NUCLEIC ACID MOLECULE INHIBITING F11 GENE EXPRESSION

[0001] The present application claims priority to the following prior applications: Patent Application No. 202410114595.7, filed with the China National Intellectual Property Administration on January 26, 2024, entitled “Nucleic Acid Molecule Inhibiting F11 Gene Expression”; and Patent Application No. 202410568807.9, filed with the China National Intellectual Property Administration on May 9, 2024, entitled “GalNAc Derivative and Oligonucleotide Conjugate Thereof”. The two prior applications are incorporated herein by reference in their entirety.

TECHNICAL FIELD

[0002] The present application relates to the field of RNAi, specifically to a nucleic acid molecule that can inhibit the expression of the F11 gene by means of RNAi and uses thereof.

BACKGROUND

[0003] RNA interference (RNAi) is a highly conserved evolutionary phenomenon characterized by the efficient and specific degradation of homologous mRNA induced by double-stranded RNA (dsRNA). dsRNA, typically 19-30 bp in length, is one of the important tools in RNAi technology. In natural organisms, long dsRNAs are usually recognized by the Dicer enzyme after entering the cell and cleaved into small RNA fragments (i.e., siRNAs) about 21 to 23 nucleotides long. The resulting dsRNA fragments unwind into single strands and form complexes with certain proteins (RISCs for short). RISC can bind to mRNA that is complementary to the antisense strand of dsRNA in the cell and cleave the mRNA, causing it to be degraded, resulting in the inability to synthesize proteins and producing a gene “silencing” phenomenon. In industrial production, people tend to chemically synthesize dsRNA and modify it to further improve the stability and efficacy of dsRNA drugs.

[0004] A thrombus is a blood clot that restricts blood flow within a blood vessel. It can occur in the arterial or venous circulation and is the common pathological basis for most myocardial infarctions, ischemic strokes, and venous thromboembolism (VTE). Currently, the main targets of commonly used anticoagulants are thrombin and FX (or coagulation factor 10), which are equally important in hemostasis and thrombosis. If an attempt is made to exert an anticoagulant effect by inhibiting these two factors, it will inevitably affect the hemostasis process. This contradiction limits the intensity of anticoagulant therapy, meaning that patients at high risk of bleeding may not be suitable for anticoagulation treatment. Clinically, there is a need for new anticoagulants that can reduce the risk of bleeding while achieving

anticoagulation, and coagulation factor XI (FXI) has become a new anticoagulation target. Compared to FX, FXI (or coagulation factor FXI or F11) plays an auxiliary role in hemostasis but a necessary role in thrombosis. Spontaneous hemorrhage, central nervous system hemorrhage or gastrointestinal hemorrhage is rarely observed even in patients with severe FXI deficiency.

[0005] **SUMMARY**

[0006] The present application provides a nucleic acid molecule that inhibits the expression of cytokine XI (FXI) gene via RNAi, as well as compositions, formulations, and uses of the nucleic acid molecule. Sufficient data in the examples of the present application demonstrate that preferred nucleic acid molecules of the present application include, but are not limited to, dsRNA molecules numbered 3, 3', 5, 8, 9, 9', 14, 14', 15, 19, 24, 24', 26, 26', 50, 70, 70' and 82; shRNA molecules whose structure and sequence comprise the aforementioned dsRNA molecules; precursor molecules of the shRNA molecules or dsRNA molecules; and modified forms of the shRNA molecules or dsRNA molecules. Preferred modifications include, but are not limited to, modification E and modification E-1 described in detail in the examples herein.

[0007] Specifically, in one aspect, the present application relates to a nucleic acid molecule which comprises or consists of a sense sequence and an antisense sequence complementary to each other, and the antisense sequence comprises a polynucleotide sequence complementary to FXI gene mRNA, wherein

[0008] when the antisense sequence hybridizes with the FXI gene mRNA at the maximum complementarity, the base pairs formed by complementary pairing between the antisense sequence and the FXI gene mRNA sequence constitute complementary region 1;

[0009] when the antisense sequence hybridizes with the sense sequence at the maximum complementarity, the base pairs formed by complementary pairing between the antisense sequence and the sense sequence constitute complementary region 2;

[0010] the complementary region 1 and the complementary region 2 share at least 18, 19 or 20 identical base pairs,

[0011] the number of base pairs in the complementary region 1 is 15 to 35 bp, and the complementary region 1 comprises the following nucleotides in the FXI gene mRNA sequence:

