CPG AMPHIPHILES AND USES THEREOF

SEQUENCE LISTING

The instant application contains a Sequence Listing which has been submitted electronically in ASCII format and is hereby incorporated by reference in its entirety. Said ASCII copy, created on _____ is named _____ and is _____ bytes in size.

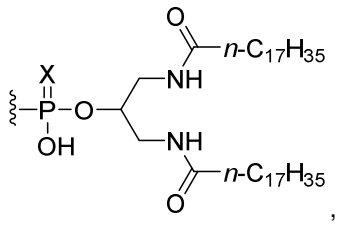
BACKGROUND OF THE INVENTION

Human papillomavirus (HPV)-related cancer is one of the fastest growing cancers in the world. Overall, 5% of all cancers world-wide can be attributed to HPV infections. There continues to be a need for further and more effective cancer treatments.

SUMMARY

The disclosure provides compounds that can be used in therapeutic methods.

Accordingly, in the first aspect, the disclosure features a compound consisting of the nucleotide sequence 5'-TCGTCGTTTTGTCGTTTTGTCGTT-3' (SEQ ID NO:1), at its 5' end, bonded or linked by a linker to the following lipid:



,

or a salt thereof,

where X is O or S.

In one example of the first aspect of the disclosure, the nucleotide sequence is bonded to the lipid.

In another example of the first aspect of the disclosure, all internucleoside groups connecting the nucleosides in 5'-TCGTCGTTTTGTCGTTTTGTCGTT-3' (SEQ ID NO:1) are phosphorothioates.

In the second aspect, the disclosure features use of the compound of the first aspect in the manufacture of a medicament for use in combinatorial therapy for treating a cancer in a human patient, wherein the medicament is to be administered in combination with a protein comprising the amino acid sequence: MHQKRTAMFQ DPQERPRKLP QLCTELQTTI HDIILECVYC KQQLLRREVY DFAFRDLCIV YRDGNPYAVG DKCLKFYSKI SEYRHYCYSL YGTTLEQQYN KPLCDLLIRC INGQKPLCPE EKQRHLDKKQ RFHNGRGRWT GRCMSCCRSS RTRRETQL (SEQ ID NO:2), and a protein comprising the amino acid sequence: MHGDTPTLHE YMLDLQPETT DLYGYGQLND SSEEEDEIDG PAGQAEPDRA HYNIVTFCCK CDSTLRLCVQ STHVDIRTLE DLLMGTLGIV CPICSQKP (SEQ ID NO:3). Also disclosed is a method of treating a cancer in a human patient. This method includes administering to the patient the compound of the first aspect of the disclosure, a protein including the amino acid sequence: MHQKRTAMFQ DPQERPRKLP QLCTELQTTI HDIILECVYC KQQLLRREVY DFAFRDLCIV YRDGNPYAVG DKCLKFYSKI SEYRHYCYSL YGTTLEQQYN KPLCDLLIRC INGQKPLCPE EKQRHLDKKQ RFHNGRGRWT GRCMSCCRSS RTRRETQL (SEQ ID NO:2), and a protein including the amino acid sequence: MHGDTPTLHE YMLDLQPETT DLYGYGQLND SSEEEDEIDG PAGQAEPDRA HYNIVTFCCK CDSTLRLCVQ STHVDIRTLE DLLMGTLGIV CPICSQKP (SEQ ID NO:3).

In one example of the second aspect of the disclosure, the cancer is human papillomavirus (HPV) positive (e.g., HPV type 16 positive).

In another example of the second aspect of the disclosure, the cancer is a head or neck squamous cell carcinoma.

In an additional example of the second aspect of the disclosure, the patient is receiving or has received platinum-containing chemotherapy. In a further example, an anti-PD-1 antibody (e.g., pembrolizumab or nivolumab) is administered to the patient.

In another example of the second aspect of the disclosure, the compound of the first aspect of the disclosure and the proteins including the amino acid sequences of SEQ ID NO:2 and SEQ ID NO:3 are administered concurrently.

