Controlling *in vivo* drug transport with pharmaceutical nanotechnology

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Nanomerics Ltd

Overview

- Aims and tools
- Gene delivery to the bladder
- Brain delivery
  - Gene delivery to the brain
- Commercialization
  - Envelta™
  - Ocular penetration enhancer
- Summary

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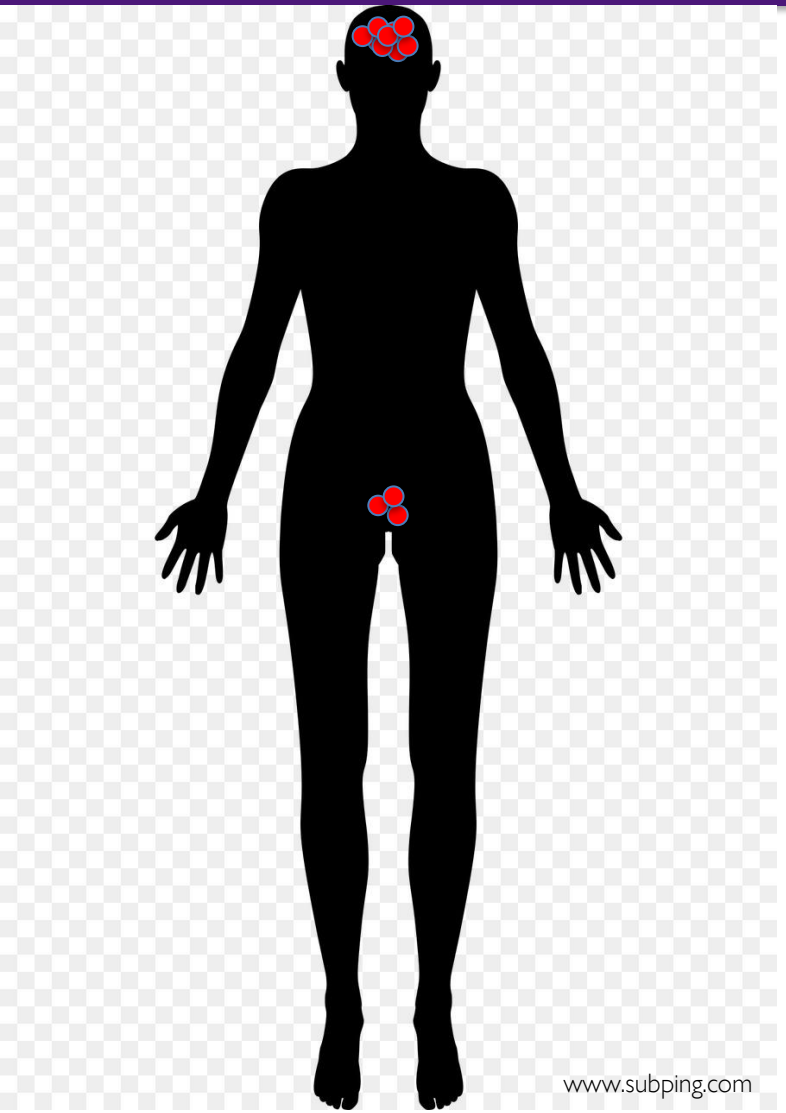
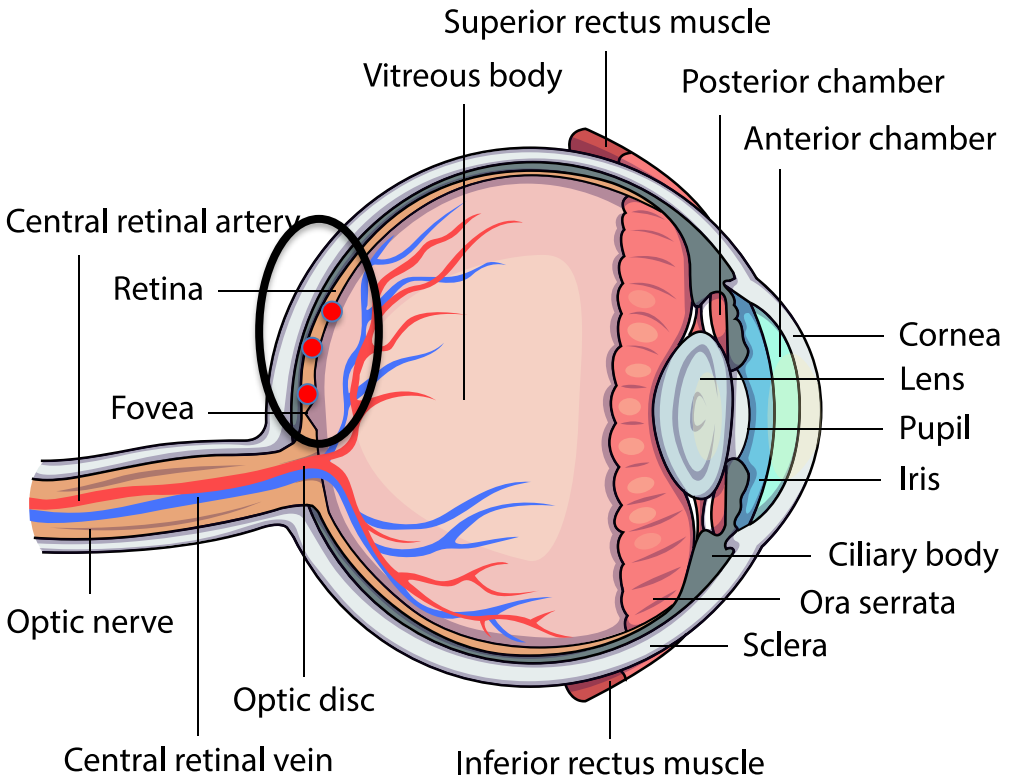
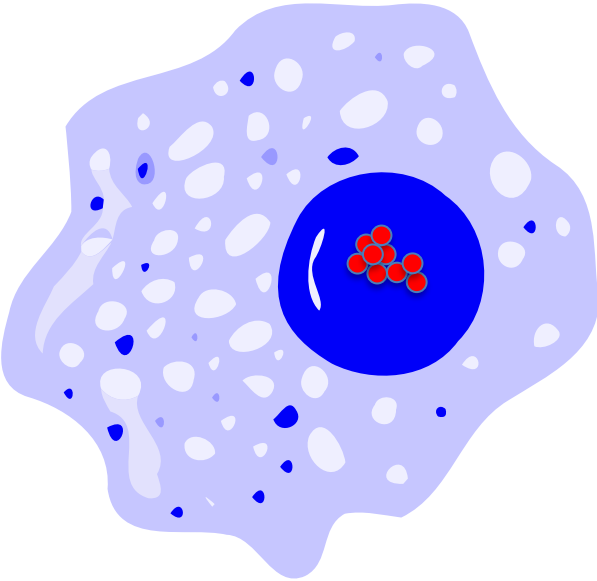
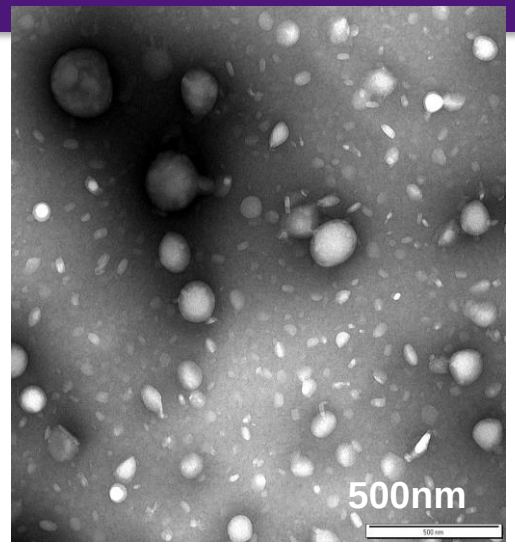
Control drug transport



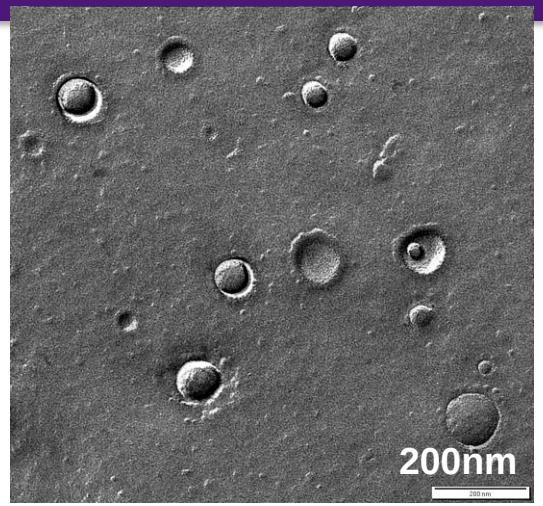




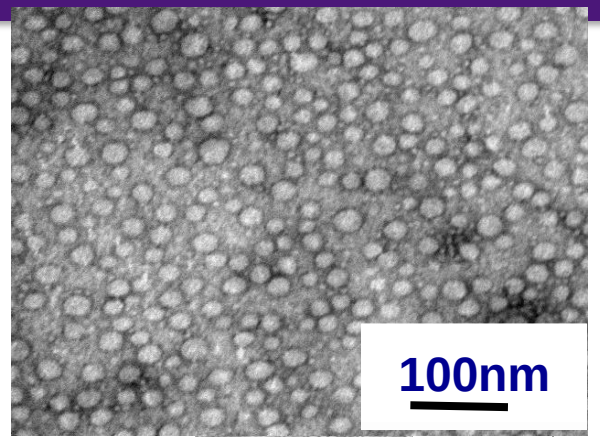
Pharmaceutical nanoparticles - tools to control drug transport



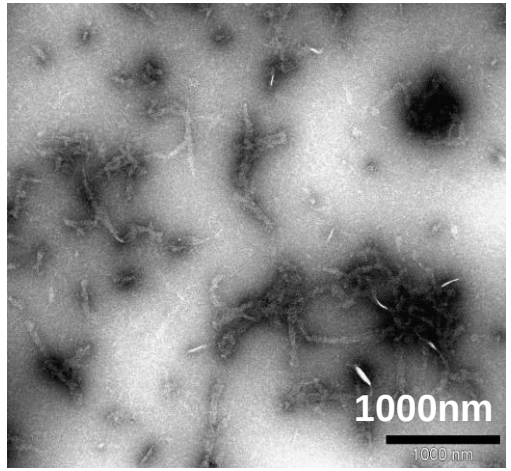
Dense Spheres



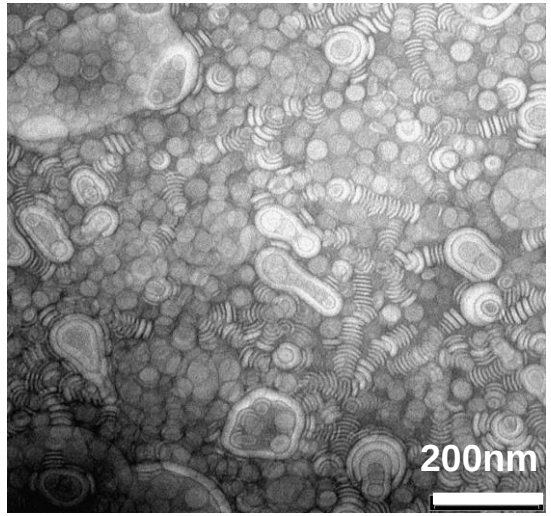
Vesicles



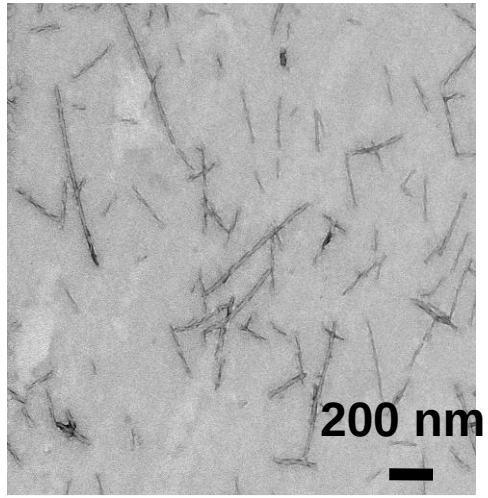
Polymeric Micelles



Worm-like



Discs



Peptide Nanofibres

Wang et al (2004) Macromolecules, 37: 9114 - 9122.
Qu et al (2008) Langmuir, 24: 9997–10004
Lalatsa et al (2012) J Control Release, 161: 523 – 536.
Mazza et al (2013) ACS Nano, 7: 1016-1026

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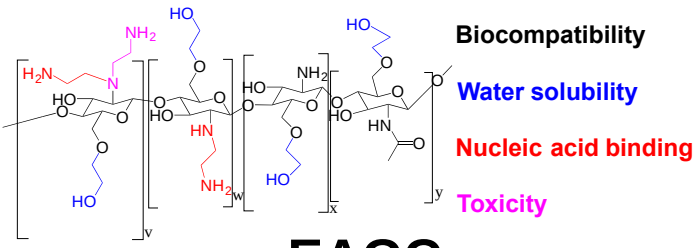
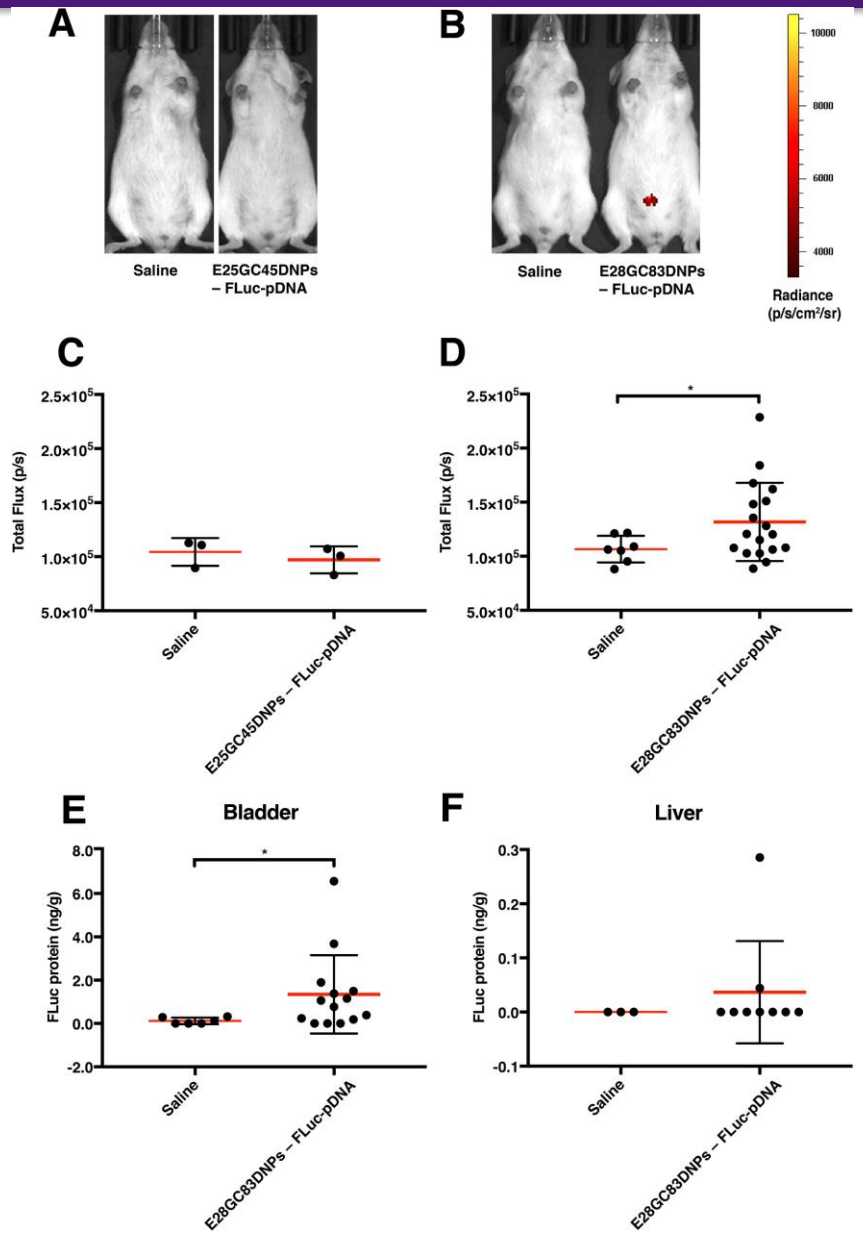
Delivering genes to a voiding bladder



EAGC

EAGC IC50 = 0.4 – 4.1 mg mL^{-1}
Lipofectamine IC50 = 0.03 mg mL^{-1}



On bladder instillation, a high molecular weight polymer (83 kDa) delivers genes to a voiding bladder

Gene expression contained in the bladder, e.g. no liver gene expression

E28GC45 – Molecular weight = 45 kDa
E28GC83 – Molecular weight = 83 kDa

Li et al (2021) J Control Release, 332, 210 -224.