[0012] nucleotides at positions 418 to 437, or 18 or 19 consecutive nucleotides therein,

[0013] nucleotides at positions 1141 to 1160, or 18 or 19 consecutive nucleotides therein,

[0014] nucleotides at positions 363 to 382, or 18 or 19 consecutive nucleotides therein,

[0015] nucleotides at positions 372 to 391, or 18 or 19 consecutive nucleotides therein,

[0016] nucleotides at positions 411 to 430, or 18 or 19 consecutive nucleotides therein,

[0017] nucleotides at positions 421 to 440, or 18 or 19 consecutive nucleotides therein,

[0018] nucleotides at positions 422 to 441, or 18 or 19 consecutive nucleotides therein,

[0019] nucleotides at positions 425 to 444, or 18 or 19 consecutive nucleotides therein,

[0020] nucleotides at positions 465 to 484, or 18 or 19 consecutive nucleotides therein,

[0021] nucleotides at positions 470 to 489, or 18 or 19 consecutive nucleotides therein,

[0022] nucleotides at positions 471 to 490, or 18 or 19 consecutive nucleotides therein, or

[0023] nucleotides at positions 518 to 537, or 18 or 19 consecutive nucleotides therein; and

[0024] the nucleotide positions of the FXI gene mRNA sequence are numbered in accordance with the corresponding nucleotides in the reference sequence of SEQ ID NO: 244.

[0025] "Consecutive 18 or 19 nucleotides therein" within a given nucleotide sequence refers to consecutive 19 nucleotides starting from the first or second nucleotide at the 5' end of the nucleotide sequence, including the first nucleotide or second nucleotide and retaining the original nucleotide order of the nucleotide sequence; or consecutive 18 nucleotides starting from the first, second or third nucleotide at the 5' end of the nucleotide sequence, including the first, second or third nucleotide and retaining the original nucleotide order of the nucleotide sequence.

[0026] In some embodiments, the sense sequence and the antisense sequence have the same length. In some embodiments, the sense sequence and/or the antisense sequence further comprise overhang nucleotides outside the complementary region 2. In some embodiments, the sense sequence is longer than the antisense sequence. In some embodiments, the antisense sequence is longer than the sense sequence. In some embodiments, the sense sequence is longer than the antisense sequence by 1, 2, 3 or 4 nucleotides. In some embodiments, the antisense sequence is longer than the sense sequence by 1, 2, 3 or 4 nucleotides. In some embodiments, the sense sequence further comprises 1 or 2 nucleotides on the 5' side and/or the 3' side of the complementary region 2. In some embodiments, the antisense sequence further comprises 1 or 2 nucleotides on the 5' side and/or the 3' side of the complementary region 2. In some embodiments, both the sense sequence and the antisense sequence further comprise 1 or 2 nucleotides on the 5' side and/or the 3' side of the complementary region 2. In some embodiments, when the antisense sequence hybridizes with the sense sequence at maximum complementarity, the sense sequence does not comprise any nucleotides that are not

complementary to the antisense sequence nucleotides between the first nucleotide and the last nucleotide of the complementary region 2. In some embodiments, when the antisense sequence hybridizes with the sense sequence at maximum complementarity, the antisense sequence does not comprise any nucleotides that are not complementary to the sense sequence nucleotides between the first nucleotide and the last nucleotide of the complementary region 2. In some embodiments, there are 2 overhang nucleotides located in the antisense sequence and adjacent to the 5' end of the complementary region 2, and the sense sequence comprises no overhang nucleotides. In some embodiments, the antisense sequence comprises only 0, 1 or 2 nucleotides outside the complementary region 1, and the sense sequence comprises only 0, 1 or 2 nucleotides outside the complementary region 2. In some embodiments, when the antisense sequence hybridizes with the mRNA sequence of the FXI gene at the maximum complementarity, the antisense sequence does not comprise nucleotides that fail to form complementary pairing with the mRNA sequence of the FXI gene between the first nucleotide and the last nucleotide of the complementary region 1. In some embodiments, when the antisense sequence hybridizes with the mRNA sequence of the FXI gene at the maximum complementarity, the mRNA sequence of the FXI gene does not comprise nucleotides that fail to form complementary pairing with the antisense sequence between the first nucleotide and the last nucleotide of the complementary region 1. In some embodiments, the 3' end of the complementary region 2 is an AU base pair. In some embodiments, the 3' end of the complementary region 2 is composed of A derived from the antisense sequence and U derived from the sense sequence (U may be written as T in the present application). In some embodiments, the 3' end of the complementary region 2 is composed of A derived from the sense sequence and U derived from the antisense sequence (U may be written as T in the present application). In some embodiments, the complementary region 1 and the complementary region 2 are uninterrupted, that is, continuous. In some embodiments, the complementary region 1 and/or complementary region 2 are discontinuous, and such discontinuity means being interrupted by one or more bubbles (a bubble refers to a non-complementary region formed by respective non-complementary nucleotides in the two sequences that constitute the complementary region).

