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Review

Chromatographic methods for the determination of therapeutic oligonucleotides☆

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ABSTRACT

Both DNA and RNA are being explored for their therapeutic potential against a wide range of diseases. As these new drugs emerge, new demands arise for the analysis and quantitation of these biomolecules. Pharmacokinetic and pharmacodynamic analysis requirements for drug approval place enormous challenges on the methods for analyzing these therapeutics. This review will focus on bioanalytical methods for DNA antisense and aptamers as well as small-interfering RNA (siRNA) therapeutics. Chromatography methods employing ultraviolet (UV), fluorescence and mass spectrometric (MS) detection along with matrix-assisted laser desorption/ionization (MALDI) will be covered. Sample preparation from biological matrices will be reviewed as well as metabolite analysis and identification. All of these techniques are important contributions toward oligonucleotide therapeutic development. They will also be important in microRNA (miRNA) biomarker discovery and RNomics in general, as more non-coding RNAs are inevitably discovered.

Contents

1. Introduction

Therapeutic oligonucleotides (oligos) have emerged as promising candidates for drug therapies for a wide range of diseases, including cancer, AIDS, Alzheimer's disease and cardiovascular disorders [1]. They have also become indispensible tools for genomic studies allowing for specific knockdown of proteins to study signaling pathways and identify therapeutic targets. Currently, there are two DNA therapeutics approved by the US FDA. Fomivirsen is an antisense oligonucleotide for the treatment of cytomegalovirus retinitis [2]. Pegaptanib is an aptamer for the treatment of neovascular age-related macular degeneration [3]. Table 1 shows the many DNA aptamer and antisense drugs, as well small-interfering RNA (siRNA) therapeutics that have been or are currently being used in human clinical trials [4].

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Table 1
Oligonucleotide therapeutics involved in clinical trials. Information includes the drug name, company sponsoring the clinical trial, the status, phase, and route of administration.

Drug	Company	Status	Phase	Administration	Disease
siRNA therapeutics					
TD101	Transderm, Inc.	Completed	I	Local injection	Pachyonychia Congenita
AGN211745	Allergan/Sirna	Completed	I/II	Local injection	Age-related macular degeneration; Choroidal neovascularization
AGN211745		Terminated	I	Intravitreal injection	Age-related macular degeneration; Choroidal neovascularization
CALAA-01	Calando Pharmaceuticals	Completed	II	IV injection	Cancer; Solid tumor
Atu027	Silence Therapeutics AG	Recruiting	I	IV injection	Advanced solid tumors
Bevasiranib	Opko Health, Inc.	Completed	II	Intravitreal injection	Diabetic macular edema
	Opko Health, Inc.	Completed	II	Intravitreal injection	Wet age-related macular degeneration
QPI-1007	Quark Pharmaceuticals	Recruiting	I	Local injection	Chronic optic nerve atrophy; Non-arteritic anterior ischemic optic neuropathy
PRO-040201	Tekmira	Terminated	I	IV injection	Hypercholesterolemia
siG12D	Silenseed	Not yet recruiting	I	Local Drug EluteR	Adenocarcinoma of the pancreas
I5NP	Quark Pharmaceuticals	Recruiting	III	IV injection	Delayed graft function in kidney transplantation
	Quark Pharmaceuticals	Active	I	IV injection	Kidney injury, Acute renal failure
SYL040012	Sylentis, S.A.	Recruiting	I/II	Opthalmic drops	Glaucoma, Ocular hypertension
Aptamer therapeutics					
ARC1905	Ophthotech Corporation	Active	I	Intravitreal injection	Dry age-related macular degeneration
E10030	Ophthotech Corporation	Recruiting	II	Intravitreal injection	Neovascular age-related macular degeneration
ARC1905	Ophthotech Corporation	Active	I	Intravitreal injection	Neovascular age-related macular degeneration
EYE001	Eyetech Pharmaceuticals	Recruiting	II/III	Intravitreal injection	Neovascular age-related macular degeneration
REG1	National Heart, Lung, and Blood Institute	Completed	I	IV injection	Anticoagulation system
Pegaptanib sodium (Macugen)	Eyetech Pharmaceuticals	Completed	IV	Intravitreal injection	Exudative age-related macular degeneration
AS1411	Antisoma Research	Recruiting	II	IV injection	Acute myeloid leukemia
NOX-E36	Noxxon Pharma AG	Completed	III	IV and Subcutaneous	Chronic inflammatory diseases, Type 2 diabetes mellitus, Systemic lupus erythematosus
NOX-A12	Noxxon Pharma AG	Recruiting	III	IV injection	Hematopoietic stem cell transplantation
ARC1779	Archemix Corp.	Completed	II	IV injection	Von Willebrand factor-related platelet function disorders
Bevacizumab	Medical University of Vienna	Recruiting	III	Intraocular injection	Diabetic retinopathy
ARC19499	Archemix Corp.	Not yet recruiting	I/II	Subcutaneous injection	Hemophilia
Antisense therapeutics					
EGFR Antisense DNA	University of Pittsburgh	Not yet recruiting	I/II	Intratumoral injection	Head and neck squamous cell carcinoma
	University of Pittsburgh	Active	I	Intratumoral injection	Head and neck squamous cell carcinoma
AEG35156	Aegera Therapeutics	Recruiting	I/II	IV injection	Advanced hepatocellular carcinoma
	Aegera Therapeutics	Recruiting	I/II	IV injection	Chronic lymphocytic leukemia
	Aegera Therapeutics	Terminated	I/II	IV injection	Advanced pancreatic cancer
	Aegera Therapeutics	Terminated	I/II	IV injection	Advanced breast cancer
	Aegera Therapeutics	Terminated	I/II	IV injection	Advanced Non-small cell lung cancer
	Aegera Therapeutics	Terminated	II	IV injection	Advanced cancer
	Aegera Therapeutics	Active	II	IV injection	Leukemia