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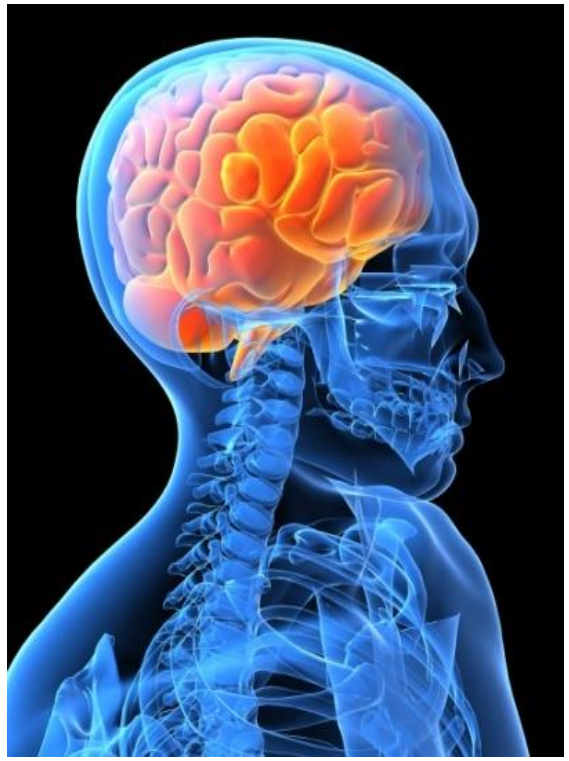
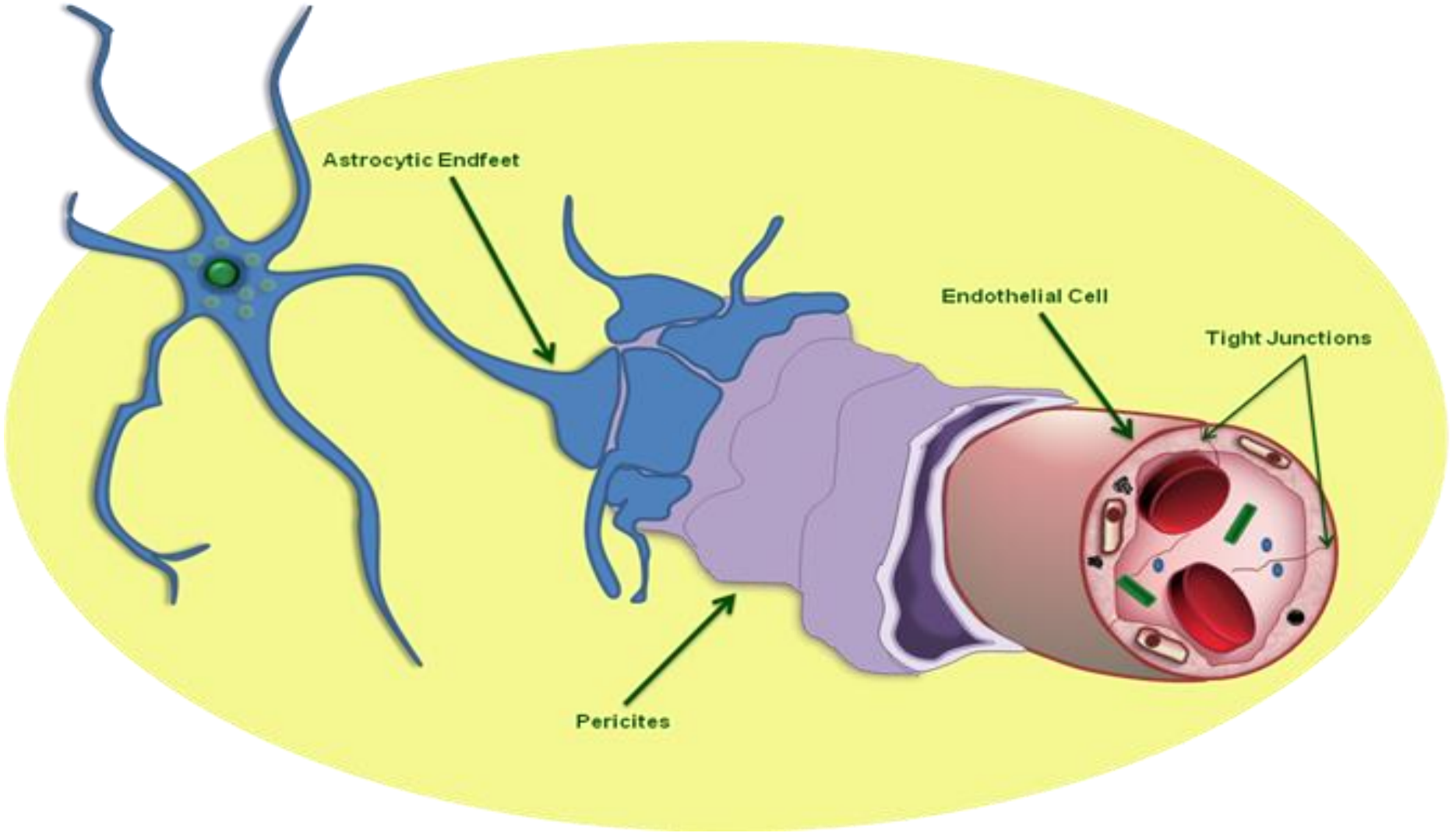
Peptides and nucleic acids in the brain



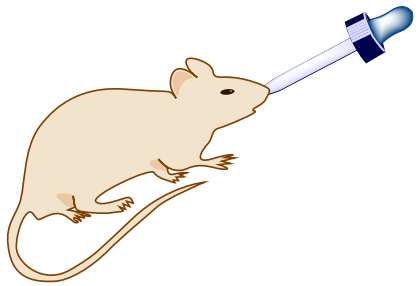


Central Nervous System Diseases

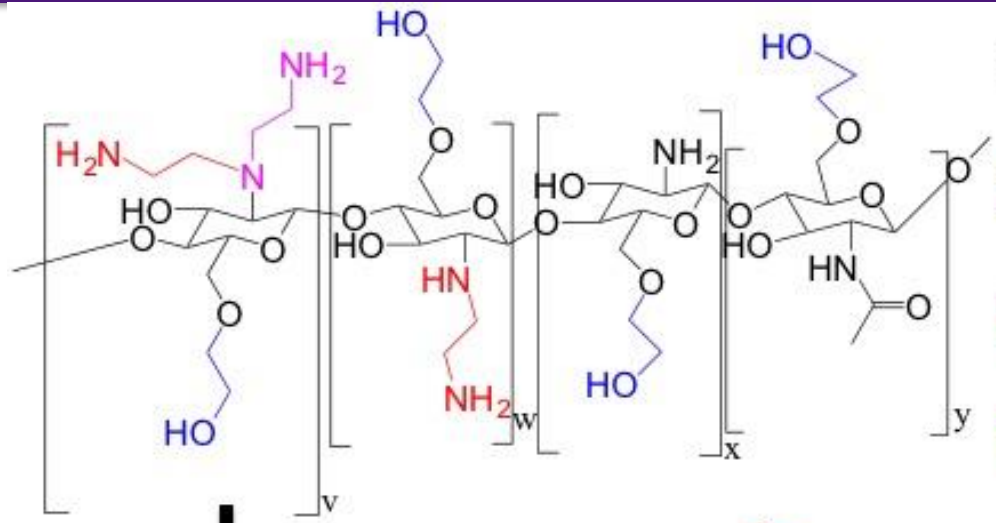
- **1 billion people living with neurological conditions including brain cancer and neurodegeneration**
- **Chronic migraine is the most common neurological disorder in the world affecting an estimated 1% of the global population**
- **50 million people living with epilepsy worldwide**
- **35 million people suffering from Alzheimer's Disease worldwide**
- **6 million patients suffer from chronic neuropathic pain in the developed world with no estimates for the global prevalence of chronic neuropathic pain**
- **Neurological disorders are predicted to grow to become the leading cause of morbidity among the world's 15 – 45 year olds in 2030**
- **Brain tumours have a poor prognosis**

- **Treatment of many CNS diseases hampered by the blood brain barrier**

Nose to brain nucleic acid delivery



Nasal delivery



Biocompatibility

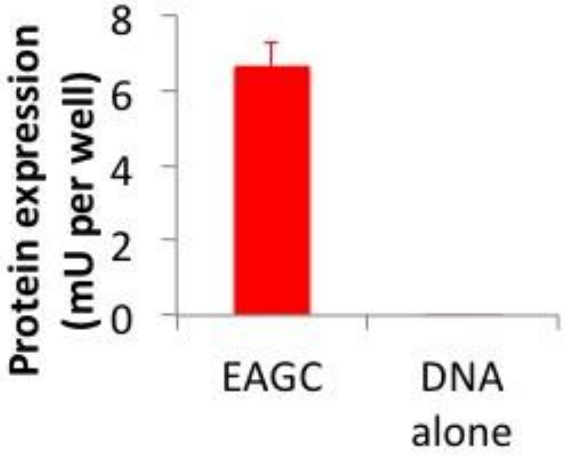
Water solubility

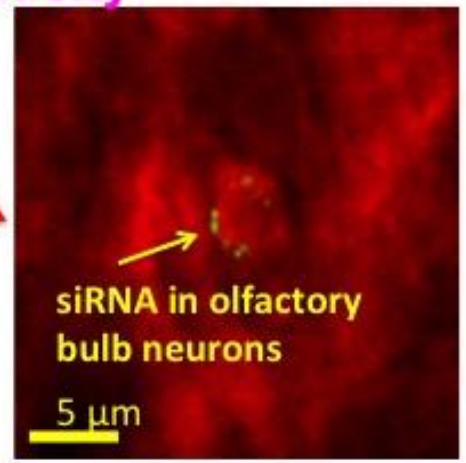
Nucleic acid binding

Toxicity

IC50 = 0.4 – 4.1 mg mL^{-1}

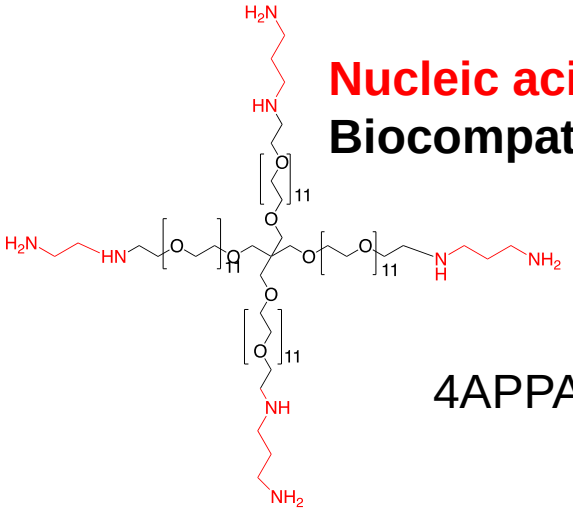
Lipofectamine IC50 = 0.03 mg mL^{-1}





Carlos et al (2017) Int J Pharm, 526, 106–124.

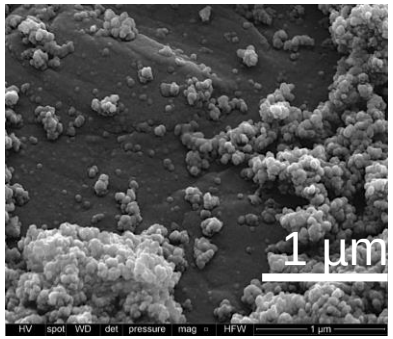
Nose to brain gene silencing

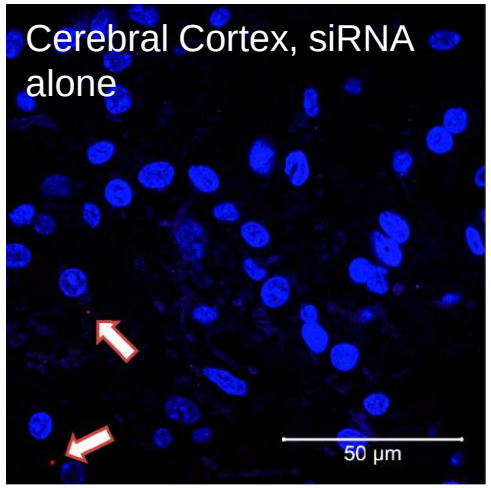


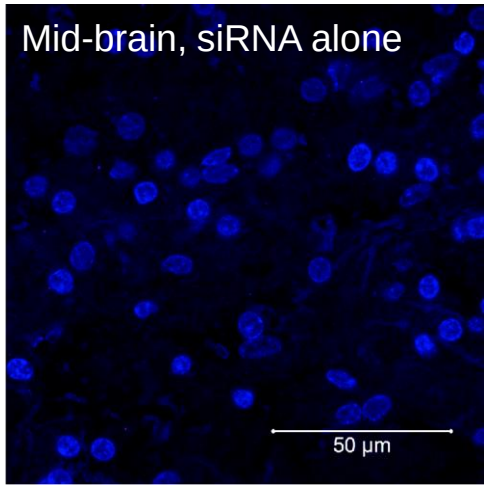
4APPA

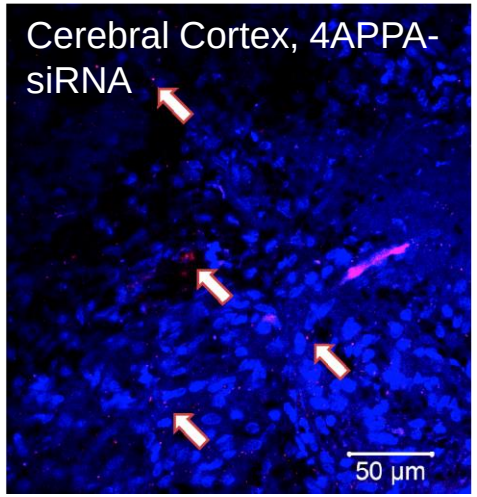
Nucleic acid binding
Biocompatibility

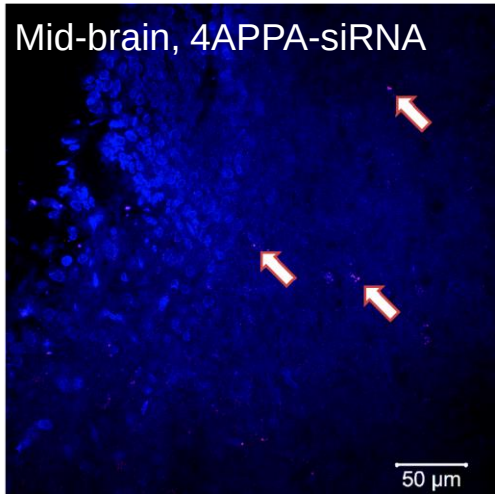
4APPA IC50 = 14 mg mL^{-1}
Lipofectamine IC50 = 0.03mg mL^{-1}