In a further example of the second aspect of the disclosure, the compound of the first aspect of the disclosure and the proteins including the amino acid sequences of SEQ ID NO:2 and SEQ ID NO:3 are administered sequentially.

In a further example, the disclosure features another method of treating a cancer in a human patient. This method includes administering to the patient the compound of the first aspect of the disclosure, a protein including the amino acid sequence:
MHQKRTAMFQ DPQERPRKLP QLCTELQTTI HDIILECVYC KQQLLRREVY DFAFRDLCIV YRDGNPYAVG DKCLKFYSKI SEYRHYCYSL YGTTLEQQYN KPLCDLLIRC INGQKPLCPE EKQRHLDKKQ RFHNGRGRWT GRCMSCCRSS RTRRETQL (SEQ ID NO:2), a protein including the amino acid sequence:
MHGDTPTLHE YMLDLQPETT DLYGYGQLND SSEEEDEIDG PAGQAEPDRA HYNIVTFCCK CDSTLRLCVQ STHVDIRTLE DLLMGTLGIV CPICSQKP (SEQ ID NO:3), and
an anti-PD-1 antibody (e.g., pembrolizumab or nivolumab).

In one example, the disclosure features a pharmaceutical composition including a compound of the first aspect of the disclosure and a pharmaceutically acceptable carrier. In a third aspect, the disclosure features a pharmaceutical composition comprising a compound of the first aspect and a pharmaceutically acceptable carrier.

In one example of the third aspect, the pharmaceutical composition further includes a protein including the amino acid sequence: MHQKRTAMFQ DPQERPRKLP QLCTELQTTI HDIILECVYC KQQLLRREVY DFAFRDLCIV YRDGNPYAVG DKCLKFYSKI SEYRHYCYSL YGTTLEQQYN KPLCDLLIRC INGQKPLCPE EKQRHLDKKQ RFHNGRGRWT GRCMSCCRSS RTRRETQL (SEQ ID NO:2), and a protein including the amino acid sequence:
MHGDTPTLHE YMLDLQPETT DLYGYGQLND SSEEEDEIDG PAGQAEPDRA HYNIVTFCCK CDSTLRLCVQ STHVDIRTLE DLLMGTLGIV CPICSQKP (SEQ ID NO:3).

In one example, the disclosure features a kit including (i) a compound of the first aspect of the disclosure or a composition of the second aspect of the disclosure and (ii) a protein including the amino acid sequence: MHQKRTAMFQ DPQERPRKLP QLCTELQTTI HDIILECVYC KQQLLRREVY DFAFRDLCIV YRDGNPYAVG DKCLKFYSKI SEYRHYCYSL YGTTLEQQYN KPLCDLLIRC INGQKPLCPE EKQRHLDKKQ RFHNGRGRWT GRCMSCCRSS RTRRETQL (SEQ ID NO:2),
and a protein including the amino acid sequence: MHGDTPTLHE YMLDLQPETT DLYGYGQLND SSEEEDEIDG PAGQAEPDRA HYNIVTFCCK CDSTLRLCVQ STHVDIRTLE DLLMGTLGIV CPICSQKP (SEQ ID NO:3). In a fourth aspect, the disclosure features a kit comprising (i) a compound of the first aspect, and (ii) a protein comprising the amino acid sequence: MHQKRTAMFQ DPQERPRKLP QLCTELQTTI HDIILECVYC KQQLLRREVY DFAFRDLCIV YRDGNPYAVG DKCLKFYSKI SEYRHYCYSL YGTTLEQQYN KPLCDLLIRC INGQKPLCPE EKQRHLDKKQ RFHNGRGRWT GRCMSCCRSS RTRRETQL (SEQ ID NO:2), and a protein comprising the amino acid sequence: MHGDTPTLHE YMLDLQPETT DLYGYGQLND SSEEEDEIDG PAGQAEPDRA HYNIVTFCCK CDSTLRLCVQ STHVDIRTLE DLLMGTLGIV CPICSQKP (SEQ ID NO:3).