[0027] In some embodiments, the complementary region 1 has 15 to 35 base pairs, for example 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34 or 35 bp. In some embodiments, the complementary region 2 has 15 to 35 base pairs (bp), for example 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34 or 35 bp. In some embodiments, the complementary region 1 and complementary region 2 have 18, 19, 20, 21,

22 or 23 identical base pairs. In some embodiments, the length of the sense sequence and/or the antisense sequence is 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29 or 30 nt. In some embodiments, the length of the sense sequence is 21 nt, and the length of the antisense sequence is 23 nt. In some embodiments, the nucleic acid molecule consists of a sense sequence of 21 nt in length and an antisense sequence of 23 nt in length. In some embodiments, the nucleic acid molecule consists of a sense sequence, an antisense sequence and a linker strand (also referred to as shRNA loop), wherein the linker strand connects the 3'-end nucleotide of the sense sequence and the 5'-end nucleotide of the antisense sequence. The selection of the linker strand is conventional in the art, for example, it may be selected in accordance with the method described in Jensen, Stig Mølgaard Rask et al. "Functional selection of shRNA loops from randomized retroviral libraries." PLoS one vol. 7,8 (2012): e43095. doi:10.1371/journal.pone.0043095, which is herein incorporated by reference in its entirety.

[0028] In some embodiments, the sense sequence and/or the antisense sequence further comprise 1 to 2 overhang nucleotides outside the complementary region 2.

[0029] In some embodiments, the antisense sequence comprises only 0, 1 or 2 nucleotides outside the complementary region 1, and the sense sequence comprises only 0, 1 or 2 nucleotides outside the complementary region 2.

[0030] In some embodiments, the 3' end of the complementary region 2 is an AU base pair. In further embodiments, the 5'-end A of the antisense strand is located outside the complementary region 1.

[0031] In some embodiments, the FXI gene mRNA sequence complementary to the antisense strand is transcribed from the coding region of a mammalian FXI gene. In some embodiments, the mammals is a primate. In some embodiments, the primate is a human or a cynomolgus monkey. In some embodiments, the mRNA comprises the polynucleotide sequence set forth in SEQ ID NO: 244. In some embodiments, the antisense sequence comprises the polynucleotide sequence set forth in SEQ ID NO: 122, 123, 126, 129, 130, 131, 136, 137, 138, 143, 149, 150, 152, 153, 177, 200, 201 or 213. In some embodiments, the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 122, 123, 126, 129, 130, 131, 136, 137, 138, 143, 149, 150, 152, 153, 177, 200, 201 or 213 plus additional 1, 2, 3 or 4 nucleotides. In some embodiments, the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 122, 123, 126, 129, 130, 131, 136, 137, 138, 143, 149, 150, 152, 153, 177, 200, 201 or 213 plus additional 1 to 2 nucleotides on the 5' and/or 3' side of the polynucleotide sequence. In some embodiments, the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 122, 123, 126, 129, 130, 131, 136, 137, 138,

143, 149, 150, 152, 153, 177, 200, 201 or 213. In some embodiments, the sense sequence comprises the polynucleotide sequence set forth in SEQ ID NO: 3, 4, 7, 10, 11, 12, 17, 18, 19, 24, 30, 31, 33, 34, 58, 81, 82 or 94. In some embodiments, the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 3, 4, 7, 10, 11, 12, 17, 18, 19, 24, 30, 31, 33, 34, 58, 81, 82 or 94 plus additional 1 to 4 (e.g., 2 or 3) nucleotides. In some embodiments, the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 3, 4, 7, 10, 11, 12, 17, 18, 19, 24, 30, 31, 33, 34, 58, 81, 82 or 94 plus additional 1 to 2 nucleotides on the 5' and/or 3' side of the polynucleotide sequence. In some embodiments, the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 3, 4, 7, 10, 11, 12, 17, 18, 19, 24, 30, 31, 33, 34, 58, 81, 82 or 94.