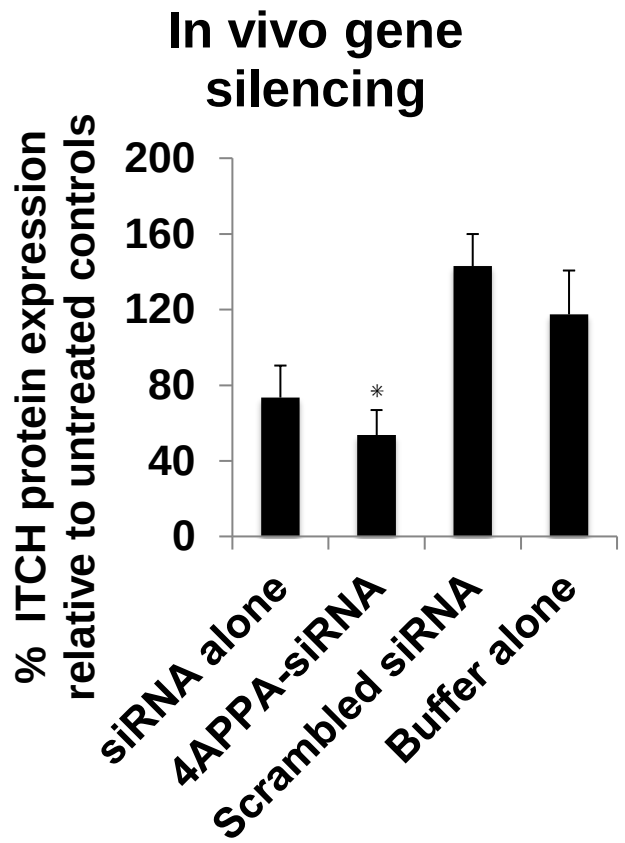








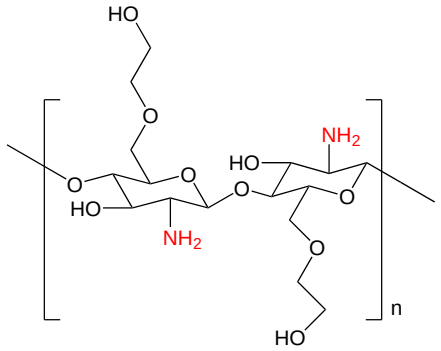




44% gene silencing

Elouzi et al, submitted

Nose to brain gene delivery

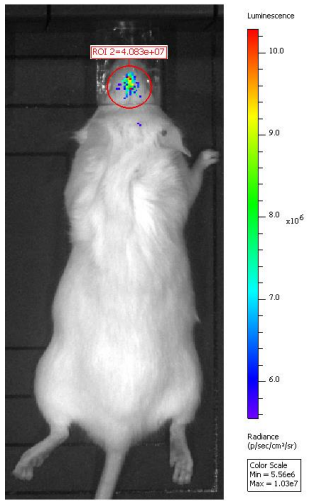


DNA binding

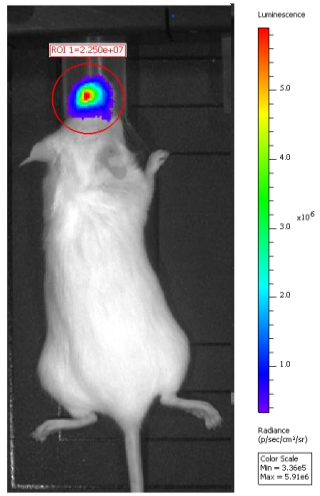
Biocompatibility

GC60 / Luc-DNA

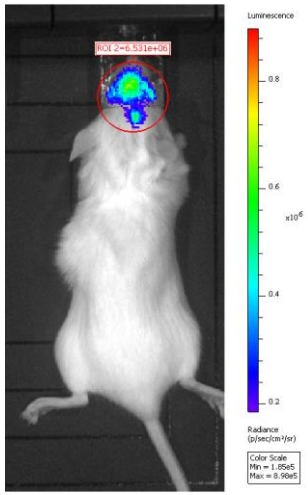
0.02 mg/Kg



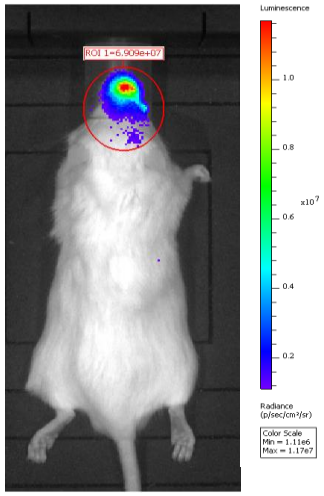
0.04 mg/Kg



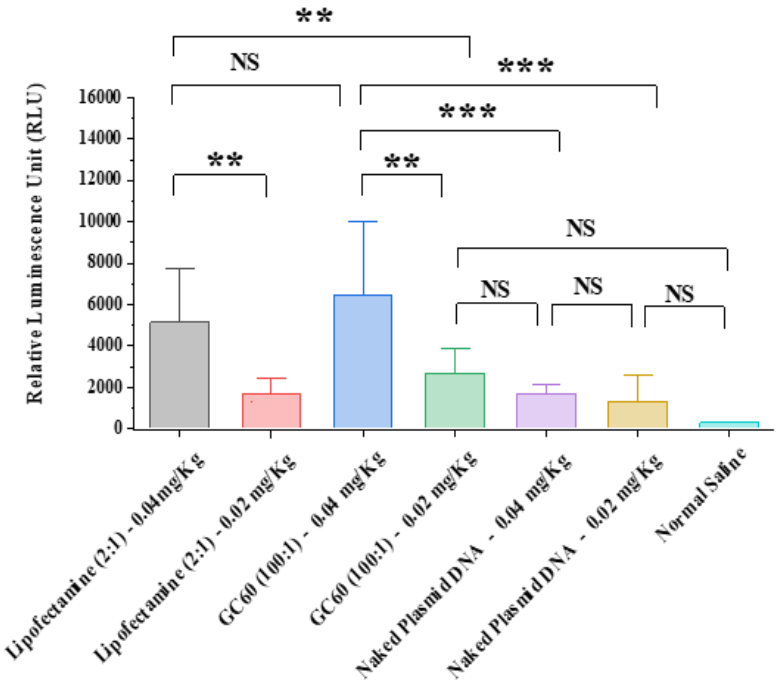
0.06 mg/Kg



0.1 mg/Kg



Cortex

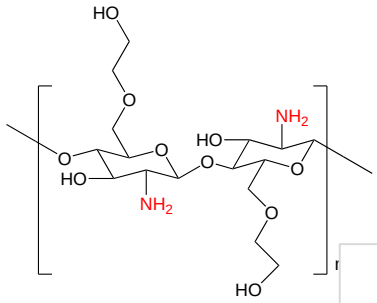


Highest gene expression consistently found in the cortex over multiple experiments

Petkova et al, 2022, Pharmaceutics, 14, 1136

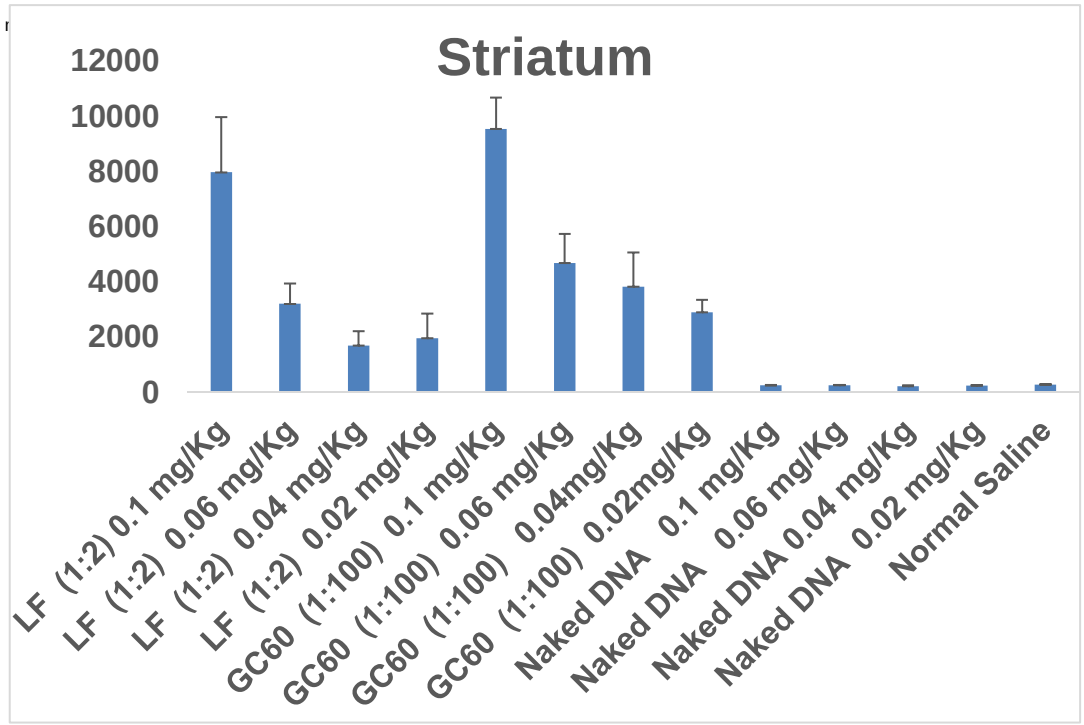
Fatani et al, in preparation

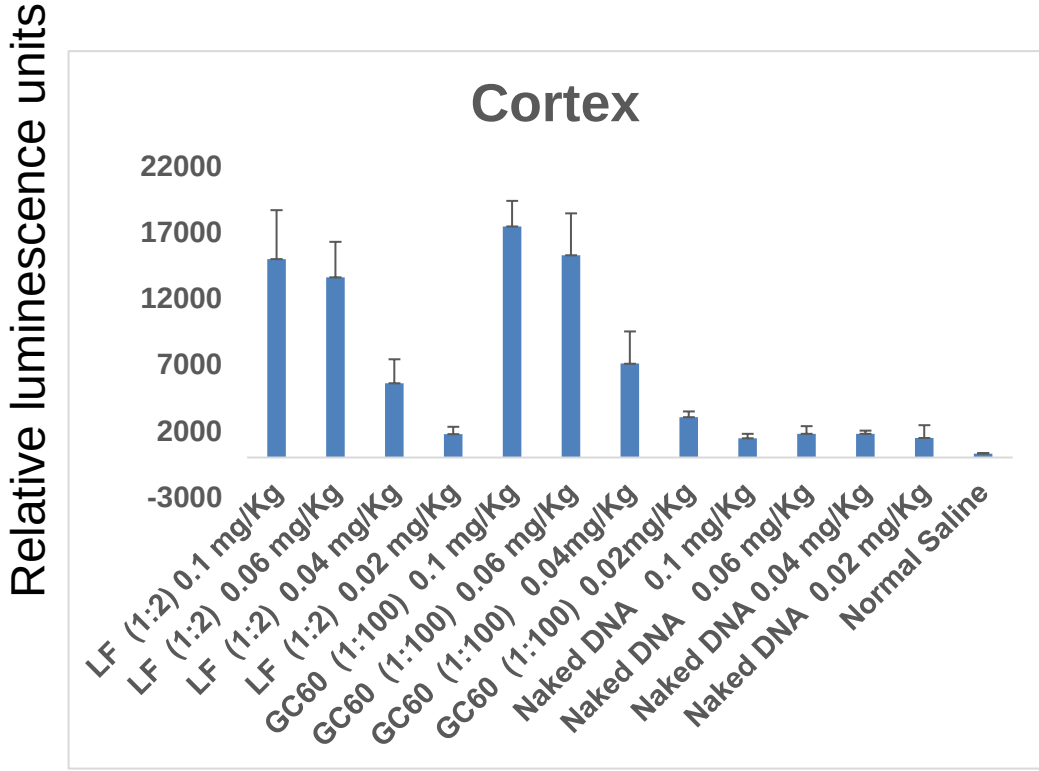
Nose to brain gene delivery



DNA binding

Biocompatibility





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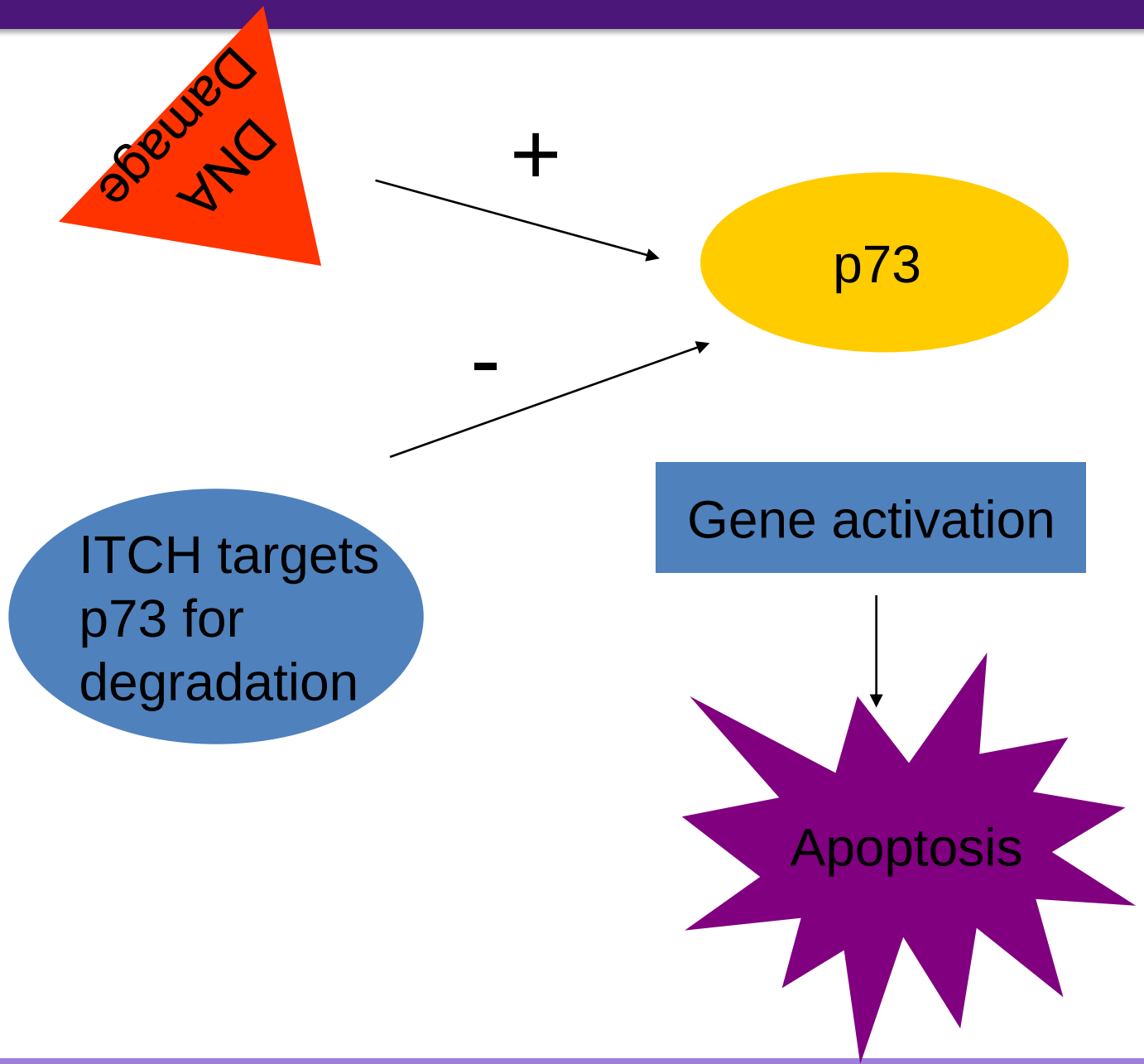
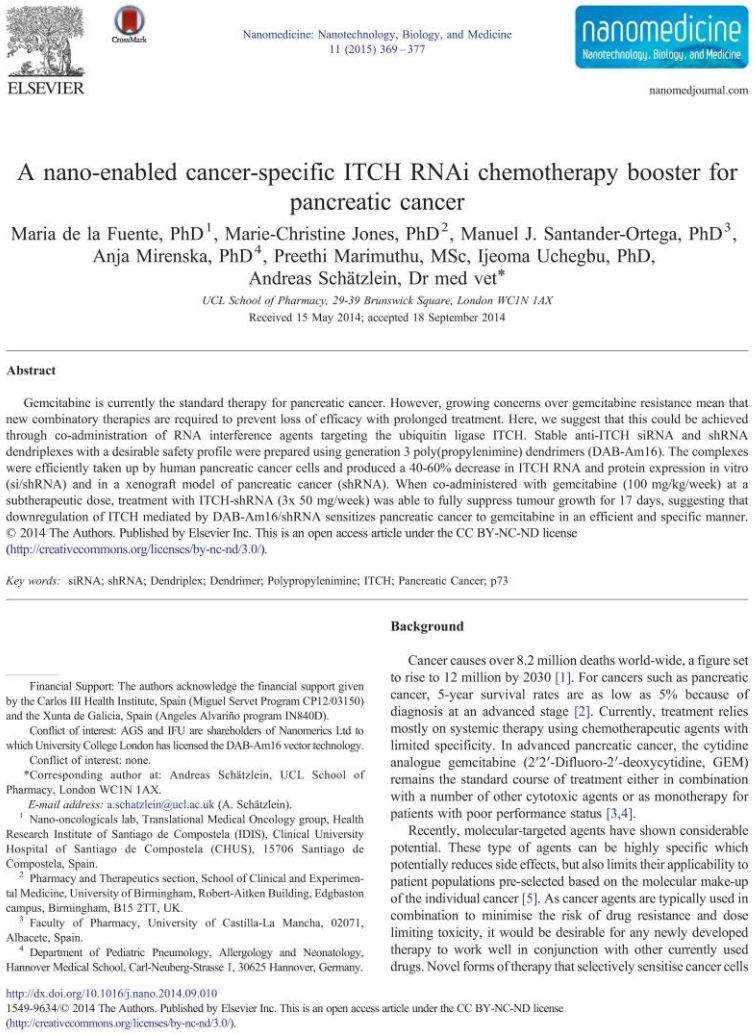
Therapeutic Intervention - ITCH as a Target



ITCH



ELSEVIER

Nanomedicine: Nanotechnology, Biology, and Medicine
11 (2015) 369–377

nanomedicine
Nanotechnology, Biology, and Medicine

nanomedjournal.com

A nano-enabled cancer-specific ITCH RNAi chemotherapy booster for pancreatic cancer

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Abstract

Gemcitabine is currently the standard therapy for pancreatic cancer. However, growing concerns over gemcitabine resistance mean that new combinatory therapies are required to prevent loss of efficacy with prolonged treatment. Here, we suggest that this could be achieved through co-administration of RNA interference agents targeting the ubiquitin ligase ITCH. Stable anti-ITCH siRNA and shRNA dendriplexes with a desirable safety profile were prepared using generation 3 poly(propylenimine) dendrimers (DAB-Am16). The complexes were efficiently taken up by human pancreatic cancer cells and produced a 40-60% decrease in ITCH RNA and protein expression in vitro (si/shRNA) and in a xenograft model of pancreatic cancer (shRNA). When co-administered with gemcitabine (100 mg/kg/week) at a subtherapeutic dose, treatment with ITCH-shRNA (3x 50 mg/week) was able to fully suppress tumour growth for 17 days, suggesting that downregulation of ITCH mediated by DAB-Am16/shRNA sensitizes pancreatic cancer to gemcitabine in an efficient and specific manner.

Key words: siRNA; shRNA; Dendriplex; Dendrimer; Polypropylenimine; ITCH; Pancreatic Cancer; p73

Background

Cancer causes over 8.2 million deaths world-wide, a figure set to rise to 12 million by 2030 [1]. For cancers such as pancreatic cancer, 5-year survival rates are as low as 5% because of diagnosis at an advanced stage [2]. Currently, treatment relies mostly on systemic therapy using chemotherapeutic agents with limited specificity. In advanced pancreatic cancer, the cytidine analogue gemcitabine (2′2′-Difluoro-2′-deoxycytidine, GEM) remains the standard course of treatment either in combination with a number of other cytotoxic agents or as monotherapy for patients with poor performance status [3,4].

Recently, molecular-targeted agents have shown considerable potential. These type of agents can be highly specific which potentially reduces side effects, but also limits their applicability to patient populations pre-selected based on the molecular make-up of the individual cancer [5]. As cancer agents are typically used in combination to minimise the risk of drug resistance and dose limiting toxicity, it would be desirable for any newly developed therapy to work well in conjunction with other currently used drugs. Novel forms of therapy that selectively sensitise cancer cells

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Conflict of interest: AGS and IFU are shareholders of Nanomerics Ltd to which University College London has licensed the DAB-Am16 vector technology.

Conflict of interest: none.