Definitions

A “linker,” as used herein, refers to a monovalent or divalent group, in which one valency is covalently bonded to one biologically functional group, and the other valency is covalently bonded to another biologically functional group. In one example, a linker connects a nucleotide sequence of, e.g., a CpG oligonucleotide, to a lipid (e.g., -P(X)(OH)-O-CH(CH_2NHCO-(CH_2)$_{16}$-CH_3)$_2$, or a salt thereof, where X is O or

S, as described herein). Such linkers can optionally include one or more nucleotides, for example, a dinucleotide (e.g., GG).

A "pharmaceutically acceptable carrier," as used herein, refers to a vehicle capable of suspending or dissolving the active compound, and having the properties of being nontoxic and non-inflammatory in a patient. Moreover, a pharmaceutically acceptable carrier may include a pharmaceutically acceptable additive, such as a preservative, antioxidant, fragrance, emulsifier, dye, or excipient known or used in the field of drug formulation and that does not significantly interfere with the therapeutic effectiveness of the biological activity of the active agent, and that is non-toxic to the patient.

The terms "treat," "treatment," and "treating" refer to therapeutic approaches in which the goal is to reverse, alleviate, ameliorate, inhibit, slow down, or stop the progression or severity of a condition associated with a disease or disorder, e.g., cancer. These terms include reducing or alleviating at least one adverse effect or symptom of a condition, disease, or disorder. Treatment is generally "effective" if one or more symptoms or clinical markers are reduced, or if a desired response (e.g., a specific immune response) is induced. Alternatively, treatment is "effective" if the progression of a disease is reduced or halted.

The disclosure provides several advantages. For example, in including lipid moieties and, optionally, a linker, certain compounds of the disclosure bind to endogenous albumin in subjects to whom they are administered, which enhances delivery of the compounds to the lymph nodes of the subjects. This facilitates the induction of a therapeutic immune response against, for example, HPV proteins administered to the subject, leading to effective cancer treatment.

Other features and advantages of the disclosure will be apparent from the following detailed description, the drawings, and the claims.

DESCRIPTION OF THE DRAWINGS

Figure 1 is a graph showing the immune response against HPV16 E6.

Figure 2 is a graph showing the immune response against HPV16 E7.

Figure 3 is a graph showing tetramer stain analysis for HPV16 E7.

Figure 4 is a series of graphs showing that administration of HPV16 E6 and HPV16 E7 with an amphiphile-CpG (aCpG) decreased tumor size compared to aCpG alone or no treatment (No Tx).

Figure 5 is a graph showing superior HPV tetramer response with aCpG vaccine compared with soluble CpG vaccine at different time points.

Figure 6 is a graph showing sustained HPV tetramer responses over time with aCpG administered once weekly or once every two weeks.

Figure 7 is a graph showing tumor size response to E7 vaccine treatment.

Figure 8 is a graph showing improved survival in E7-vaccine treated C57BL6 mice implanted with TC-1 tumor cells.

Figure 9 is a series of graphs showing serum cytokine level changes after dosing.

Figure 10 is a graph showing tetramer responses over time for an HPV16 E7/aCpG vaccine.

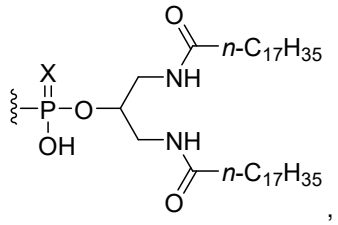
Figure 11 is a graph showing tumor growth response to HPV16 E7/aCpG vaccination plus or minus administration of an anti-PD-1 antibody.

Figure 12 is a graph showing the effects of HPV16 E7/aCpG vaccination plus or minus administration of an anti-PD-1 antibody on survival in TC-1 tumor bearing mice.

Figure 13 is a graph showing tetramer analysis for aCpG dose escalation.