[0032] In some embodiments, the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 3, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 122; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 4, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 123; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 7, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 126; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 10, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 129; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 11, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 130; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 12, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 131; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 17, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 136; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 18, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 137; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 19, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 138; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 24, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 143; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 30, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 149; the sense sequence comprises or is the

polynucleotide sequence set forth in SEQ ID NO: 31, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 150; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 33, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 152; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 34, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 153; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 58, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 177; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 81, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 200; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 82, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 201; or the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 94, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 213.

[0033] In some embodiments, the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 3 plus additional 1, 2, 3 or 4 nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 122 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 4 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 123 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 7 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 126 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 10 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 129 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 11 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 130 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 12 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 131 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set

forth in SEQ ID NO: 17 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 136 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 18 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 137 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 19 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 138 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 24 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 143 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 30 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 149 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 31 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 150 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 33 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 152 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 34 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 153 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 58 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 177 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 81 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 200 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 82 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 201 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; or the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 94 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense

sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 213 plus additional 1 to 4 (e.g., 2 or 3) nucleotides.

[0034] In some embodiments, the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 3 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 122; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 4 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 123; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 7 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 126; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 10 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 129; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 11 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 130; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 12 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 131; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 17 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 136; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 18 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 137; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 19 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 138; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 24 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 143; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 30 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 149; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 31 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 150; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 33 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 152; the sense sequence consists of the

polynucleotide sequence set forth in SEQ ID NO: 34 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 153; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 58 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 177; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 81 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 200; the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 82 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 201; or the sense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 94 plus additional 1 to 4 (e.g., 2 or 3) nucleotides, and the antisense sequence is the polynucleotide sequence set forth in SEQ ID NO: 213.

[0035] In some embodiments, the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 3, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 122 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 4, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 123 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 7, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 126 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 10, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 129 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 11, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 130 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 12, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 131 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 17, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 136 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 18, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 137 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 19, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 138 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense

sequence is the polynucleotide sequence set forth in SEQ ID NO: 24, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 143 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 30, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 149 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 31, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 150 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 33, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 152 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 34, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 153 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 58, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 177 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 81, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 200 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 82, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 201 plus additional 1 to 4 (e.g., 2 or 3) nucleotides; or the sense sequence is the polynucleotide sequence set forth in SEQ ID NO: 94, and the antisense sequence consists of the polynucleotide sequence set forth in SEQ ID NO: 213 plus additional 1 to 4 (e.g., 2 or 3) nucleotides.

[0036] In some embodiments:

[0037] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 34, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 153;

[0038] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 58, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 177; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 33, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 152;

[0039] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 3, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 122;

[0040] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 4, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 123;

[0041] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 7, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 126;

[0042] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 10, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 129;

[0043] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 11, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 130;

[0044] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 12, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 131;

[0045] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 17, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 136;

[0046] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 18, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 137;

[0047] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 19, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 138;

[0048] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 24, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 143;

[0049] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 30, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 149;

[0050] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 31, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 150;

[0051] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 81, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 200;

[0052] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 82, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 201; or

[0053] the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 94, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 213.

[0054] In some embodiments, the nucleic acid molecule is a dsRNA or shRNA. In some embodiments, the dsRNA is an siRNA. In some embodiments, one or more nucleotides in the nucleic acid molecule are chemically modified. In some embodiments, the chemical modification makes the nucleic acid molecule more stable in the cellular or in vivo environment. In some embodiments, the chemical modification prolongs the in vivo and/or in vitro half-life of the nucleic acid molecule. In some embodiments, the chemical modification comprises, or consists of, one or more modifications selected from the group consisting of:

[0055] locked nucleic acid modification, unlocked nucleic acid modification, 2'-methoxyethyl modification, 2'-O-methyl modification (2'-OMe), 2'-O-allyl modification, 2'-C-alkyl modification, 2'-C-allyl modification, 2'-fluoro modification (2'-F), 2'-deoxy modification (d), phosphorothioate modification (s), 2'-amino modification, morpholino modification, phosphoramidate modification, methylphosphonate modification, tetrahydropyran modification, 1,5-anhydrohexitol modification, 5'-vinyl phosphate modification, and cyclohexenyl modification.