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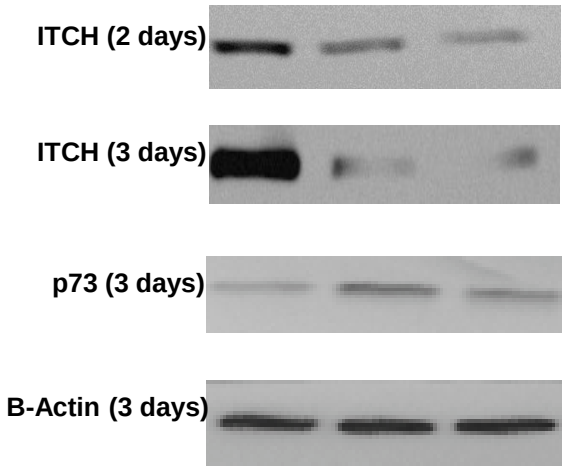
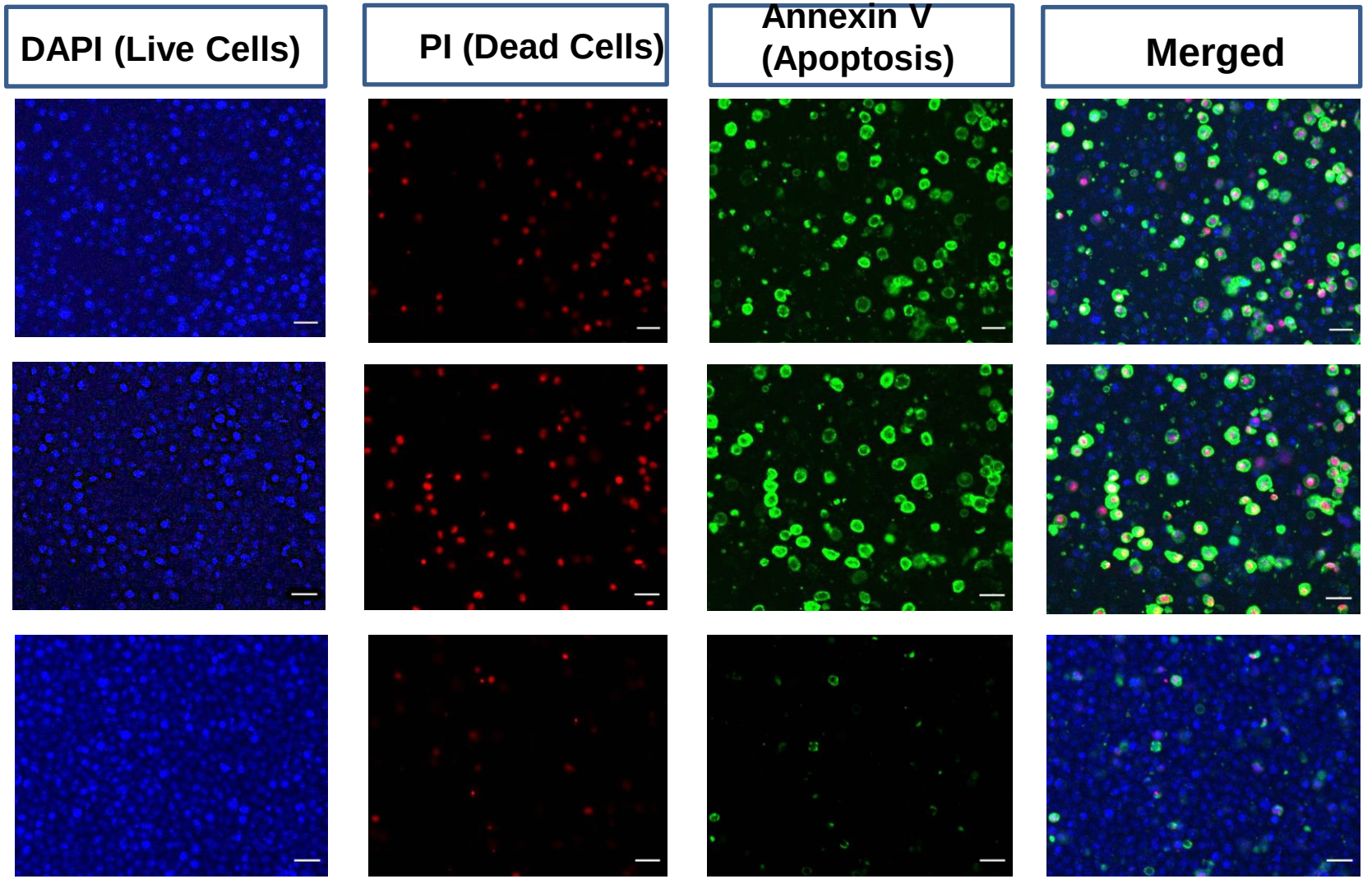
http://dx.doi.org/10.1016/j.nano.2014.09.010
1549-9634/

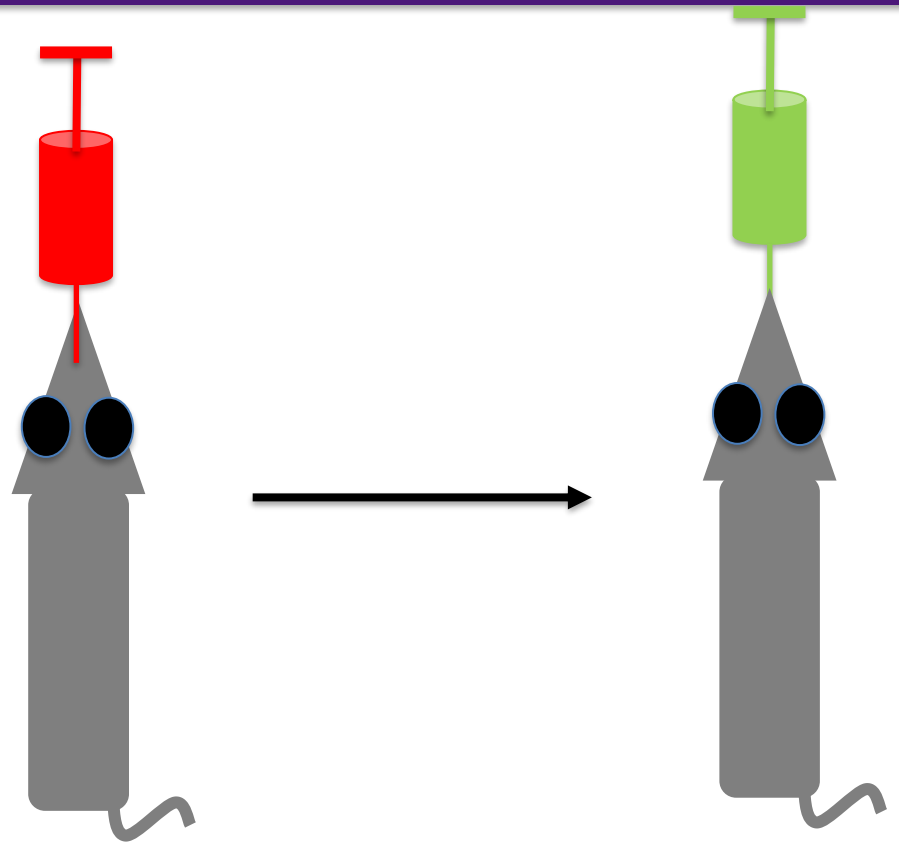
A combination of ITCH shRNA and a sub-therapeutic dose of gemcitabine suppressed tumour growth for 17 days

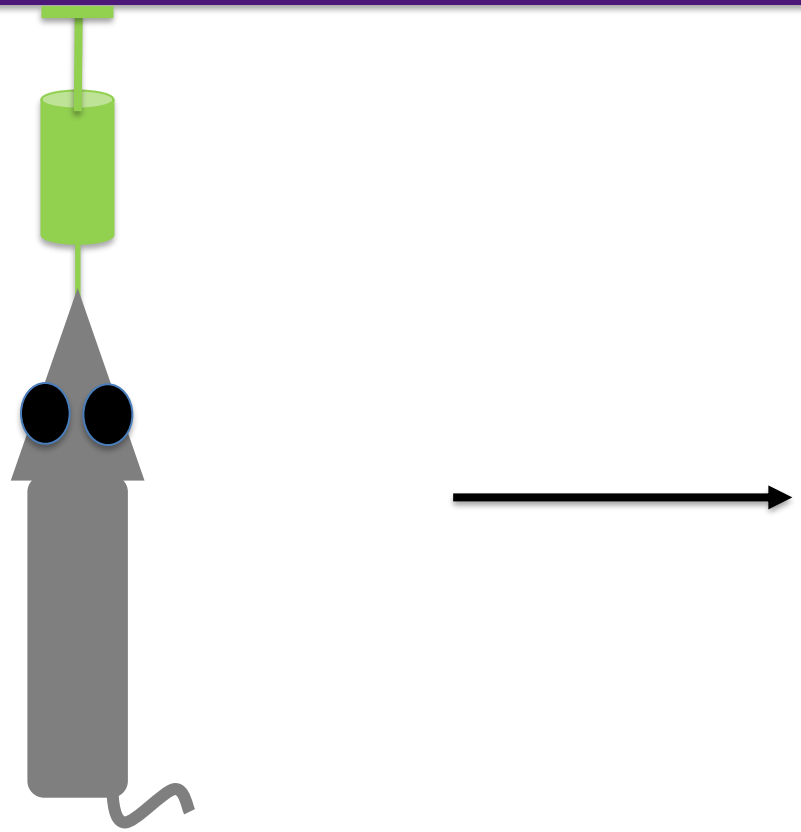
ITCH, p73 and apoptosis





Therapeutic strategy





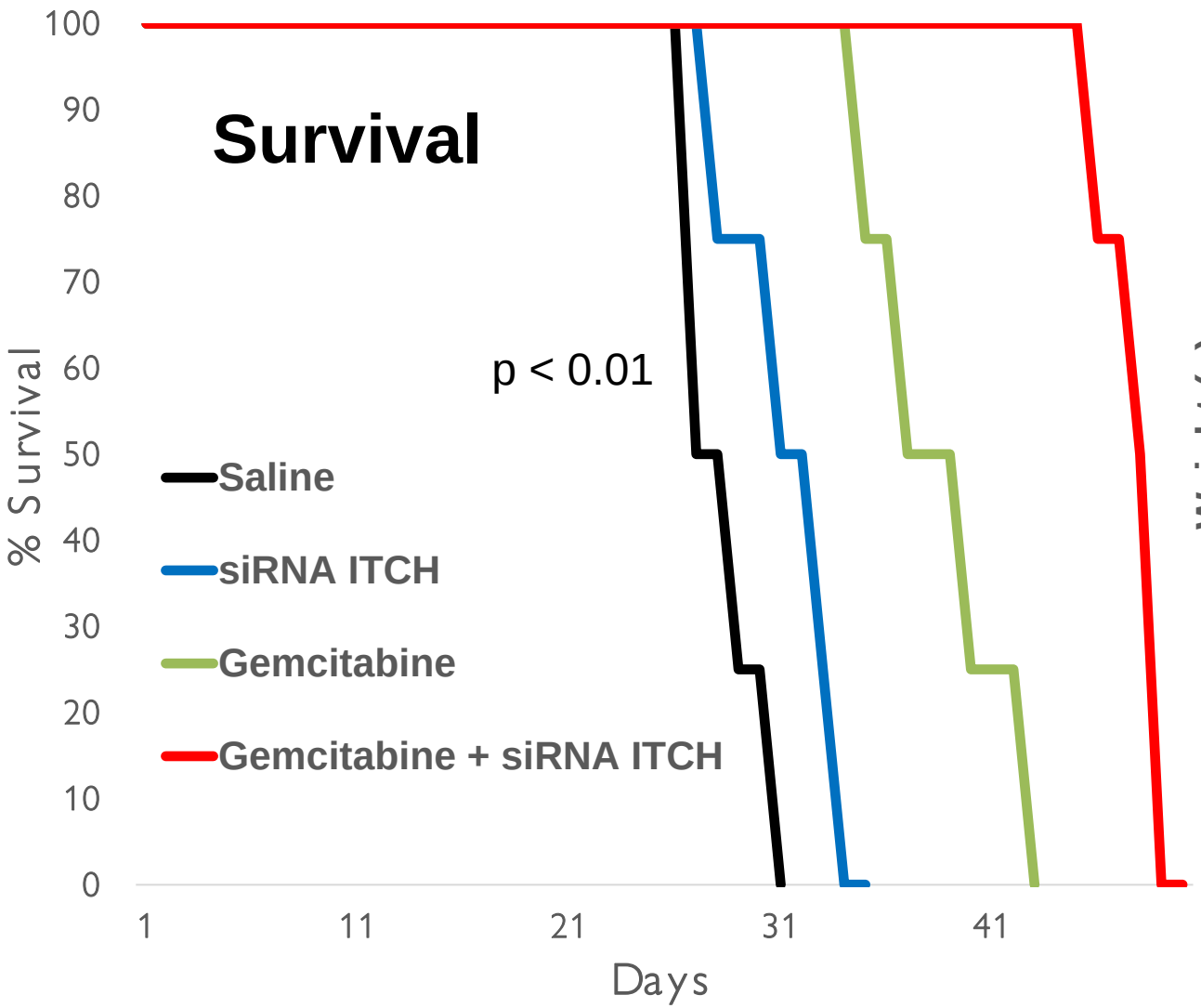
Day 0 – Day 50
a) clinical scoring
b) weighing

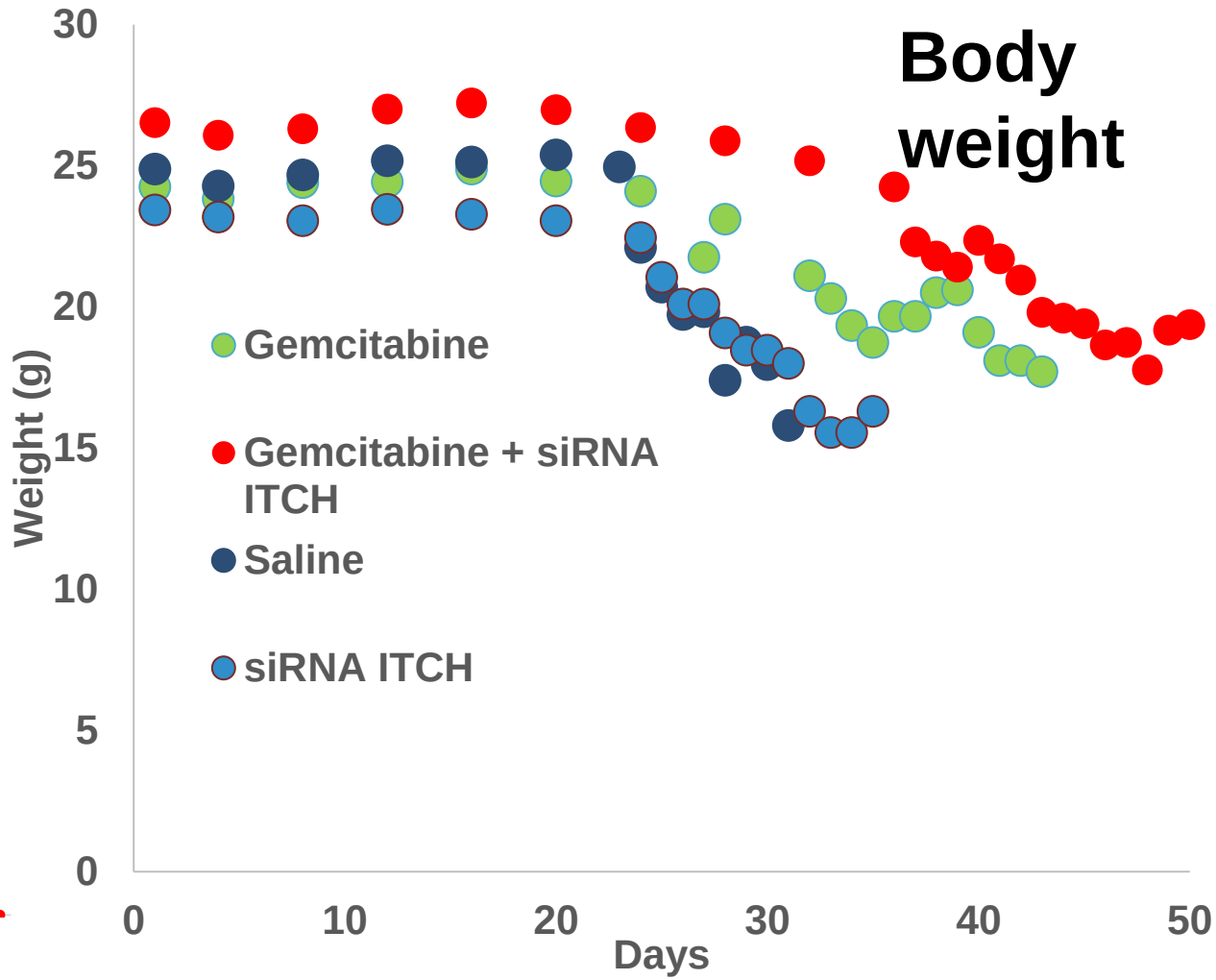
Day 0
Intracranial tumour cell (U87MG) implantation

Day 4
Intranasal treatment:
a) Saline
b) SiRNA ITCH
c) Gemcitabine
d) Gemcitabine + siRNA ITCH

Fatani, in preparatio

Brain gene therapy





Fatani, in preparation

Brain gene therapy

Treatment	Survival
Saline	26 days
siRNA ITCH	27 days
Gemcitabine	34 days
Gemcitabine + siRNA ITCH	45 days

Survival extended by 33% when siRNA ITCH combined with gemcitabine

Fatani, in preparation

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nanomerics
...deliver more

Molecular Envelope Technology

Nanomerics Leadership Team

Andreas Schätzlein
Professor of Translational Therapeutics, PhD
Chief Executive Officer

Co-founder

UCL

Ijeoma Uchegbu FMedSci, HonFRSC
Professor of Pharmaceutical Nanoscience, PhD
Chief Scientific Officer

