

SUSTAINED DELIVERY OF POLYIONIC BIOACTIVE AGENTS

FIELD OF THE INVENTION

The field of the invention is drug delivery, including nucleic acid delivery.

BACKGROUND OF THE INVENTION

Delivery of one or more types of biologically active molecule to an animal or to an animal tissue forms the basis of modern pharmacology. In order to achieve the fullest therapeutic or prophylactic effect, the composition and method used to deliver the bioactive agent must provide the proper amount of the agent to the appropriate tissue(s) of the animal, in an active or activatable form, at an appropriate point in time, and for a sufficient duration. Despite thousands of years of pharmacological research and practice, there remains a critical need for compositions and methods of delivering polyionic bioactive agents, particularly in a sustained-release manner.

Numerous polyionic bioactive agents are known in the art, including both polyanionic and polycationic bioactive agents. Nucleic acids, in particular, have proven difficult to deliver effectively to the animal and in a sustained-release manner. The difficulties in delivering nucleic acids have persisted despite the intense and increasing desire of researchers and clinicians to be able to deliver nucleic acids to animal tissues, particularly to human tissues. In December, 1995, the U.S. National Institutes of Health issued a "Report and Recommendations of the Panel to Assess the NIH Investment in Research on Gene Therapy (Orkin et al., 1995, National Institutes of Health, Bethesda, Md.). In this Report, it was recognized that the development of gene therapy approaches to disease treatment was being inhibited, in part, by a dearth of effective gene transfer vectors. The Report recognized a need for further research applied to improving vectors for gene delivery.

Among the physiological phenomena which can inhibit administration of a nucleic acid to an animal tissue are the following.

Inability to direct the nucleic acid to cells of the desired tissue.

Inability of the nucleic acid to cross membranes of cells of the desired tissue.

Nucleolytic digestion of the nucleic acid prior to its delivery to cells of the desired tissue.

Nucleolytic digestion of the nucleic acid within cells of the desired tissue prior to transfer of the nucleic acid to a location within the cells at which the nucleic acid may exert its intended effect.

Clearance of the nucleic acid from the animal's system before the nucleic acid has been delivered to a sufficient fraction of cells of the desired tissue.

Inability to achieve an adequate dosage of the nucleic acid at the desired tissue.

A desirable nucleic acid vector will permit administration that is not significantly inhibited by these phenomena.

Numerous compositions and methods are known for delivering a nucleic acid to an animal tissue. Such compositions include "naked" (i.e. non-complexed) nucleic acids, nucleic acids complexed with cationic molecules such as polylysine and liposome-forming lipids, and virus vectors.

Naked nucleic acids can be taken up by various animal cells, but are subject to nucleolysis, both inside and outside of cells that take them up. For example, it is known that cells

in wounded tissue (e.g. cells lining an incision made in a tissue) are particularly amenable to taking up naked nucleic acids. Examples of such cells include, but are not limited to, fibroblasts, capillary endothelial cells, capillary pericytes, mononuclear inflammatory cells, segmented inflammatory cells, and granulation tissue cells.

The use of nucleic acid analogs which are relatively resistant to nucleolysis is known. Such analogs include, for example, phosphorothioate nucleic acid analogs. However, in some situations, particularly where incorporation of the nucleic acid into the genome of the target cell is desired, the use of nucleic acid analogs can be undesirable. Targeting of naked nucleic acid vectors to particular animal tissues can be difficult, particularly in situations in which the tissue is normally bathed by a liquid in which the vector may be carried away from the tissue site.

Compositions for sustained release of naked nucleic acids are known, but such compositions have many of the same drawbacks of other naked nucleic acid vectors, namely, that the nucleic acids released from the compositions may not be efficiently taken up by cells of the desired tissue and that the nucleic acids released from the compositions are susceptible to nucleolysis. Examples of such compositions include compositions comprising naked nucleic acids in a biodegradable polymer matrix. Another shortcoming of such compositions is that they can be difficult to target to specific tissues in order to achieve localized delivery of the nucleic acid. Such compositions generally occur in liquid form, which must be injected at the desired site, but is capable of flowing from the site of administration to other sites.

Numerous vectors comprising a nucleic acid complexed with a compound to improve stability or uptake of the nucleic acid by a target cell have been described. Such compounds include, by way of example, calcium phosphate, polycations such as diethylaminoethyl-dextran, polylysine, or polybrene, and liposome-forming lipids such as didocylmethylammonium bromide and Lipofectamine™. Many of these compounds are toxic, or produce undesired reactions, when administered to patients. Thus, while nucleic acid vectors comprising a nucleic acid complexed with one of these compounds may be useful for transfection of cultured cells, these vectors are not useful for delivering nucleic acids to cells in an animal tissue.

Virus vectors are generally regarded as the most efficient nucleic acid vectors. Recombinant replication-defective virus vectors have been used to transduce (i.e., infect) animal cells both in vitro and in vivo. Such vectors have included retrovirus, adenovirus, adeno-associated virus vectors, and herpes virus vectors. While highly efficient for gene transfer, a major disadvantage associated with the use of virus vectors is the inability of many virus vectors to infect non-dividing cells. Another serious problem associated with the use of virus gene vectors is the potential for such vectors to induce an immune response in a patient to whom they are administered. Such an immune response limits the effectiveness of the virus vector, since the patient's immune system rapidly clears the vector upon repeated or sustained administration of the vector. Furthermore, insertion of a gene into the genome of a cell by a virus vector may induce undesirable mutations in the cell. Other problems associated with virus gene vectors include inability to appropriately regulate gene expression over time in transfected cells, potential production and transmission to other humans of harmful virus particles, local and general toxicity, undesirable immunogenicity, and unintended disruption of target or other cell metabolism.

What is needed are compositions and methods which can be used to deliver nucleic acids to cells of a desired tissue in