[0056] In some embodiments, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 12 of the sense strand comprise 2'-fluoro modifications, and/or the nucleotides at positions 2, 14 and 16 of the antisense strand comprise 2'-fluoro modifications. In some embodiments, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 12 of the sense strand comprise 2'-fluoro modifications, and/or the nucleotides at positions 2, 14 and 16 of the antisense strand comprise 2'-fluoro modifications, and the remaining nucleotides comprise 2'-O-methyl modifications. In some embodiments, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 12 of the sense strand are modified with 2'-fluoro, and/or the nucleotides at positions 2, 14 and 16 of the antisense strand are modified with 2'-fluor, the remaining nucleotides are modified with 2'-O-methyl, and the nucleic acid molecule comprises no additional modifications other than phosphorothioate modifications.

[0057] In some embodiments, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 11 of the sense strand comprise 2'-fluoro modifications, and/or the nucleotides at positions 2, 14 and 16 of the antisense strand comprise 2'-fluoro modifications. In some embodiments, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 11 of the sense strand comprise 2'-fluoro modifications, and/or the nucleotides at positions 2, 14 and 16 of the antisense strand comprise 2'-fluoro modifications, and the remaining nucleotides comprise 2'-O-methyl modifications. In some embodiments, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 11 of the sense strand are modified with 2'-fluoro, and/or the nucleotides at positions 2, 14 and 16 of the antisense strand are modified with 2'-fluor, the remaining nucleotides are modified with 2'-O-methyl, and the nucleic acid molecule comprises no additional modifications other than phosphorothioate modifications.

[0058] In some embodiments, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 11 of the sense strand comprise 2'-fluoro modifications, and/or the nucleotides at positions 2, 6, 14 and 16 of the antisense strand comprise 2'-fluoro modifications. In some embodiments, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 11 of the sense strand comprise 2'-fluoro modifications, and/or the nucleotides at positions 2, 6, 14 and 16 of the antisense strand comprise 2'-fluoro modifications, and the remaining nucleotides comprise 2'-O-methyl modifications. In some embodiments, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 11 of the sense strand are modified with 2'-fluoro, and/or the nucleotides at positions 2, 6, 14 and 16 of the antisense strand are modified with 2'-fluor, the remaining nucleotides are modified with 2'-O-methyl, and the nucleic acid molecule comprises no additional modifications other than phosphorothioate modifications.

[0059] In some embodiments, the nucleic acid molecule comprises a motif selected from the group consisting of:

[0060] (1) sense sequence: NmNmNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmNfNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmNmNm

[0061] (2) sense sequence: NmNmNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm, antisense sequence: NmNfNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmNmNm; and

[0062] (3) sense sequence: NmNmNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm, antisense sequence: NmNfNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmNmNm;

[0063] wherein in each sequence, N from 5' to 3' independently represents the nucleotide at the corresponding position in the sequence; Nm denotes a ribonucleotide modified with 2'-O-methyl, and Nf denotes a ribonucleotide modified with 2'-fluoro; it shall be understood that the present embodiment does not limit whether each nucleotide comprises any modifications other than those indicated herein.

[0064] In some embodiments, the above embodiments further comprise at least one phosphorothioate modification between the first and second nucleotides, and between the second and third nucleotides at the 5' end of the sense sequence; and comprise at least one phosphorothioate modification between the first and second nucleotides, between the second and third nucleotides, between the last and penultimate nucleotides, and between the penultimate and antepenultimate nucleotides at the 5' end of the antisense sequence.

[0065] In some embodiments, the nucleic acid molecule comprises a motif selected from any one of (1) to (9) below:

[0066] (1) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0067] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than 2'-F, 2'-OMe and phosphorothioate modifications;

[0068] (2) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0069] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than 2'-F, 2'-OMe and phosphorothioate modifications;

[0070] (3) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0071] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than 2'-F, 2'-OMe and phosphorothioate modifications;

[0072] (4) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmsNmsNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0073] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmsNmsNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than 2'-F, 2'-OMe and phosphorothioate modifications;

[0074] (5) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmsNmsNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0075] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmsNmsNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than 2'-F, 2'-OMe and phosphorothioate modifications;

[0076] (6) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmsNmsNm, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0077] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmsNmsNm, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than 2'-F, 2'-OMe and phosphorothioate modifications;

[0078] (7) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0079] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than 2'-F, 2'-OMe and phosphorothioate modifications;

[0080] (8) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0081] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than 2'-F, 2'-OMe and phosphorothioate modifications;

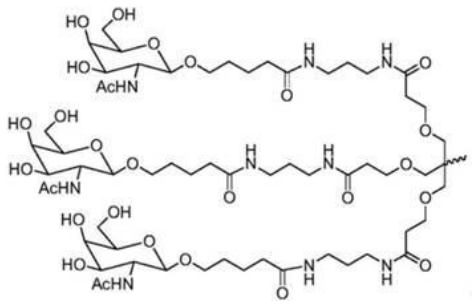
[0082] (9) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0083] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm,

wherein each nucleotide comprises no modifications other than 2'-F, 2'-OMe and phosphorothioate modifications.