Co-founder and inve

UCL

Andrea Mica MSc
Non Executive Director

Business strategy

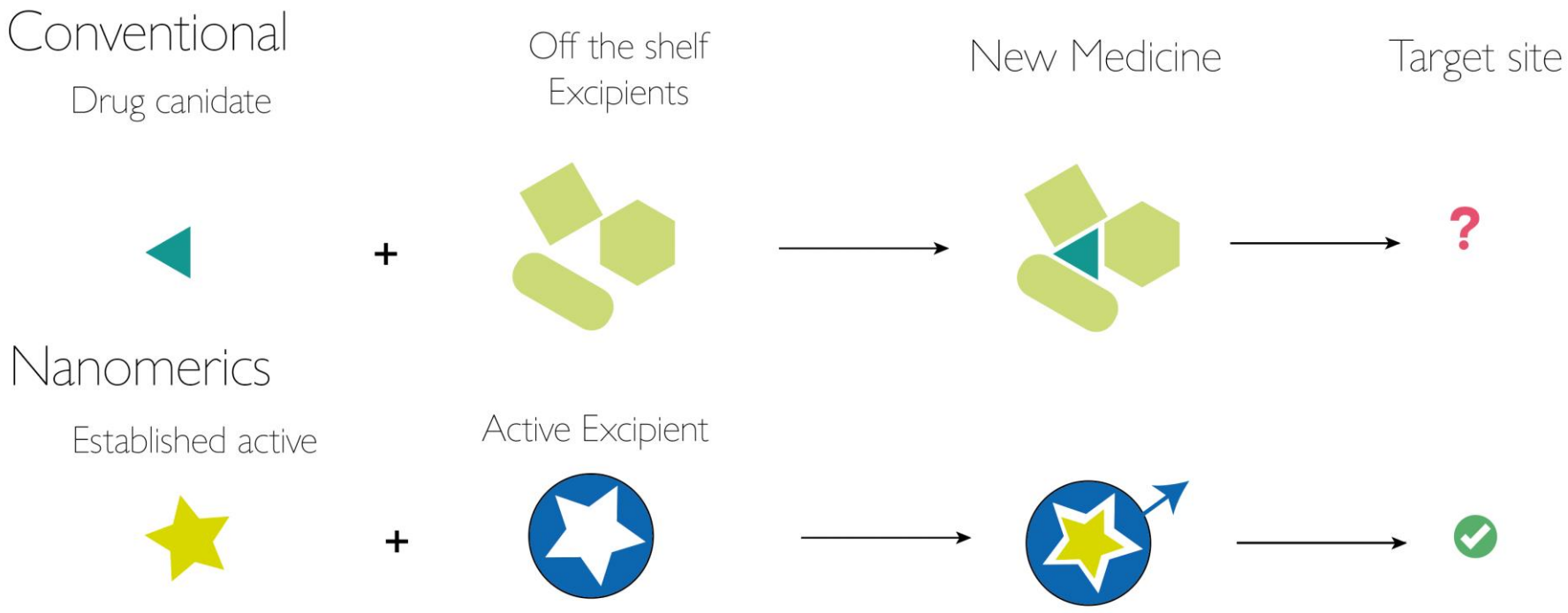
oxford technology

Alan Bye BSc, PhD
Non Executive Director

40 compounds from concept to market

gsk

Nanomerics' active excipient platform accelerates the development of precision medicines



Molecular Envelope Technology (MET) – Ophthalmology, Neurology and More

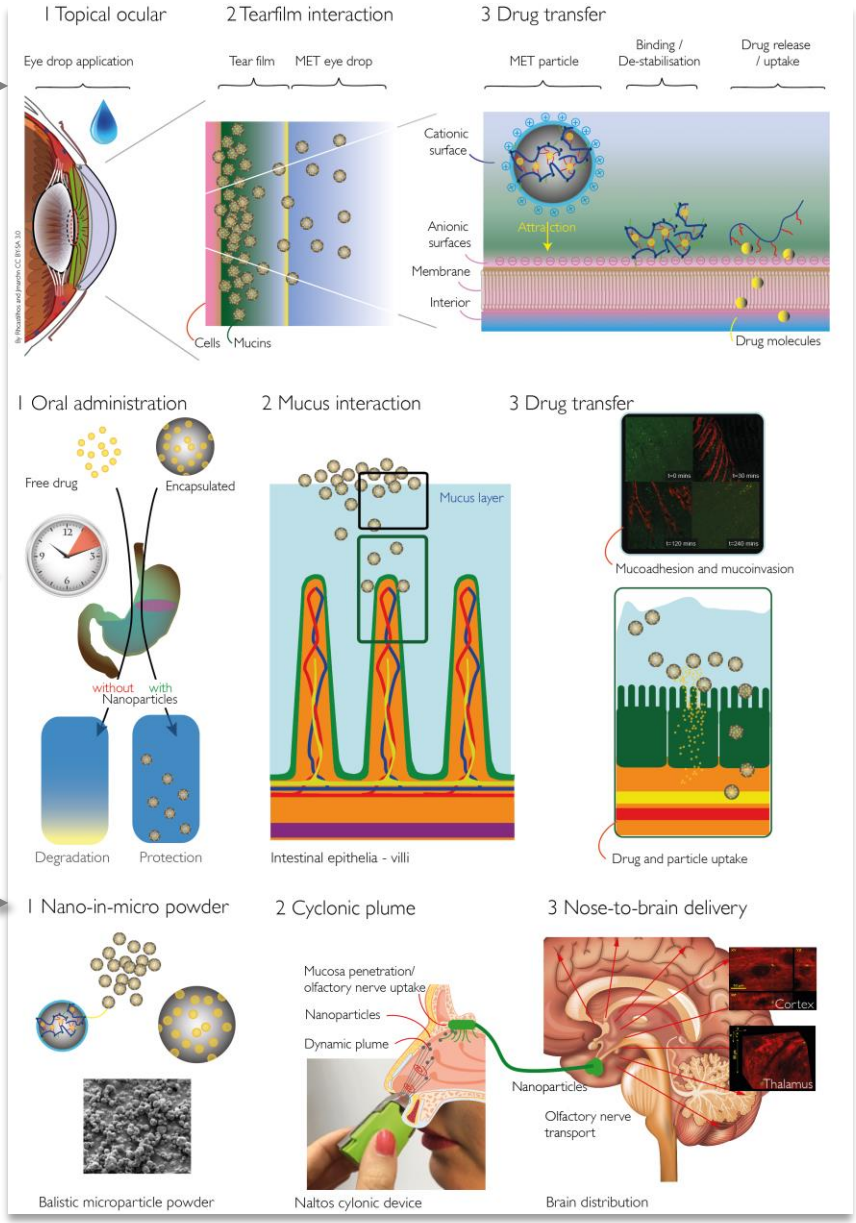
Nasal spray targeting brain diseases

Non-irritant permeation enhancer targeting front and back of the eye ocular diseases

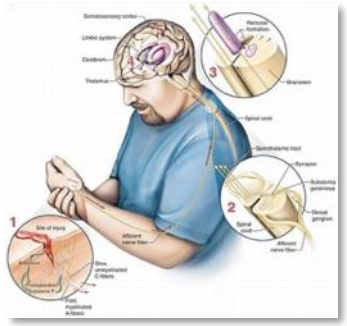
Intravenous to oral switch

Applications

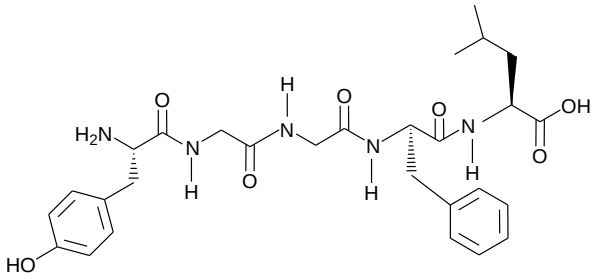
- **Ocular delivery**
  - Topical (eye drops) delivery of drugs to the front **and back** of the eye
  - 5 – 18X more drug delivered compared to conventional eye drops
  - No plasma exposure - efficacy without systemic side effects or ocular injections
- **Oral delivery**
  - Switch from intravenous to oral medicines
  - Convenience without medical infrastructure
  - 3 – 38X increase in Cmax
- **Nose-to-brain delivery**
  - Delivery of drugs to the brain bypassing the blood-brain-barrier with minimal plasma exposure
  - Efficacy without systemic side effects



Pain – A solution

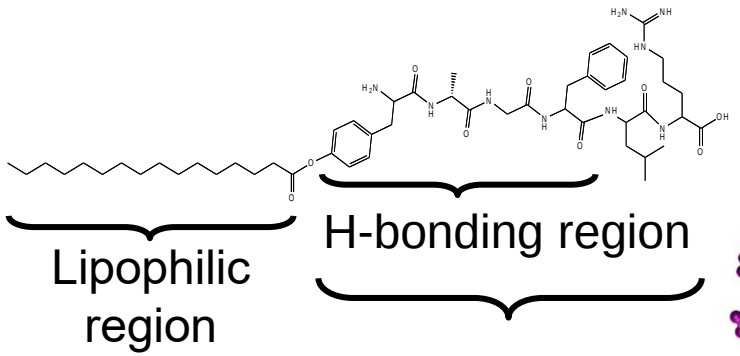


- Acute Pain
  - Breakthrough pain (70% of cancer patients)
- Severe chronic pain
  - Cancer and neuropathic pain (diabetic neuropathy and post herpetic neuralgia)
  - 8.2 Million US patients
  - 20% of European Adults
- Efficacy Problems
  - Only 25% neuropathic pain patients experience pain relief
- Mostly mu opioid receptor agonists
  - 80% patients experience side effects: constipation, nausea, sedation
  - Analgesic tolerance
  - 3 – 19% of patients become addicted to opioids
  - Respiratory depression
    - 425,000 opioid emergency admissions and15,000 deaths annually in the US
- **Hypothesis**
  - **Efficacy plus reduced side effects = a differentiated product**

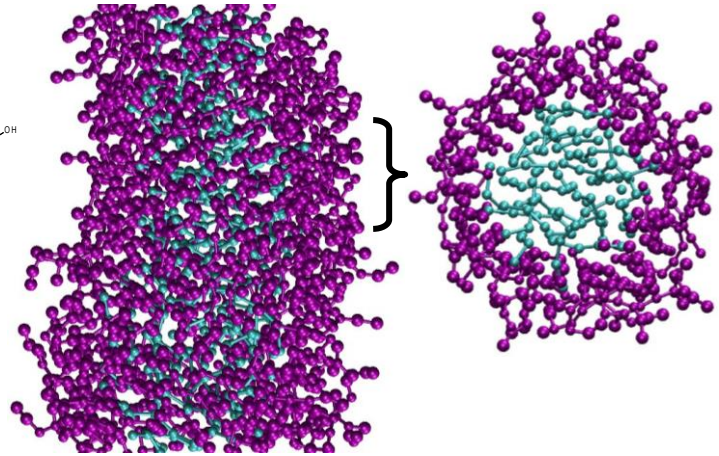


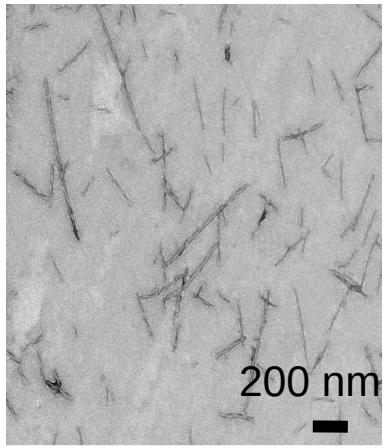
Leucine^5-Enkephalin

Brain delivery of peptides - peptide nanofibres

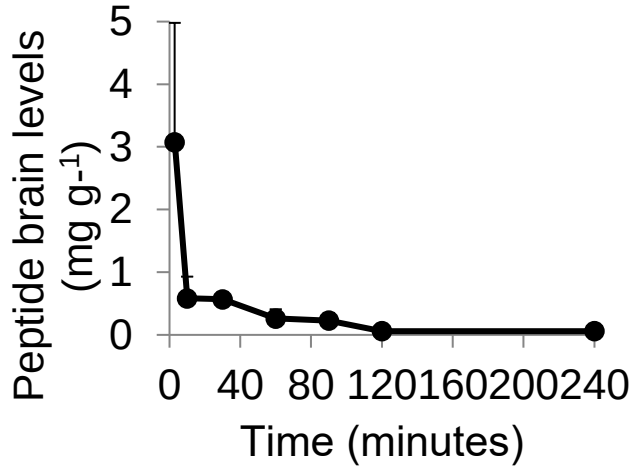


Palmitoyl dalargin (pDal)

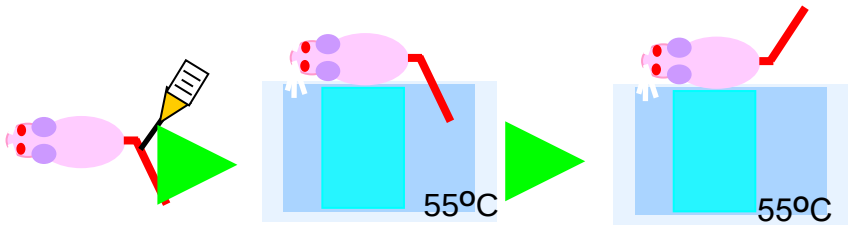


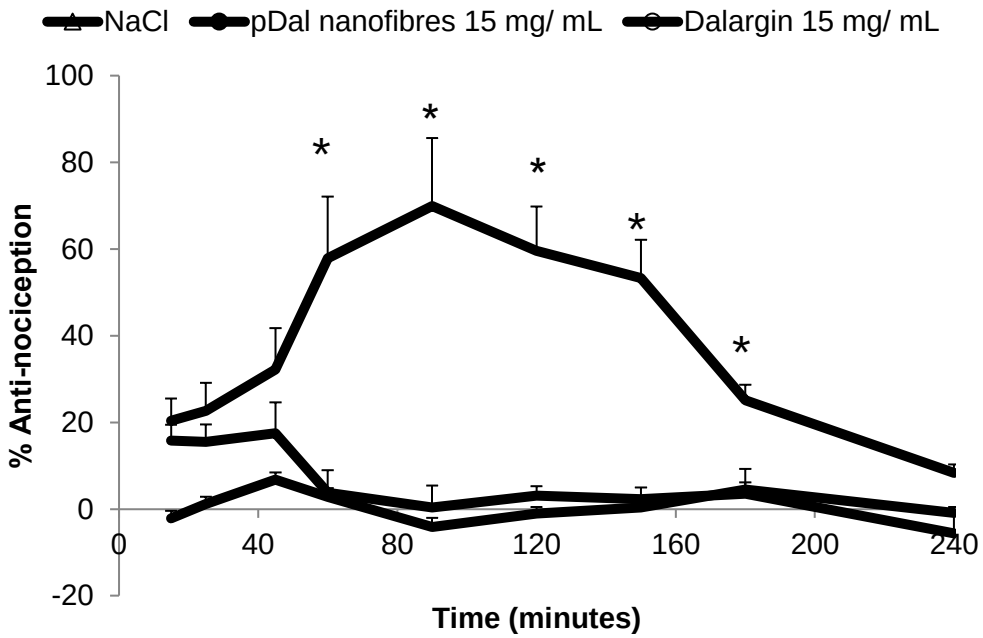


Peptide Nanofibres



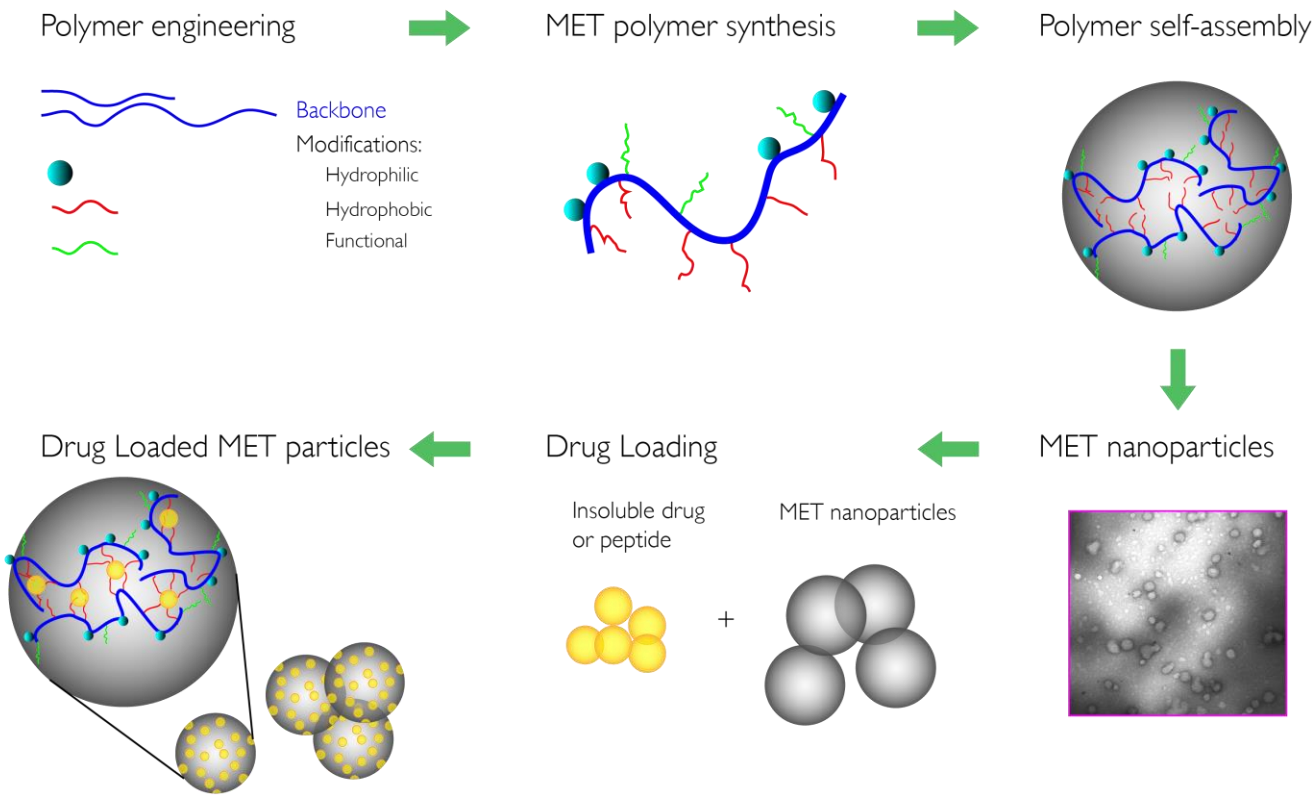
Peptide alone is not detected in the brain





Mazza et al (2013) ACS Nano, 7: 1016 – 1026.

