



LJMU Research Online

Abdellatif, AAH, Scagnetti, G, Younis, MA, Bouazzaoui, A, Tawfeek, HM, Aldosari, BN, Almurshedi, AS, Alsharidah, M, Rugaie, OA, Davies, MPA, Liloglou, T, Ross, K and Saleem, I

Non-coding RNA-directed therapeutics in lung cancer: Delivery technologies and clinical applications

<http://researchonline.ljmu.ac.uk/id/eprint/22937/>

Article

Citation (please note it is advisable to refer to the publisher's version if you intend to cite from this work)

Abdellatif, AAH, Scagnetti, G, Younis, MA, Bouazzaoui, A, Tawfeek, HM, Aldosari, BN, Almurshedi, AS, Alsharidah, M, Rugaie, OA, Davies, MPA, Liloglou, T, Ross, K and Saleem, I (2023) Non-coding RNA-directed therapeutics in lung cancer: Delivery technologies and clinical applications.

LJMU has developed **LJMU Research Online** for users to access the research output of the University more effectively. Copyright © and Moral Rights for the papers on this site are retained by the individual authors and/or other copyright owners. Users may download and/or print one copy of any article(s) in LJMU Research Online to facilitate their private study or for non-commercial research. You may not engage in further distribution of the material or use it for any profit-making activities or any commercial gain.

The version presented here may differ from the published version or from the version of the record. Please see the repository URL above for details on accessing the published version and note that access may require a subscription.

For more information please contact researchonline@ljmu.ac.uk

<http://researchonline.ljmu.ac.uk/>

Tables

Table 1. List of FDA-approved oligonucleotide therapeutics as of 2022, their approval year, indication and administration route.[18] ^(a)

Oligonucleotide type	Brand name, Drug name	Manufacturer	Indication	Administration route	Approval year
ASO	Vitravene (Fomivirsen)	Ionis	Cytomegalovirus infection	Intravitreal	1998 ^(b)
Aptamer	Macugen (Pegaptanib)	Eyetech/Pfizer	Neovascular, elderly macular degeneration	Intravitreal	2004
ASO	Kynamro (Mipomersen)	Ionis	Hypercholesterolemia	Subcutaneous	2013
ASO	Exondys 51 (Eteplirsen)	Sarepta	DMD	Intravenous	2016
ASO	Spinraza (Nusinersen)	Ionis	Spinal muscular atrophy	Intrathecal	2016
ssON	Defitelio (Defibrotide)	Jazz	Hepatic veno-occlusive disease	Intravenous	2016
ASO	Tegsedi (Inotirsen)	Ionis	hATTR	Subcutaneous	2018
siRNA	Onpattro (Patisiran)	Alnylam	hATTR	Intravenous	2018
ASO	Waylivra (Volanorsen)	Ionis /Akcea	Familial chylomicronaemia syndrome	Subcutaneous	2019
ASO	Vyondys 53 (Golodirsen)	Sarepta	DMD	Subcutaneous	2019
siRNA	Givlaari (Givosiran)	Alnylam	Acute hepatic porphyriasis	Subcutaneous	2019
ASO	Viltolarsen (Viltepso)	NS pharma	DMD	Intravenous	2020
siRNA	Oxlumo (Lumasiran)	Alnylam	Primary hyperoxaluria type 1	Subcutaneous	2020
ASO	Amondys 45 (Casimersen)	Sarepta	DMD	Subcutaneous	2021
siRNA	Leqvio (Inclisiran)	Alnylam /Novartis	Hypercholesterolemia	Subcutaneous	2021

^(a) The data were collected from FDA database and classified by the authors.

^(b) The drug was discontinued in 2002.

Abbreviations: ASO, Anti-sense oligonucleotide; ssON, Single-stranded oligonucleotide; siRNA, Small interfering RNA; hATTR, Hereditary transthyretin-mediated amyloidosis; DMD, Duchenne muscular dystrophy.

Table 2. A summary of the major challenges that encounter ncRNAs-targeting oligonucleotide therapeutics and some recent approaches to tackle them.