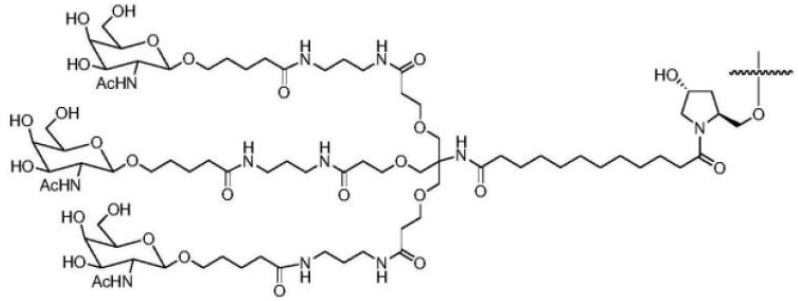
[0084] In all motifs of the present application, Nm represents a ribonucleotide modified with 2'-O-methyl, Nf represents a ribonucleotide modified with 2'-fluoro, and s represents a phosphorothioate modification.

[0085] In some embodiments, the nucleic acid molecule is linked to an organ-targeting ligand, such as a liver-targeting ligand. In some embodiments, the nucleic acid molecule is linked to at least one asialoglycoprotein receptor (ASGPR) ligand. In some embodiments, the organ-targeting ligand is linked to the 5' end or 3' end of the sense sequence. In some embodiments, the ASGPR ligand is one or more GalNAc derivatives linked via a divalent or trivalent branched structure. In some embodiments, the GalNAc derivative comprises the following structure:



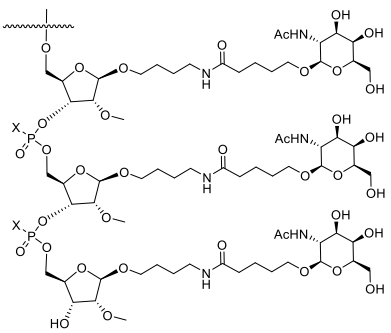
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[0086] In some embodiments, the GalNAc derivative is L96, and the structure of the L96 is shown below:



.

[0087] In some embodiments, the GalNAc derivative is ligand 1, and the structure of the ligand 1 is shown below:

[0088]  (X=S^- or O^-) (Formula II);

[0089] wherein 〰 in formula I and formula II represents linkage to the 3' end of the sense sequence or antisense sequence of the nucleic acid molecule; preferably, the linkage is formed via a phosphodiester bond or a phosphorothioate diester bond.

[0090] More preferably, the nucleic acid molecule has a modification motif according to any one of (1) to (6):

[0091] (1) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96; antisense strand: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0092] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96; antisense strand: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm; wherein each nucleotide comprises no modifications other than L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0093] (2) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1; antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0094] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm; wherein each nucleotide comprises no modifications other than ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0095] (3) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNm-L96; antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0096] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNm-L96; antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

wherein each nucleotide comprises no modifications other than L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0097] (4) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1; antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0098] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;
wherein each nucleotide comprises no modifications other than ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0099] (5) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNm-L96; antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[00100] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNm-L96; antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;
wherein each nucleotide comprises no modifications other than L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[00101] (6) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1; antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[00102] in other embodiments: sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;
wherein each nucleotide comprises no modifications other than ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications.

[0100] In some embodiments, the nucleic acid molecule comprises a 5'-phosphate or 5' phosphate mimetic modification, such as a phosphate or phosphate mimetic at the 5' end of the antisense strand; preferably, the 5' phosphate mimetic modification is 5'-VP.

[0101] In some embodiments, the nucleic acid molecule has the following modification motifs:

[0102] GN-E20VP:

[0103] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1

[0104] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0105] in other embodiments, sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1

[0106] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications.