Molecular Envelope Technology (MET)



- Control of polymer properties through chemistry
- Stability for safety and predictability
- Charge controlled surface interactions
- GMP manufacture

MET: Safety Demonstrated in GLP Regulatory Tox Studies

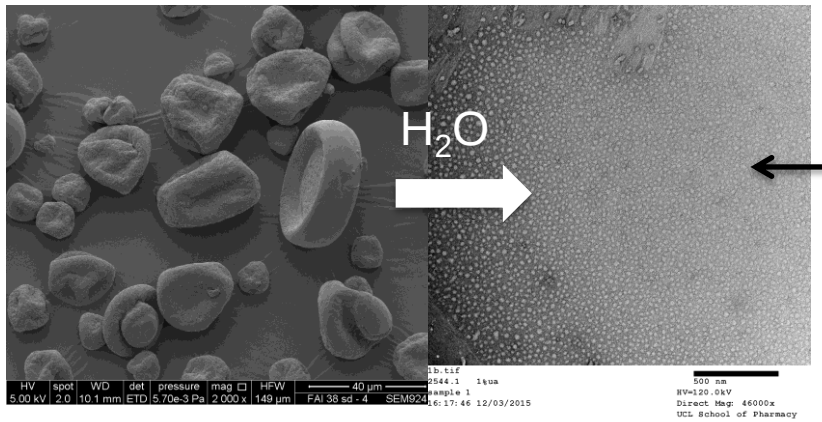
Test	Outcome
Single intravenous dose in the rat	Maximum tolerated dose = 150 mg kg^{-1}
7 Day repeat intravenous dose study in the rat	Maximum tolerated dose = 100 mg kg^{-1}
GLP Mutagenicity testing Ames test	Negative
GLP Mutagenicity testing Mouse Lymphoma Test	Negative
GLP Intravenous Rat Irwin Study	Nothing abnormal detected at 100 mg kg^{-1}
GLP cardiovascular safety pharmacology	*
GLP Intravenous respiratory safety pharmacology in the rat	NOAEL = 40 mg kg^{-1}
Oral 7 day repeat dose ranging dog study	NOAEL = 300 mg kg-1 (top dose studied)
GLP oral 28 day repeat dose dog study	NOAEL = 150 mg kg^{-1} (top dose studied)
Oral 7 day repeat dose ranging rat study	NOAEL = 200 mg kg^{-1}
GLP oral 28 day repeat dose rat study	NOAEL = 200 mg kg^{-1} (top dose studied)
Intranasal 7 day repeat dose ranging rat study	NOAEL = 30 mg kg^{-1} (Reduced weight gain at 50 mg kg^{-1})
GLP 28 day intranasal dose in the rat	NOAEL = 18 mg kg^{-1}
6 day topical Ocular tolerability study in the rabbit	NOAEL = 40 mg mL^{-1} (top concentration studied)

* Access through licensee

Overview

- Aims and tools
- Gene delivery to the bladder
- Brain delivery
  - Gene delivery to the brain
- Commercialization
  - **Envelta™**
  - Ocular penetration enhancer
- Summary

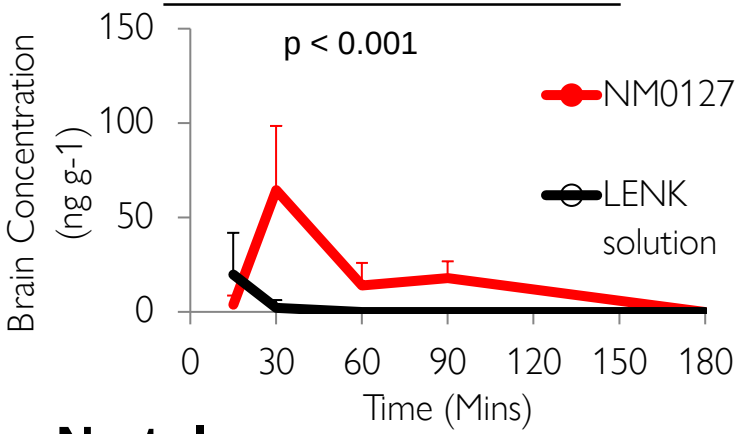
Envelta™ - Molecular Envelope Technology + leucine enkephalin



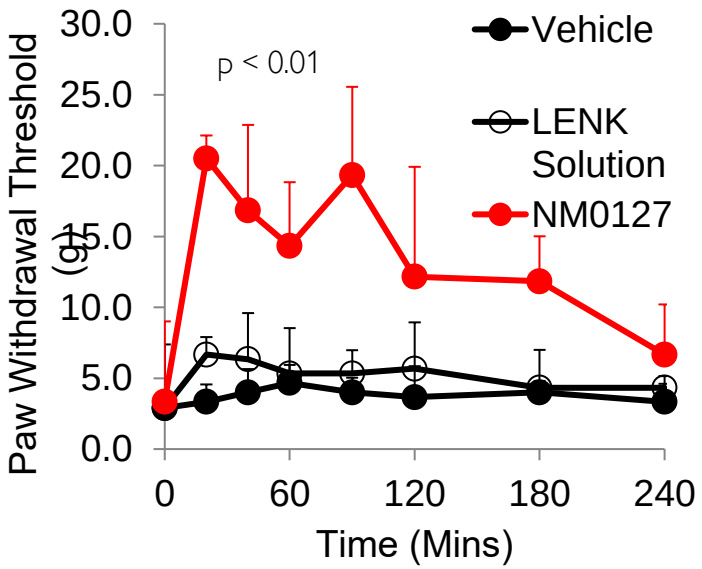
Manufacture
Nano-in Micro powder

Particulate Nose to Brain Delivery

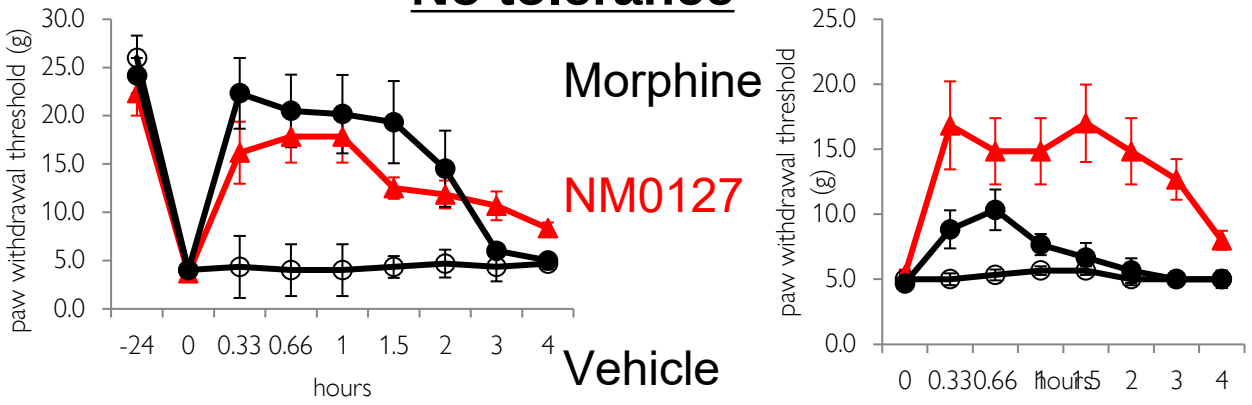
Pharmacokinetics



Pharmacodynamics



No tolerance



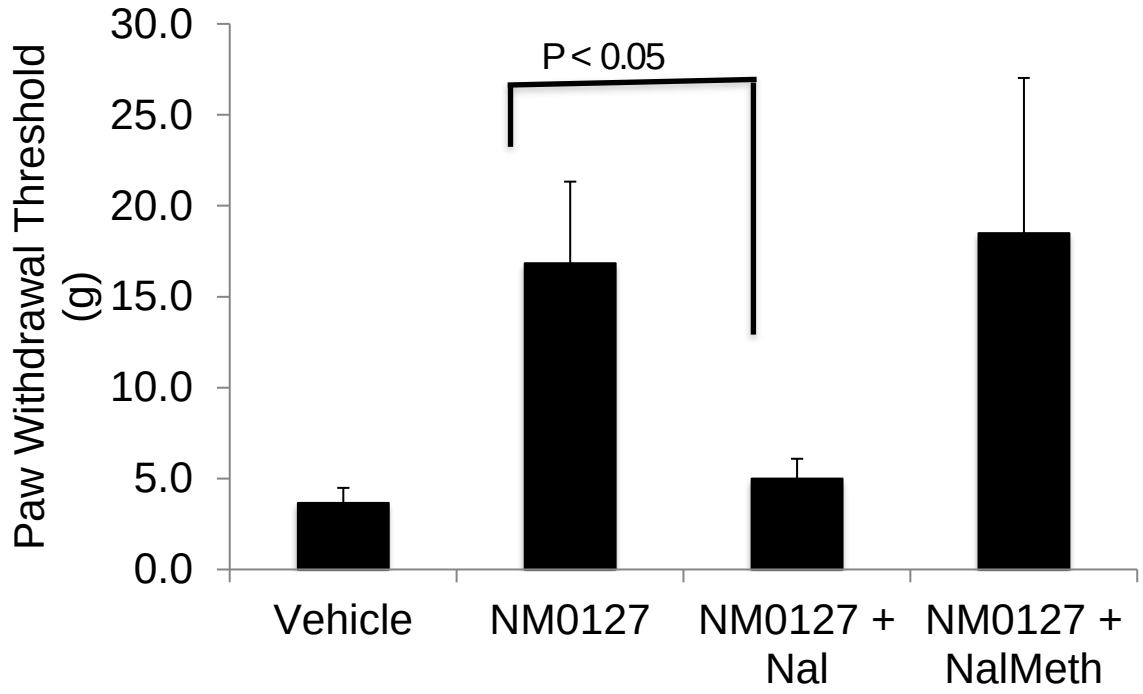
Dosed on Day 1 and challenged with von Frey hairs.

Dosed twice daily

Dosed on Day 5 and challenged again with von Frey hairs.

Godfrey et al, *J Control Release* **2017**, 270, 135-144.

Centrally Acting

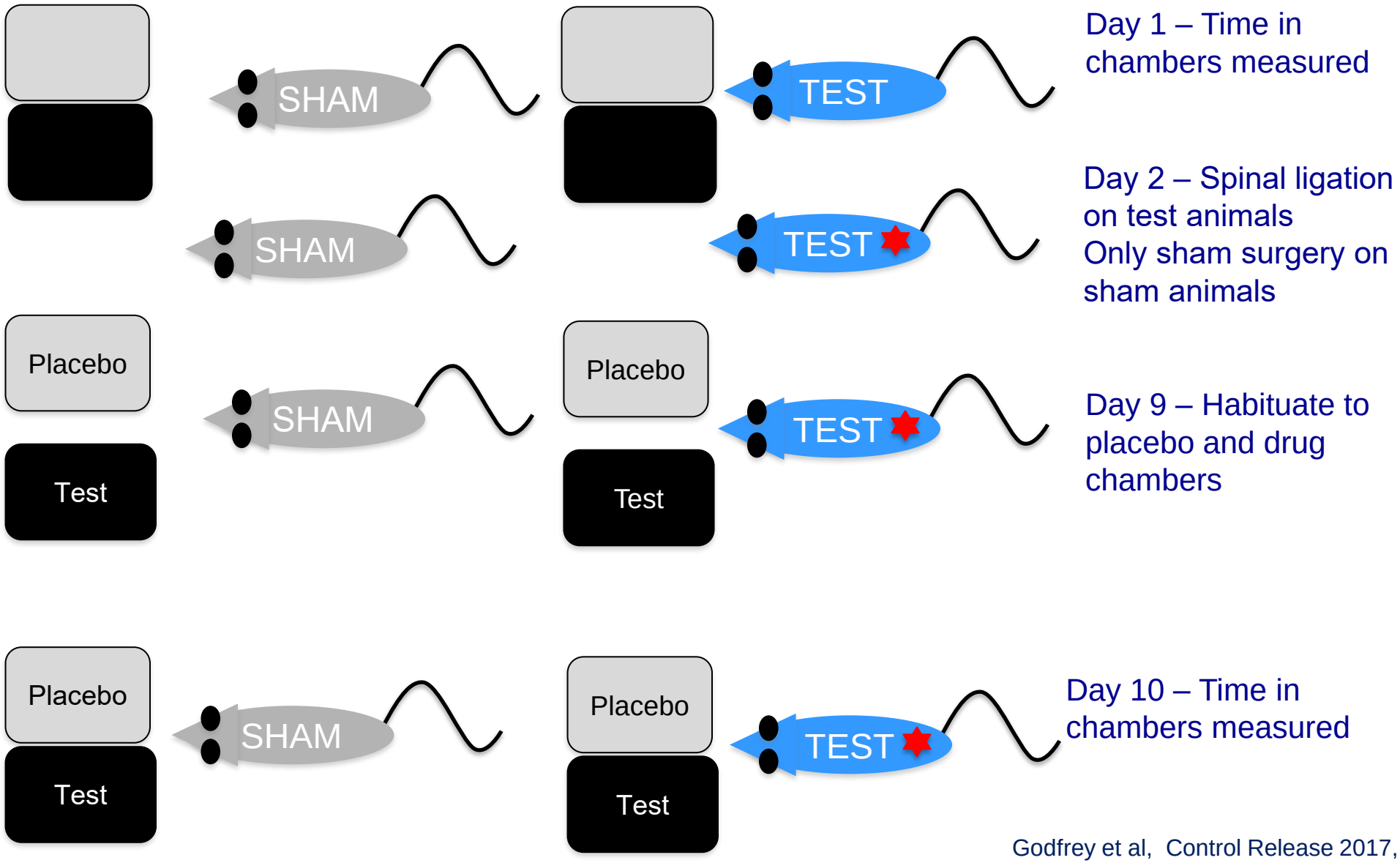


Intranasal NM0127 acts exclusively on the central opioid receptors and is only inhibited by the central inhibitor Naloxone (Nal) and is not inhibited by the peripheral inhibitor (Naloxone Methiodide – NalMeth).

NM0127 unlikely to cause constipation as the constipation side effect is largely peripherally mediated.

Godfrey et al, Control Release 2017, 270, 135-144.

Conditioned Placement Preference Behavioural Experiments



Day 1 – Time in chambers measured

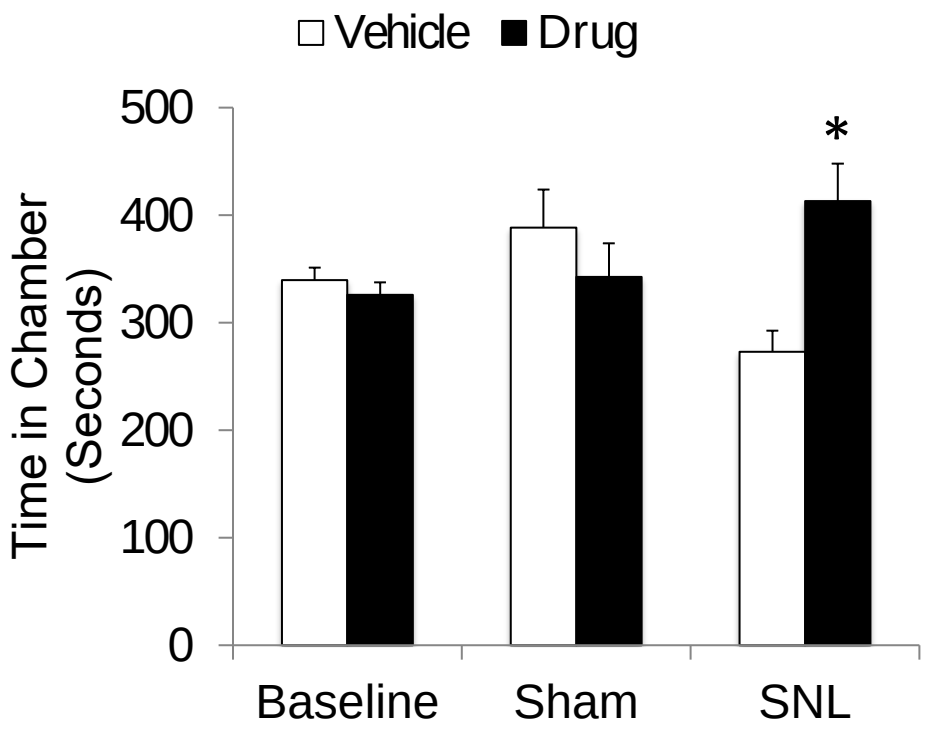
Day 2 – Spinal ligation on test animals
Only sham surgery on sham animals

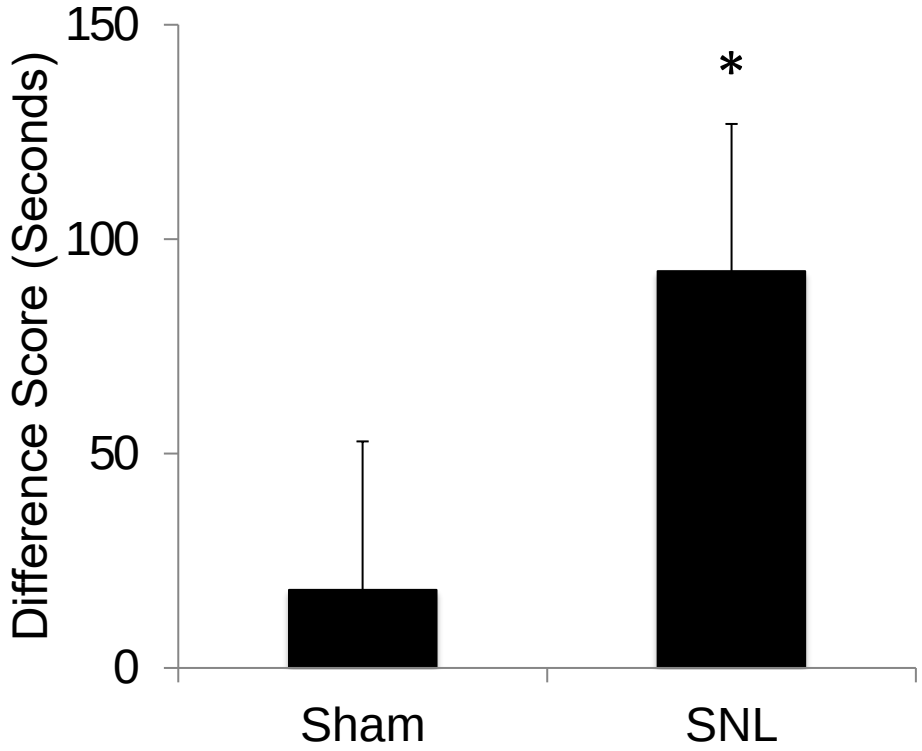
Day 9 – Habituate to placebo and drug chambers

Day 10 – Time in chambers measured

Godfrey et al, Control Release 2017, 270, 135-144.