Nature of the challenge	Challenge	Approach to overcome	References
Experimental	Poor <i>in vitro-in vivo</i> correlation	3D cell culture models Dynamic culture models	[7] [192]
<i>In vivo</i>	Poor stability Poor pharmacokinetics Immunogenicity	Chemically-modified oligonucleotides Nanocarriers	[193] [7, 19, 20]
	Poor selectivity	Targeted delivery systems	[4, 9]
Cellular	Poor cellular uptake	Targeted delivery systems Cell-penetrating peptides	[9, 191] [194]
Intracellular	Lysosomal degradation	Endosomal escape devices Fusogenic lipids	[6] [151]
	Poor nuclear penetration	Nuclear-localization signals	[114]
Translational	Poor scale-up	Ligand-free targeting Microfluidic devices	[5] [7, 14]
	Poor animal-human correlation	Representative animal models Patient-derived xenografts	[4, 195] [196, 197]

Table 1. oligonucleotide therapeutics delivered by local routes of administration that reached advanced clinical trials.[96] ^(a)

Oligonucleotide type	Oligonucleotide name	Administration route	clinical Phase	Indication	ClinicalTrials.gov Identifier
ASO	Mongersen	Orally	Phase II ^(b)	Chron	NCT02685683
ASO	Exc 001	Intradermally	Phase IIb	Hypertrophic scar	NCT01038297
ASO	Alicaforsen	Rectally	Phase III	Chronic antibiotic refractory pouchitis	NCT02525523
miRNA mimic	Remlarsen	Intradermally	Phase II	Skin Fibroplasia	NCT03601052
siRNA	Cotsiranib	Intradermally	Phase II	Hypertrophic scar	NCT02956317

(a) The data were collected from National Institute of Health (NIH) database and classified by the authors.

(b) Withdrawn later due to lack of effectiveness.

Table 4. A summary of the most common chemical modifications of oligonucleotides, their description, and the advantages and disadvantages they brought about in the oligonucleotide therapeutics.

Site of modification	Mode of modification	Description	Advantages	Disadvantages	Application
Nitrogenous base	Phosphorothioate (PS)	Non-bridging oxygen atom is replaced with sulphur group	Improved stability against nucleases; Improved pharmacokinetics; Increased binding to plasma proteins; Increased hydrophobicity	Reduced binding affinity to the mRNA targets	ASO
	Phosphoester	Neutral phosphoester oligonucleotides are incorporated within siRNA to generate a prodrug (short interfering ribonucleic neutral molecules)	Massive RNAi response (due to the cleavage of Phosphoester group once the molecule is internalized)	-	siRNA
Sugar	Peptide nucleic acids (PNA)	Sugar backbone is replaced with synthetic poly ethyleneimine scaffold with nucleobase acetic acid connected to every second backbone nitrogen atom via amine bond	Improved stability against nucleases and proteases; Stronger affinity to RNA targets	-	siRNA, ASO
	2'-OMe	2'-OH group of the ribose is replaced with 2'-methoxy group	Increased affinity to RNA; Increased stability against nucleases	Sensitivity to serum nucleases	
	2'-F	2'-OH group of the ribose is replaced with 2'-fluoro	Increased binding affinity to RNA	-	

2'-MOE	2'-OH group of the ribose is replaced with 2'-methoxyethoxy group	Increased affinity to RNA; Increased serum stability; Increased miRNA inhibitory activity		
Locked Nucleic Acid (LNA)	2'-OH group of the ribose is replaced with 2',4'-o-methylene bridge	Increased RNA affinity (reached by reducing the conformational flexibility of nucleotides)	Anti-miRNA activity is only slightly higher	
Combination of chemical modifications	More than one of the abovementioned modifications co-exist in the same oligonucleotide molecule	Combined benefits of multiple modifications in a single molecule	Complexity of synthesis	All types

Abbreviations: RNAi, RNA interference.

Table 5. Preclinical studies regarding oligonucleotide therapeutics delivered via nanoparticles as novel treatments for lung cancer.