[0107] In some embodiments, the nucleic acid molecule has the following modification motifs:

[0108] GN-E04VP:

[0109] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1

[0110] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0111] in other embodiments, sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1

[0112] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications.

[0113] In some embodiments, the nucleic acid molecule has the following modification motifs:

[0114] GN-E05VP:

[0115] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1

[0116] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0117] in other embodiments, sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1

[0118] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications.

[0119] In some embodiments, the nucleic acid molecule has the following modification motifs:

[0120] LN-E20VP:

[0121] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNm-L96

[0122] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0123] in other embodiments, sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNm-L96

[0124] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications.

[0125] In some embodiments, the nucleic acid molecule has the following modification motifs:

[0126] LN-E04VP:

[0127] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96

[0128] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0129] in other embodiments, sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96

[0130] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications.

[0131] In some embodiments, the nucleic acid molecule has the following modification motifs:

[0132] LN-E05VP:

[0133] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96

[0134] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0135] in other embodiments, sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96

[0136] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications.

[0137] In some embodiments, the naked sequences of the nucleic acid molecules specifically provided herein include the sense sequence: SEQ ID NO: 58 and the antisense sequence: SEQ ID NO: 177 (siRNA No. 50), which adopt any one of the modification patterns listed above.

[0138] In some embodiments, the naked sequences of the nucleic acid molecules specifically provided herein include the sense sequence: SEQ ID NO: 58 and the antisense sequence: SEQ ID NO: 177 (siRNA No. 50), which adopt the following modification patterns:

[0139] GN-E20:

[0140] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0141] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0142] GN-E20VP:

[0143] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0144] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0145] GN-E04:

[0146] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0147] antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0148] GN-E04VP:

[0149] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0150] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0151] GN-E05:

[0152] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0153] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm; or

[0154] GN-E05VP:

[0155] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0156] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm.

[0157] In some embodiments, the naked sequences of the nucleic acid molecules specifically provided herein include the sense sequence: SEQ ID NO: 58 and the antisense sequence: SEQ ID NO: 177 (siRNA No. 50), which adopt the following modification patterns:

[0158] GN-E20:

[0159] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0160] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm; wherein each nucleotide comprises no modifications other than ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0161] GN-E20VP:

[0162] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0163] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0164] GN-E04:

[0165] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0166] antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0167] GN-E04VP:

[0168] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0169] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0170] GN-E05:

[0171] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0172] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications; or

[0173] GN-E05VP:

[0174] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0175] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications.

[0176] In some embodiments, the naked sequences of the nucleic acid molecules provided herein include the sense sequence: SEQ ID NO: 34 and the antisense sequence: SEQ ID NO: 153 (siRNA No. 26'), which adopt any one of the modification patterns listed above.

[0177] In some embodiments, the naked sequences of the nucleic acid molecules provided herein include the sense sequence: SEQ ID NO: 34 and the antisense sequence: SEQ ID NO: 153 (siRNA No. 26'), which adopt the modification patterns shown below:

[0178] GN-E20:

[0179] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0180] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0181] GN-E20VP:

[0182] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0183] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0184] GN-E04:

[0185] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0186] antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0187] GN-E04VP:

[0188] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0189] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0190] GN-E05:

[0191] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0192] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm; or

[0193] GN-E05VP:

[0194] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0195] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm.

[0196] In some embodiments, the naked sequences of the nucleic acid molecules specifically provided herein include the sense sequence: SEQ ID NO: 34 and the antisense sequence: SEQ ID NO: 153 (siRNA No. 26'), which adopt the following modification patterns:

[0197] GN-E20:

[0198] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0199] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

wherein each nucleotide comprises no modifications other than ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0200] GN-E20VP:

[0201] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0202] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0203] GN-E04:

[0204] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0205] antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0206] GN-E04VP:

[0207] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0208] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications;

[0209] GN-E05:

[0210] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1,

[0211] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications; or

[0212] GN-E05VP:

[0213] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1,

[0214] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, ligand-1, 2'-F, 2'-OMe and phosphorothioate modifications.

[0215] In some embodiments, the naked sequences of the nucleic acid molecules specifically provided herein include the sense sequence: SEQ ID NO: 58 and the antisense sequence: SEQ ID NO: 177 (siRNA No. 50), which adopt the following modification patterns:

[0216] LN-E20:

[0217] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0218] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0219] LN-E20VP:

[0220] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0221] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0222] LN-E04:

[0223] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0224] antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0225] LN-E04VP:

[0226] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0227] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0228] LN-E05:

[0229] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0230] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm; or

[0231] LN-E05VP:

[0232] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96,

[0233] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm.

