenicity
  - Non-replicative
- **Streamlines** manufacturing and quality processes
  - Non-biological transfection technology reduces variation
  - Readily extractable from final cell product
  - Eliminates need for new vector development – Single transfection vehicle for all cellular gene-editing systems
- **Versatile** loading profile effective for intracellular delivery of:
  - Gene-editing complexes
  - Oligonucleotides
  - Peptides/Proteins
- **Improves** cell yield
  - Reduced toxicity vs. viral or detergent-mediated transfection
  - Improved cell proliferation

MGMR® Discover its Value in Your Process

Seamlessly Combine MGMR® with your Technology in 3 Phases

1 Send Biopact Your Payload Molecule
- Provide Biopact with your gene-editing technology:
  - CRISPR
  - TALEN
  - ZfN
- Biopact **loads MGMR** with your payload at **No Cost**

2 Biopact Evaluates Performance
- Biopact will determines:
  - Loading efficiency on MGMR
  - Transfection efficiency in your target cell
- Biopact **reports performance of MGMR** to you at **No Cost**

3 Put MGMR® to the Test
- Order MGMR loaded with your molecule ($10k-$20k)
- Readily sufficient for *in vitro*, *ex vivo* or *in vivo* testing
- Evaluate MGMR loaded with your payload in your own studies

biopact™