

GRAFT COPOLYMER POLYELECTROLYTE COMPLEXES FOR DRUG DELIVERY

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application is a Continuation-in-Part under 35 U.S.C. §120 of U.S. patent application Ser. No. 12/744,824, filed on May 26, 2010, which is the U.S. National Stage application under 35 U.S.C. §371 of International Application Serial No. PCT/US2008/84995, filed on Nov. 26, 2008, which claims the benefit of priority under 35 U.S.C. §119(e) of U.S. Provisional Application Ser. No. 60/990,606, filed on Nov. 27, 2007.

[0002] This application also claims the benefit of priority under 35 U.S.C. §119(e) of U.S. Provisional Application Ser. No. 61/619,234, filed on April 02, 2012. The entire disclosures of all of the above applications are incorporated herein by reference.

STATEMENT REGARDING FEDERALLY SPONSORED RESEARCH OR DEVELOPMENT

[0003] This invention was made with government support under Contract No. W81XWH-10-2-0139 and the FY10 Defense Medical Research and Development Program Basic Research Award for Project D61_I_10_J2_235, both of which were awarded by the Department of Defense. The government has certain rights in the invention.

FIELD OF THE INVENTION

[0004] This invention relates to materials and methods for intracellular delivery of drug molecules which are otherwise difficult to deliver, including for example, therapeutic cationic peptides, oligonucleotides, other nucleic acids, plasmid DNA encoding genes or ribozymes, peptide nucleic acids, aminoglycoside antibiotics, glycopeptide antibiotics, and lipopeptide antibiotics. Further, co-delivery of other therapeutic agents, including for example, local anesthetics, anti-tumor agents, imaging agents, fluorescent dyes and quantum dots, can be achieved via incorporation of a liposome.

BACKGROUND OF INVENTION

[0005] The ability of exogenously administered nucleotide molecules to mediate gene silencing was discovered nearly 30 years ago. Ever since, this technology has been utilized as a research tool to study gene function, and over time it has been developed for treatment of diseases arising from the abnormal over-expression or over-activity of a particular gene, such as cancer, autoimmune and cardiovascular diseases, wound healing and viral infections. This technology, referred to herein as antisense therapeutics, includes a range of technologies differentiated by the approaches they use to break down the mRNA. Approaches of interest include, for example, RNA interference (RNAi), micro-RNA, and the use of conventional antisense deoxynucleotide technologies.

[0006] Further progress in this field requires improvement in the systemic and cellular delivery of these antisense therapeutics to their targets. Some of the barriers at the systemic level include survival against unfavorable interactions with serum proteins present in the bloodstream, avoidance of accumulation in non-target organs such as lung, liver and kidney, and targeting of the diseased or infected cells. Once the antisense therapeutics have overcome these barriers, they must maneuver their way into the target cell and finally to the target

mRNA within the cell. Some of the challenges to antisense therapeutic delivery at the cellular level include efficient entry into the cell, escape from degradative lysosomes, and release into the cytoplasm.

[0007] While viral vectors are also being used for delivery of such antisense therapeutics and gene delivery, safety concerns persist. Although iterative design of non-viral vectors has endowed them with attributes for overcoming some of the systemic and cellular barriers in the delivery of antisense therapeutics, their delivery efficiencies are generally too low and their cytotoxicities are generally too high. A major barrier to the intracellular delivery of antisense therapeutics is their sequestration in endosomes, which eventually fuse with lysosomes, leading to degradation of their contents.

[0008] Accordingly, there is a continuing need in the art for methods and materials that improve intracellular delivery of antisense therapeutics, as well as oligonucleotides and other nucleic acids in general.

[0009] Similarly, there is a need in the art for methods and materials to improve intracellular delivery of cationic peptides, peptide nucleic acids, and various antibiotic molecules (aminoglycosides, glycopeptides and lipopeptide antibiotics), with or without co-delivery of other therapeutic agents (drugs).

BRIEF SUMMARY OF THE INVENTION

[0010] A first aspect of the invention provides innovative graft polymers designed for the efficient delivery of polynucleotides into biological cells, and for maintaining the biological activity of these molecules while in serum and other aqueous environments. Polynucleotides can include plasmid DNA, synthetic natural or chemically modified oligonucleotides used for gene silencing via antisense or RNA interference mechanisms, ribozymes, aptamers, microRNAs, decoys, etc. Such polymers can comprise an anionic graft polymer comprising an anionic polymer backbone with pendant carboxylic acid groups and pendent chains comprising amphipathic or hydrophilic polymers covalently bonded to a portion of said pendent carboxylic acid groups. The instant polymers preferably have a graft density of between about 0.1 and about 25 mole percent, more preferably between about 0.5 and about 10 mole percent, and most preferably between about 0.5 to about 5 mole percent.

[0011] Suitable anionic backbone polymers include, but are not limited to, polyanhydrides, poly(acrylic acids), poly(alkylacrylic acids), carboxymethylcellulose, polyglutamic acids, polyaspartic acids, vinyl copolymers, or combinations thereof. In the preferred embodiments the backbone of the instant polymer comprises poly(propylacrylic acid) or poly(methacrylic acid). Suitable polymers for use as pendent chains, include but are not limited to, polyetheramines, poly(alkylene oxides), or combinations thereof.

[0012] In one specific embodiment, such polymers can comprise a backbone comprising a poly(alkylacrylic acid) and one or more polyetheramine pendent chains covalently attached to said polymer backbone via said acrylic acid groups predominantly comprising ethylene oxide repeating units, wherein said polymer has a graft density between about 0.1 and about 25 mole percent.

[0013] A second aspect of the invention provides a vector for intracellular delivery of therapeutic polynucleotide molecules, including but not limited to antisense molecules, comprising a graft polymer as described above and at least one cationic agent for delivery of polynucleotides. Suitable cat-