CLAIMS

1. A compound consisting of the nucleotide sequence 5'-TCGTCGTTTTGTCGTTTTGTCGTT-3' (SEQ ID NO:1), at its 5' end, bonded or linked by a linker to the following lipid:



,

or a salt thereof,

wherein X is O or S.

2. The compound of claim 1, wherein the nucleotide sequence is bonded to the lipid.

3. The compound of claim 1 or 2, wherein all internucleoside groups connecting the nucleosides in 5'-TCGTCGTTTTGTCGTTTTGTCGTT-3' (SEQ ID NO:1) are phosphorothioates.

4. Use of the compound of claim 1 or 2 in the manufacture of a medicament for use in combinatorial therapy for treating a cancer in a human patient, wherein the medicament is to be administered in combination with a protein comprising the amino acid sequence:

MHQKRTAMFQ DPQERPRKLP QLCTELQTTI HDIILECVYC KQQLLRREVY DFAFRDLCIV YRDGNPYAVG DKCLKFYSKI SEYRHYCYSL YGTTLEQQYN KPLCDLLIRC INGQKPLCPE EKQRHLDKKQ RFHNGRGRWT GRCMSCCRSS RTRRETQL (SEQ ID NO:2),

and a protein comprising the amino acid sequence:

MHGDTPTLHE YMLDLQPETT DLYGYGQLND SSEEEDEIDG PAGQAEPDRA HYNIVTFCCK CDSTLRLCVQ STHVDIRTLE DLLMGTLGIV CPICSQKP (SEQ ID NO:3).

5. The use of claim 4, wherein the cancer is human papillomavirus (HPV) positive.

6. The use of claim 5, wherein the cancer is HPV type 16 positive.

7. The use of claim 4, wherein the cancer is a head or neck squamous cell carcinoma.

8. The use of claim 4, wherein the patient is receiving or has received platinum-containing chemotherapy.

9. The use of claim 4, wherein the compound and the proteins comprising the amino acid sequences of SEQ ID NO:2 and SEQ ID NO:3 are to be administered concurrently.

10. The use of claim 4, wherein the compound and the proteins comprising the amino acid sequences of SEQ ID NO:2 and SEQ ID NO:3 are to be administered sequentially.

11. A pharmaceutical composition comprising a compound of claim 1 or 2 and a pharmaceutically acceptable carrier.

12. The pharmaceutical composition of claim 11, wherein the composition further comprises a protein comprising the amino acid sequence:
MHQKRTAMFQ DPQERPRKLP QLCTELQTTI HDIILECVYC KQQLLRREVY DFAFRDLCIV YRDGNPYAVG DKCLKFYSKI SEYRHYCYSL YGTTLEQQYN KPLCDLLIRC INGQKPLCPE EKQRHLDKKQ RFHNGRGRWT GRCMSCCRSS RTRRETQL (SEQ ID NO:2),
and a protein comprising the amino acid sequence:
MHGDTPTLHE YMLDLQPETT DLYGYGQLND SSEEEDEIDG PAGQAEPDRA HYNIVTFCCK CDSTLRLCVQ STHVDIRTLE DLLMGTLGIV CPICSQKP (SEQ ID NO:3).

13. A kit comprising (i) a compound of claim 1 or 2, and (ii) a protein comprising the amino acid sequence:
MHQKRTAMFQ DPQERPRKLP QLCTELQTTI HDIILECVYC KQQLLRREVY DFAFRDLCIV YRDGNPYAVG DKCLKFYSKI SEYRHYCYSL YGTTLEQQYN KPLCDLLIRC INGQKPLCPE EKQRHLDKKQ RFHNGRGRWT GRCMSCCRSS RTRRETQL (SEQ ID NO:2),
and a protein comprising the amino acid sequence:
MHGDTPTLHE YMLDLQPETT DLYGYGQLND SSEEEDEIDG PAGQAEPDRA HYNIVTFCCK CDSTLRLCVQ STHVDIRTLE DLLMGTLGIV CPICSQKP (SEQ ID NO:3).