Activity in a Spinal Ligation Model of Neuropathic Pain





NM0127 activity in a neuropathic pain spinal nerve ligation (SNL) model, BL = Baseline.

SNL animals prefer the NM0127 paired chamber whereas Sham animals show no preference.

There is an absence of reward seeking behaviour

Godfrey et al, Control Release 2017, 270, 135-144.

Envelta™

Nanoparticle technology enables exclusive brain exposure to amphiphilic peptides via the nasal route

- Activity in all pain models
- No reward seeking behaviour
- No tolerance
- Active in morphine tolerants
- Centrally acting so unlikely to cause constipation

Journal of Controlled Release 270 (2018) 135–144

Contents lists available at ScienceDirect

Journal of Controlled Release

journal homepage: www.elsevier.com/locate/jconrel

Nanoparticulate peptide delivery exclusively to the brain produces tolerance free analgesia

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[b] Nanomerics Ltd., 1394 High Road, London N20 9YZ, UK
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[d] Department of Biomedical Sciences, College of Osteopathic Medicine, University of New England, 11 Hills Beach Rd, Biddeford, ME 04005, USA
[e] Department of Pharmacology, College of Medicine, University of Arizona, 1501 N. Campbell Ave, Tucson, AZ 85724, USA
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ARTICLE INFO

Keywords:
Nanoparticles
Microparticles
Chitosan amphiphiles
Leucine[5]-enkephalin
Intranasal
Analgesia
Brain delivery
Delta opioid receptor

ABSTRACT

The delivery of peptide drugs to the brain is challenging, principally due to the blood brain barrier and the low metabolic stability of peptides. Exclusive delivery to the brain with no peripheral exposure has hitherto not been demonstrated with brain quantification data. Here we show that polymer nanoparticles encapsulating leucine[5]-enkephalin hydrochloride (LENK) are able to transport LENK exclusively to the brain via the intranasal route, with no peripheral exposure and nanoparticle localisation is observed within the brain parenchyma. Animals dosed with LENK nanoparticles (NM0127) showed a strong anti-nociceptive response in multiple assays of evoked and on going pain whereas animals dosed intranasally with LENK alone were unresponsive. Animals did not develop tolerance to the anti-hyperalgesic activity of NM0127 and NM0127 was active in morphine tolerant animals. A microparticulate formulation of clustered nanoparticles was prepared to satisfy regulatory requirements for nasal dosage forms and the polymer nanoparticles alone were found to be biocompatible, via the nasal route, on chronic dosing.

1. Introduction

The delivery of peptides to the brain is challenging, not merely because of the blood brain barrier but also because peptides have a very short plasma half life and are frequently not detected in the plasma on intravenous administration [1]. The intranasal to brain route of administration has emerged as an interesting route for the administration of compounds directly into the brain [2] even though the dose (100 μL or 25 mg) is a limitation. Here we show that the use of an intranasal nanoparticle delivery system enables the delivery of a metabolically unstable [3], δ selective opioid receptor (DOR) [4][5] agonist, leucine[5]-enkephalin hydrochloride (LENK), directly and exclusively to the brain. The δ selectivity has been well studied by Toll and others [5] using cloned human μ, δ and κ receptors in Chinese Hamster Ovary (CHO) cells with EC50 values, in ^{35}SGTPγS binding assays, of 25.5 ± 0.8 nM and 1.35 ± 0.2 nM for μ-CHO and δ-CHO cell membranes respectively. Similar results were obtained with guinea pig brain membranes with Ki values of 21.7 ± 1.4 nm and 1.6 ± 0.5 nm obtained when using the μ (^{3}H DAMGO) and δ (^{3}H DPDPE) selective agonists respectively [5]. DORs reside in the cerebral cortex, putamen, caudate nucleus, nucleus accumbens and hippocampus of humans [6] and hence exclusive brain delivery, via a non-parenteral route of administration, enables LENK to be considered as a potential analgesic. Chronic pain affects 19% of European adults, with nearly half being poorly managed by current therapies and with devastating consequences on their quality of life [7]. While neuropathic pain (diabetic, post herpetic or human immunodeficiency virus related) affects 6 million patients in the seven major markets (United States, Japan, France, Germany, Italy, Spain and the UK), only 25% of these patients experience pain relief with the current approved medicines [8]. Furthermore breakthrough pain is highly prevalent in analgesic treated cancer patients with one study reporting prevalence rates of 74% [9] and 45% of the patients studied were unable to predict the onset of excruciating breakthrough pain [9]. Breakthrough pain requires remedies with a rapid onset of action.

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Received 18 October 2017; Received in revised form 24 November 2017; Accepted 25 November 2017
Available online 27 November 2017

Godfrey et al, J Control Release 2017, 270, 135-144.

Envelta™ out-licensed to Virpax Pharmaceuticals (NASDAQ:VRPX)

In-licensed Naltos delivery device

AP

Virpax Begins IND Enabling Studies of Envelta(TM)

Virpax Begins IND Enabling Studies of Envelta(TM)

February 23, 2021

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Business

Virpax Reports Favorable Preclinical Safety Data for Envelta™ for the Treatment of Acute and Chronic Pain

10 February 2022, 14:00 GMT

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Virpax Reports Favorable Preclinical Safety Data for Envelta™ for the Treatment of Acute and Chronic Pain

- Preclinical study data indicate that intranasal administration of enkephalin (Envelta) is well-tolerated and safe -

Business Wire

BERWYN, Pa. -- February 10, 2022

Virpax® Pharmaceuticals, Inc. ("Virpax" or the "Company") (NASDAQ: VRPX), a company specializing in developing non-addictive product candidates for pain management, as well as PTSD, CNS disorders and anti-viral indications, reported promising results from preclinical dose range finding studies for Envelta (NES100), Virpax's endogenous enkephalin intranasal spray for acute and chronic pain, including pain associated with cancer. These findings complement previous positive preclinical toxicology results and support the Company's continuing development of this potential non-addictive treatment for acute and chronic pain.

Investigational New Drug (IND) enabling studies for Envelta are being performed under a Cooperative Research and Development Agreement (CRADA) with the National Institutes of Health's (NIH) National Center for Advancing Translational Sciences (NCATS) entered into by Virpax and the NIH in August of 2020. Under the cooperation agreement, the studies are performed and funded by

Parameter	Envelta™	Fentanyl	Morphine
Fast acting within (15 minutes)	✔	✔	X
Non-invasive route of administration	✔	✔	X ✔
Active ingredient efficacious in humans	✔	✔	✔
Absence of constipation side effects	✔ (as centrally acting)	X	X
Absence of respiratory depression	✔ (delta opioid receptor agonist)	X	X
Absence of analgesic tolerance	✔	X	X
Active in morphine tolerants	✔	X (only at high doses)	X
Absence of reward seeking behaviour	✔	X	X

Overview

- Aims and tools
- Gene delivery to the bladder
- Brain delivery
  - Gene delivery to the brain
- Commercialization
  - Envelta™
  - Ocular penetration enhancer
- Summary

MET Comparative Advantage – Corneal Deposition

pharmaceutics

MDPI

Article

Polymeric Micelles for the Enhanced Deposition of Hydrophobic Drugs into Ocular Tissues, without Plasma Exposure

Ijeoma F. Uchegbu [1,2,*], Jan Breznikar [1], Alessandra Zaffalon [1], Uche Odunze [1] and Andreas G. Schätzlein [1,2]

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Abstract: Commercial topical ocular formulations for hydrophobic actives rely on the use of suspensions or oil in water emulsions and neither of these formulation modalities adequately promote drug penetration into ocular tissues. Using the ocular relevant hydrophobic drug, cyclosporine A (CsA), a non-irritant ocular penetration enhancer is showcased, which may be used for the formulation of hydrophobic actives. The activity of this penetration enhancer is demonstrated in a healthy rabbit model. The Molecular Envelope Technology (MET) polymer (N-palmitoyl-N-monomethyl-N,N-dimethyl-N,N,N-trimethyl-6-O-glycolchitosan), a self-assembling, micelle-forming polymer, was used to formulate CsA into sterile filtered nanoparticulate eye drop formulations and the stability of the formulation tested. Healthy rabbits were dosed with a single dose of a MET–CsA (NM133) 0.05% formulation and ocular tissues analyzed. Optically clear NM133 formulations were prepared containing between 0.01–0.1% *w/v* CsA and 0.375–0.75% *w/v* MET polymer. NM133 0.01%, NM133 0.02% and NM133 0.05% were stable for 28 days when stored at refrigeration temperature (5–6 °C) and room temperature (16–23 °C), but there was evidence of evaporation of the formulation at 40 °C. There was no change in drug content when NM133 0.05% was stored for 387 days at 4 °C. On topical dosing to rabbits, corneal, conjunctival and scleral AUC_{0-24} levels were 25,780 ng.h g^{-1}, 12,046 ng.h g^{-1} and 5879 ng.h g^{-1}, respectively, with NM133 0.05%. Meanwhile, a similar dose of Restasis 0.05% yielded lower values of 4726 ng.h/g, 4813 ng.h/g and 1729 ng.h/g for the drug corneal, conjunctival and scleral levels, respectively. NM133 thus delivered up to five times more CsA to the ocular surface tissues when compared to Restasis. The MET polymer was non-irritant up to a concentration of 4% *w/v*. The MET polymer is a non-irritant ocular penetration enhancer that may be used to deliver hydrophobic drugs in optically clear topical ocular formulations.

Keywords: cyclosporine A; Molecular Envelope Technology (MET); N-palmitoyl-N-monomethyl-N,N-dimethyl-N,N,N-trimethyl-6-O-glycolchitosan (GCPQ); eye; penetration enhancer

Citation: Uchegbu, I.F.; Breznikar, J.; Zaffalon, A.; Odunze, U.; Schätzlein, A.G. Polymeric Micelles for the Enhanced Deposition of Hydrophobic Drugs into Ocular Tissues, without Plasma Exposure. *Pharmaceutics* **2021**, *13*, 744. https://doi.org/10.3390/pharmaceutics13050744

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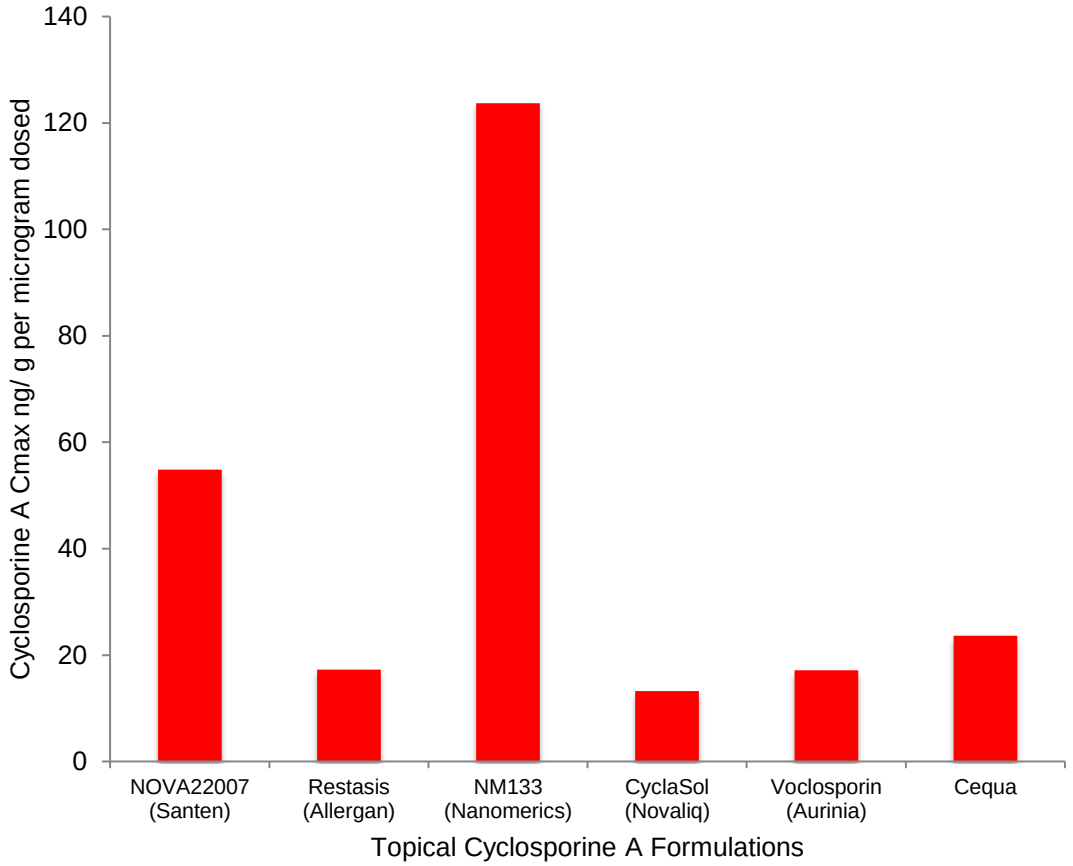
1. Introduction

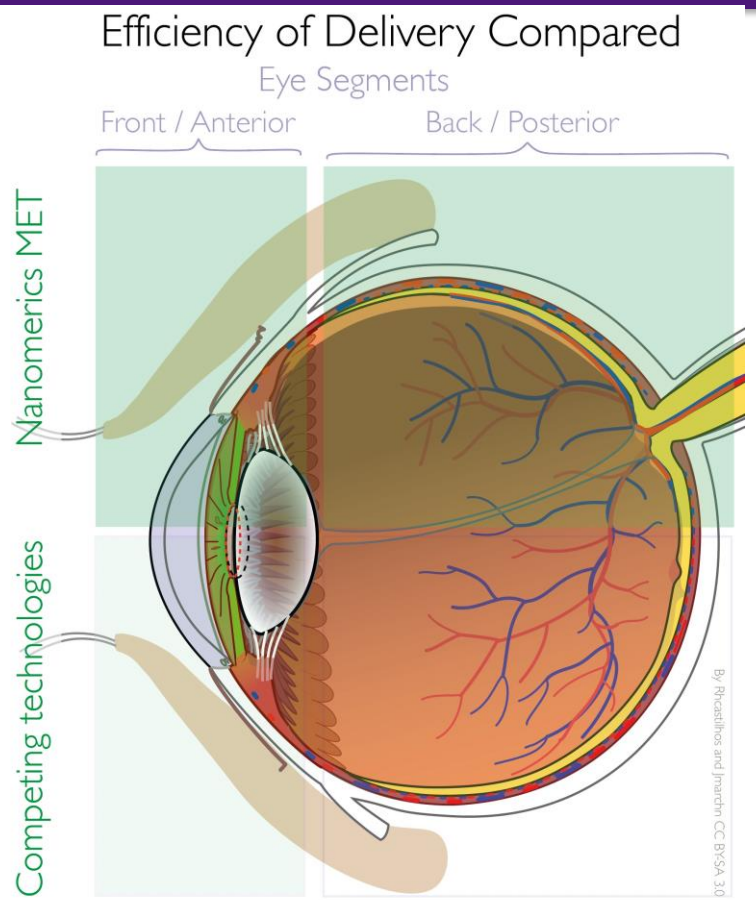
The topical ocular delivery of drugs is usually accomplished using eye drop formulations; however, these formulations have a short ocular residence time, draining through the nasolacrimal duct within 1–3 min [1]. Although ocular clearance may be delayed by up to 30–50 min through the inclusion of viscosifying polymer excipients [1], eye drops are largely inefficient at delivering drugs to the tissue due to the small volume of the eye drop (<50 µL), the rapid clearance of the formulation from the ocular surface and the water and lipid barriers that make up the tear film [2]. Strategies to improve permeation of molecules into the ocular tissues are thus highly desirable. Various nanosystems have been described as experimental formulation agents [3]. However, usually for clinical applications, the formulation of hydrophobic drugs into topical ocular formulations is either



Ocular penetration enhancement

Uchegbu et al, 2021, Pharmaceutics, 13, doi:10.3390/pharmaceutics13050744.