Study	Context	oligonucleotide cargo	Nanocarrier type	Delivery route/animal model
(Loira-Pastoriza <i>et al.</i> , 2021)[198]	Immunotherapy of metastatic lung cancer	Phosphorothioate-linked CpG ODNs (C274 and B1826)	Cationic liposomes	Intraperitoneally or intratracheally in murine model of metastatic lung cancer
(Perry <i>et al.</i> , 2020)[104]	Immunotherapy of metastatic lung cancer	CpG ODNs	Polymeric nanoparticles	Orotracheal instillation in mice instilled with KAL-LN2E1 cells
(Zhang <i>et al.</i> , 2020)[153]	Targeted delivery of anticancer, homoharringtonine (HHT), to lung cancer cells	EGFR aptamer (as a targeting ligand)	Stimuli-responsive PLGA nanoparticles	Intraperitoneal injection in A549 athymic mouse xenograft
(Cheng <i>et al.</i> , 2018)[199]	Dual Modulation of Bcl-2 and Akt-1 in Lung and Cervical Carcinomas	ASO	T7 Peptide-Conjugated LNPs	Tail vein injection (IV) in A549 athymic mouse xenograft
(Yang <i>et al.</i> , 2018)[152]	Anti-MicroRNA-155 delivery for lung cancer therapy	miR-155 inhibitor	PEA NPs	IV in A549 athymic mouse xenograft
(Cheng <i>et al.</i> , 2017)[200]	Knocking down Bcl-2 for treatment of lung cancer	Chemically-modified ASO (G3139-GAP)	LNPs	IV in A549 athymic mouse xenograft
(Yung <i>et al.</i> , 2016)[150]	Therapeutic delivery of AntimiR-21 for lung cancer	Phosphorothioate-modified antimiR-21	LNPs	IV in A549 athymic mouse xenograft
(Garbuzenko <i>et al.</i> , 2010)[201]	Targeting MRP1 and BCL2 to suppress chemoresistance in lung cancer	ASO	Liposomes	Inhalation in an orthotopic murine model of human lung carcinoma

Abbreviations: PEA, Poly(ester amine); EGFR, Epidermal growth factor receptor; MRP1, Multidrug resistance-associated protein 1; Bcl-2, B-cell lymphoma 2 protein.

Table 6. Some recent studies on chitosan modifications which are beneficial for oligonucleotides delivery.

Study	Cargo	Application	Chitosan modification	Experimental model	Advantages of the modification
(Motiei <i>et al.</i> , 2021)[202]	miRNA (miR-34a)	Breast cancer	Chitosan grafted with PGA	MDA-MB-231 cells	Increased core stability against pH variation and improved the nanoparticle loading capacity
(Khatami <i>et al.</i> , 2021)[158]	Anti-miR-21	Colorectal cancer	Chitosan functionalised with anti-miR-21	MCF-7, C26 cells, and mice	Increased uptake of nanoparticles into target cells, reduced tumor growth <i>in vivo</i>
(G. Huang <i>et al.</i> , 2021)[168]	Anti-miR-21	Tyrosine kinase inhibitors-resistant NSCLC	α -Linolenic acid-modified chitosan	H1975 cells and mice	Increased nucleic acid complexation, cellular uptake, and tumor accumulation
(Zhu <i>et al.</i> , 2020)[167]	MB ONT	Detection of miR-155-5p and imaging of lung cancer	Chitosan is self- assembled with MB ONT	A549, SPC-A1, H446 cells, and mice	High detection and imaging efficiency
(Dowaidar <i>et al.</i> , 2017)[203]	siRNA	Gene delivery to cancer cells	Chitosan conjugated with a cell-penetrating peptide and used to coat Fe ₃ O ₄ NPs	HeLa cells	Improved colloidal stability and transfection efficiency
(Dong <i>et al.</i> , 2011)[166]	ASO	Inhibition of telomerase activity in lung cancer	Chitosan-coated PLGA NPs	A549, primary NSCLC, and patient-derived tissues	High complexation of ASO and high delivery efficiency
(Nafee <i>et al.</i> , 2007)[164]	ASO	Gene delivery to cancer cells	Chitosan-coated PLGA NPs	A549 cells	Improved colloidal characteristics of NPs, high loading capacity, increased cellular uptake

Abbreviations: PGA, Polyglutamate; NSCLC, Non-small cell lung cancer; MB ONT, Molecular Beacon Oligonucleotide.