MET Ocular Delivery vs. Eye Drops - Key USPs



Parameter	MET	Suspensions	Emulsions
Hydrophobic drugs	✓	✓	✓
Penetration Enhancement	✓	X	X
Front of eye enhanced	✓	X	X
Back of eye	✓	X	X
Reduced dosing frequency	✓	✓	X
No irritation	✓	Stabiliser specific	X
No loss of visual acuity	✓	✓	X
No need to shake	✓	X	✓
Ease of manufacture	✓	✓	X

OC134

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A polymeric aqueous tacrolimus formulation for topical ocular delivery

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ARTICLE INFO

Keywords:
TAC
MET
Cornea
Chitosan
Allergic ophthalmic disorders

ABSTRACT

Tacrolimus (TAC) suspension is used to treat moderate to severe atopic keratoconjunctivitis (AKC) and vernal keratoconjunctivitis (VKC). The objectives of this study were to formulate the hydrophobic compound TAC (TAC) in an aqueous eye drop formulation and study its ocular biodistribution on topical ocular application to a healthy rabbit model, with the overall aim of using the formulation to treat AKC and VKC. A thin-film hydration method was used to encapsulate TAC within the chitosan-based amphiphile: N-palmitoyl-N-monomethyl-N,N-dimethyl-N,N,N-trimethyl-6-O-glycolchitosan (Molecular Envelope Technology – MET) in an aqueous formulation. The formulation was characterized, and its stability studied under three storage conditions for one month. The ocular distribution of the formulation was studied in healthy rabbits and the ocular tissues and the whole blood analyzed by LC-MS/MS. A 200 nm nanoparticle formulation (MET-TAC) containing 0.1 ± 0.002% w/v TAC was produced with viscosity, osmolarity and pH within the ocular comfort range, and the formulation was stable on refrigeration for one month. On topical application, the TAC concentrations in rabbit cornea and conjunctiva one hour after dosing were 4452 ± 2289 and 516 ± 180 ng/g of tissue, respectively. A topical ocular aqueous TAC eye drop formulation has been prepared with the ability to deliver sufficient drug to the relevant ocular surface tissues.

1. Introduction

Allergic conjunctivitis affects 6–30% of the general population in Europe, with 25% of cases involving severe and persistent disease (Leonardi et al., 2015). Allergic ocular symptoms affect 40% of the US population at least once in their lifetime, with a prevalence rate of 29.7% (Singh et al., 2010). The majority of allergic conjunctivitis patients (55–81%) suffer from seasonal allergic conjunctivitis, while the more severe forms of the disease: atopic keratoconjunctivitis (AKC) and vernal keratoconjunctivitis (VKC) affect 4–39% of allergic conjunctivitis patients, depending on geographical location; with particularly high numbers of VKC (39% of allergic conjunctivitis patients) and AKC (39% of allergic conjunctivitis patients) in Brazil (Leonardi et al., 2015). VKC affects children, resolves around puberty and is more prevalent in boys with a prevalence of 1.16–10.55 per 10,000 of the general population in Western Europe (Bremond-Gignac et al., 2008) and 18% in Nigerian primary school children (Duke et al., 2016). VKC is a sight-threatening disease with no overall gold standard form of therapy (Addis and Jeng, 2018). AKC is also a sight-threatening condition which affects adults mostly and is usually present as a co-morbidity with atopic dermatitis (Guglielmetti et al., 2010), with 67.5% of atopic dermatitis patients diagnosed with AKC in one Japanese study (Dogru et al., 1999). Notwithstanding the rare diseases of AKC and VKC, an estimated 25% of ocular allergy patients have frequent episodes (more than four times a week) for more than four weeks and are classed as having a severe disease which impacts negatively on their quality of life (Leonardi et al., 2015). Patients with severe disease are treated with anti-allergy drugs, and corticosteroids and treatments are frequently inappropriate (Leonardi et al., 2015). Prolonged steroid use is associated with glaucoma (Kersey and Broadway, 2006) and hence is not ideal especially with the younger patients.

TAC acts by binding to FK506 binding protein forming a complex which inhibits calcineurin (Thomson et al., 1995). This inhibition of calcineurin suppresses dephosphorylation of nuclear factor activated T-cells, resulting in suppression of the interleukin 2 gene, interferon-gamma and interleukin 4 and interleukin 5; ultimately inhibiting the proliferation of T-cells (Zhai et al., 2011). TAC also inhibits the release of histamine from mast cells (Sengoku et al., 2000). These mechanisms contribute to the effectiveness of TAC in allergic conjunctivitis. In a 56 patient randomized controlled trial involving severe allergic conjunc-

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0378-5173/

- Topical OC134 (0.1%) eye drops for allergic conjunctivitis
- 15-20% of the population experience allergic conjunctivitis
- 35% of cases not controlled by antihistamines

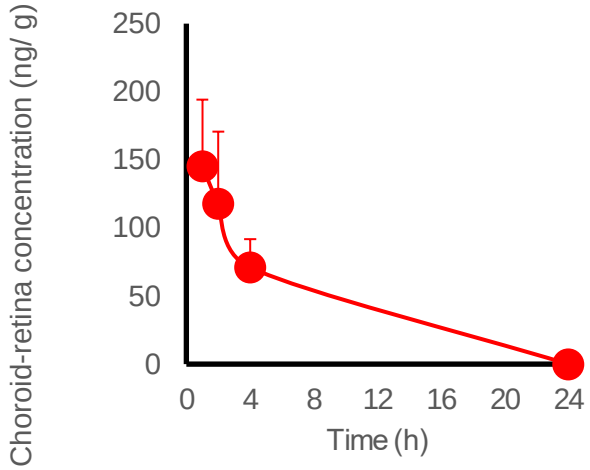
- Superior drug deposition to the commercial preparation
  - ***OC134 delivers 5X more drug to the conjunctiva***
    - *Competitor 0.1% eye drops results in 80ng/ g in the conjunctiva1 hour after dosing.*
  - ***OC134 delivers 18X more drug to the cornea***
    - *Competitor 0.1% eye drops results in 250ng/ g in the cornea 1 hour after dosing.*
  - ***OC134 delivers drug to the aqueous humour***
    - *Competitor 0.1% eye drops are not detectable in the aqueous humour.*
  - ***OC134 does not result in plasma exposure***

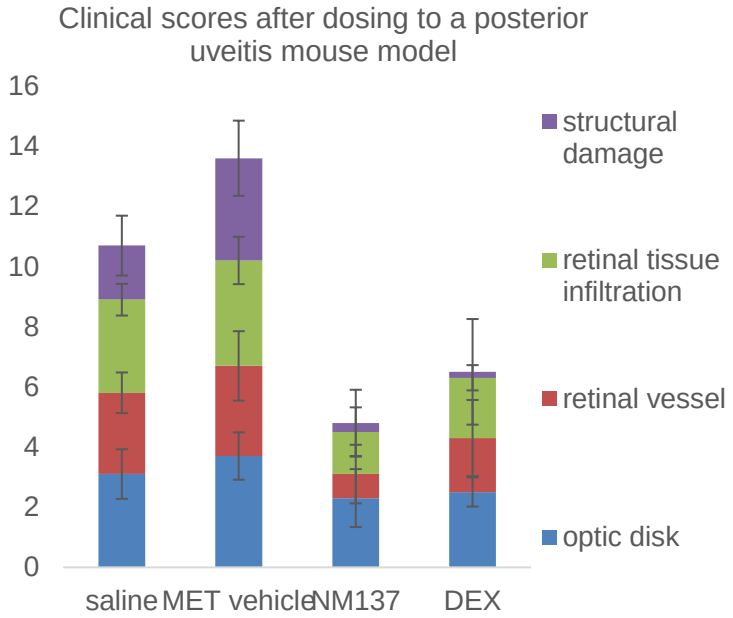
  - Non-opaque formulation
  - Higher response rates anticipated
  - Targeting an orphan drug indication
  - Innovative Licensing Access Pathway (ILAP) passport awarded

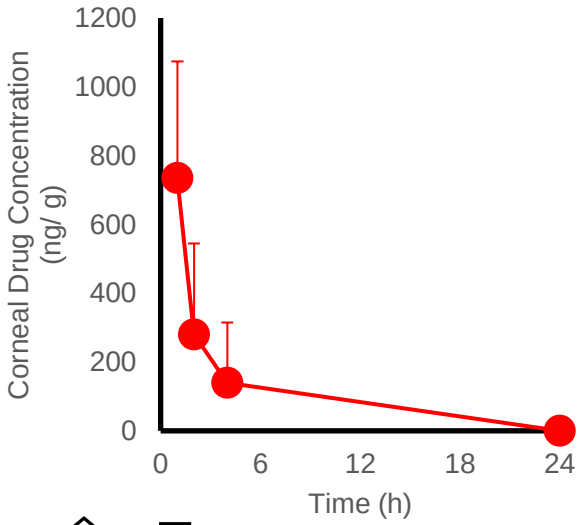
  - **Clinical trials project started in 2021**
    - Quotient Sciences

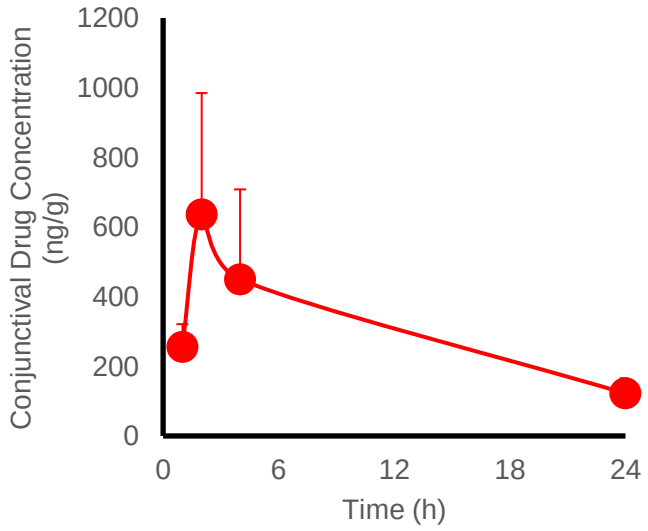
Badr et al, Int J Pharm, 2021,120364. doi:10.1016/j.ijpharm.2021.120364

OC137







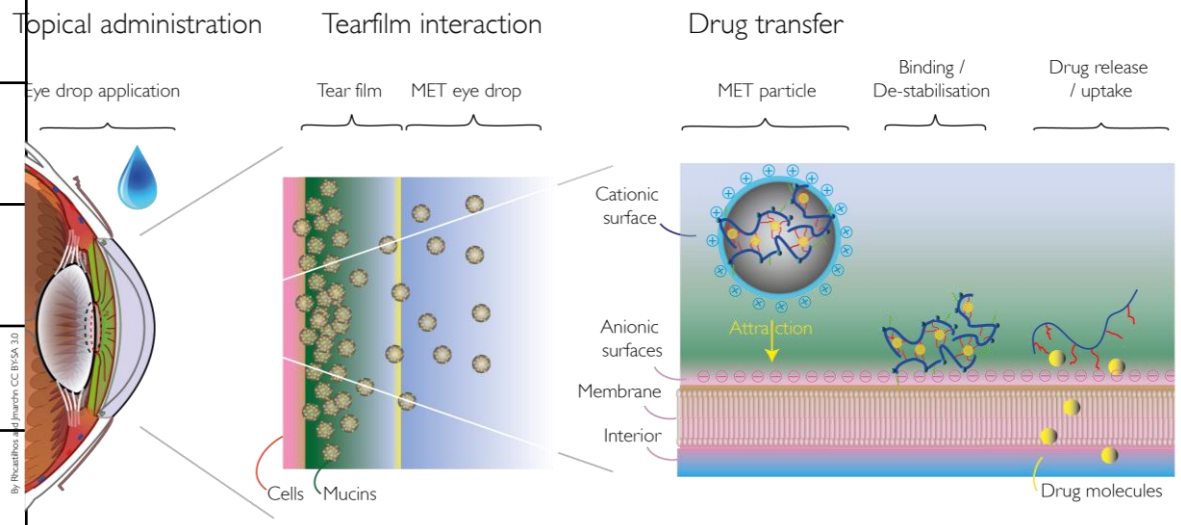


- OC137 (0.2%) eye drops for non-infectious posterior uveitis
- Prevalence of 0.12%, causes 5 – 25% of global blindness
- Drug efficacious on intravitreal delivery
- Topical delivery to the back of the eye with proven preclinical efficacy in a posterior uveitis model
- Upregulates Treg and downregulates Th17
- Superior to the intravitreal formulation that is currently in development
  - Ease of use in the home without the need for medical personnel administered intravitreal injections
  - Targeting an orphan indication
- Status
  - Preclinical proof of concept

Badr et al, Int J Pharm, 2022, 621, 121755

Ocular MET for Hydrophobic Drugs - Competitive Advantage

Parameter	Nanomerics' MET	Drug suspensions	Oil in water emulsions
High hydrophobic drug capacity	✓	X	✓
Formulation ease of manufacture	✓	✓	X
Permeation enhancement	✓	X	X
No ocular irritation	✓	✓/X	X
Optically clear	✓		X
No shaking before use	✓	X	✓



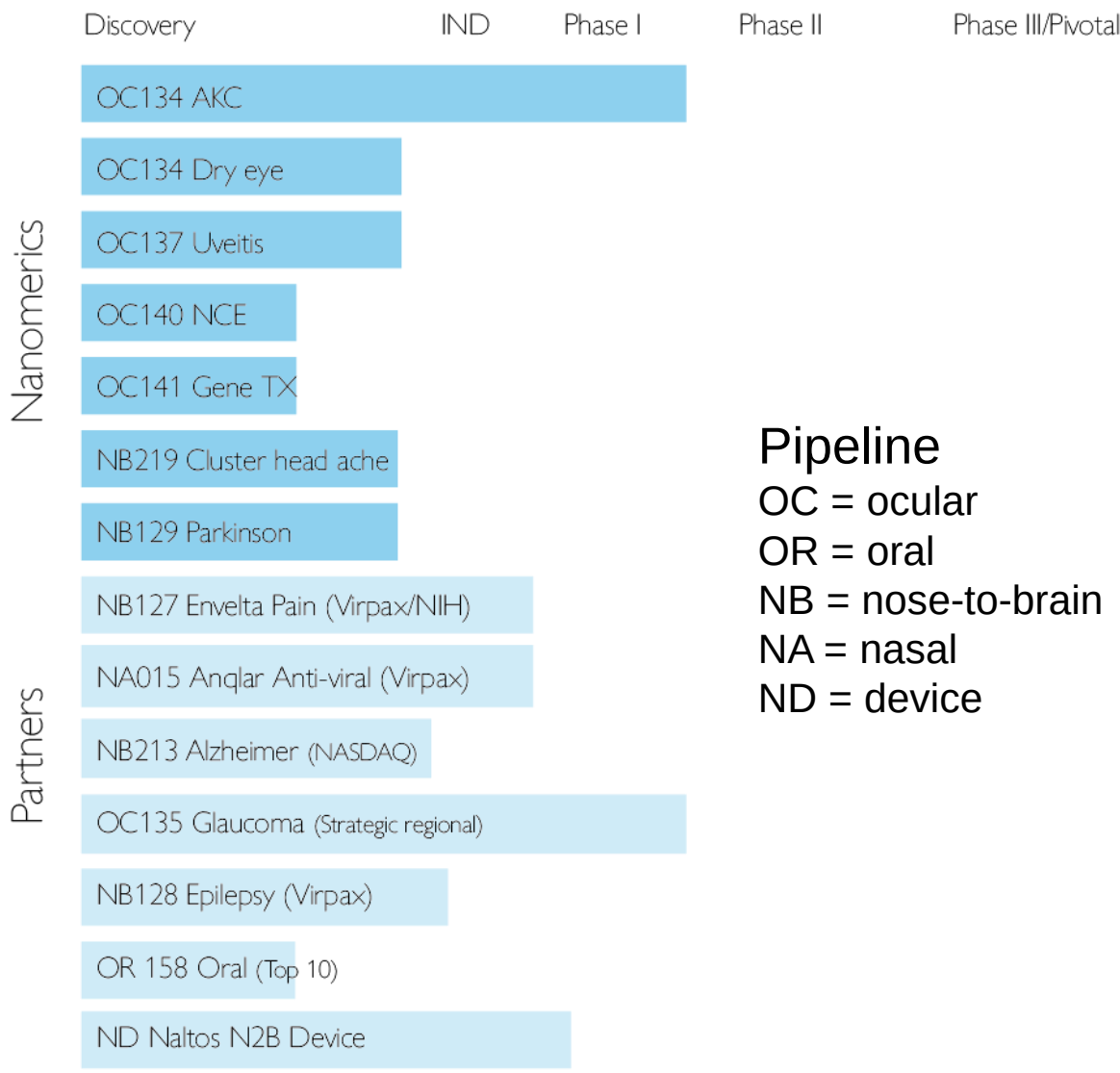
Pipeline with 4 Phase I ready/ IND-enabling assets

- **Ocular own**
  - OC134
- **Ocular partnered**
  - OC135
- **Nose-to-brain partnered**
  - Envelta™ and Naltos device

VIRPAX PHARMACEUTICALS · NIH National Center for Advancing Translational Sciences · alchemypharmatech devices that deliver

- **Nasal partnered**
  - AnQlar

VIRPAX PHARMACEUTICALS



Pipeline
OC = ocular
OR = oral
NB = nose-to-brain
NA = nasal
ND = device

Overview

- Aims and tools
- Gene delivery to the bladder
- Brain delivery
  - Gene delivery to the brain
- Commercialization
  - Envelta™
  - Ocular penetration enhancer
- **Summary**

Summary

Delivering metabolically labile peptide drugs to the brain

Pain therapeutics

Delivering drugs to the retina using eye drops

Reducing the need for intravitreal formulations

Targeting drugs to ocular surface tissues with no plasma exposure using eye drops

Reduced systemic side effects

Delivering nucleic acids to the brain

Neurological disorders

Delivering genes to the bladder

Bladder cancers

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