[0234] In some embodiments, the naked sequences of the nucleic acid molecules specifically provided herein include the sense sequence: SEQ ID NO: 58 and the antisense sequence: SEQ ID NO: 177 (siRNA No. 50), which adopt the following modification patterns:

[0235] LN-E20:

[0236] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0237] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0238] LN-E20VP:

[0239] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0240] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0241] LN-E04:

[0242] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0243] antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0244] LN-E04VP:

[0245] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0246] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0247] LN-E05:

[0248] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96,

[0249] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications; or

[0250] LN-E05VP:

[0251] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96,

[0252] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications.

[0253] In some embodiments, the naked sequences of the nucleic acid molecules provided herein include the sense sequence: SEQ ID NO: 34 and the antisense sequence: SEQ ID NO: 153 (siRNA No. 26'), which adopt the modification patterns shown below:

[0254] LN-E20:

[0255] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0256] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0257] LN-E20VP:

[0258] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0259] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0260] LN-E04:

[0261] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96,

[0262] antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0263] LN-E04VP:

[0264] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNm-L96,

[0265] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0266] LN-E05:

[0267] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNm-L96,

[0268] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm; or

[0269] LN-E05VP:

[0270] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNm-L96,

[0271] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm.

[0272] In some embodiments, the naked sequences of the nucleic acid molecules specifically provided herein include the sense sequence: SEQ ID NO: 34 and the antisense sequence: SEQ ID NO: 153 (siRNA No. 26'), which adopt the following modification patterns:

[0273] LN-E20:

[0274] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0275] antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0276] LN-E20VP:

[0277] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96,

[0278] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0279] LN-E04:

[0280] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96,

[0281] antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0282] LN-E04VP:

[0283] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96,

[0284] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications;

[0285] LN-E05:

[0286] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96,

[0287] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications; or

[0288] LN-E05VP:

[0289] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96,

[0290] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm, wherein each nucleotide comprises no modifications other than vp, L96, 2'-F, 2'-OMe and phosphorothioate modifications.

[0291] In addition, the present application further provides a precursor of the aforementioned nucleic acid molecule, which may be an shRNA or dsRNA.

[0292] In a second aspect, the present application provides a second nucleic acid molecule. The second nucleic acid molecule can be transcribed into a dsRNA or shRNA precursor in cells, and the dsRNA or shRNA is the nucleic acid molecule according to the first aspect. In some embodiments, the second nucleic acid molecule is a circular or linear nucleic

acid molecule. In some embodiments, the second nucleic acid molecule is a circular or linear plasmid. In some embodiments, the nucleic acid molecule is derived from an artificially constructed viral genome, including but not limited to lentiviral vectors or other retroviral vectors, adenoviral vectors, AAV vectors, poxviral vectors, baculoviral vectors and herpes simplex viral vectors. In some embodiments, the nucleic acid molecule is derived from a cellular genome, such as a nuclear genome, mitochondrial nucleic acid or cytoplasmic free nucleic acid.

[0293] In a third aspect, the present application further provides a nucleic acid delivery vehicle, which comprises the nucleic acid molecule according to the first aspect or the second nucleic acid molecule according to the second aspect. In some embodiments, the nucleic acid delivery vehicle is a liposome, lipid nanoparticle, other polymer, endosome, exosome or vesicle.

[0294] Also provided are viral particles comprising the second nucleic acid molecule according to the second aspect. In some embodiments, the viral particles are enveloped virus particles or togavirus particles. In some embodiments, the viral particles are pseudotyped viral particles. In some embodiments, the viral particles are AAV, baculovirus, poxvirus, herpesvirus, alphavirus, lentivirus or other retrovirus particles.

[0295] Also provided are cells comprising the second nucleic acid molecule according to the second aspect. In some embodiments, the cells are prokaryotic cells. In some embodiments, the cells are eukaryotic cells, such as stem cells, including hematopoietic stem cells, mesenchymal stem cells and the like.

[0296] The present application further provides a pharmaceutical composition, which comprises the nucleic acid molecule according to the first aspect or a salt thereof, and a pharmaceutically acceptable carrier or diluent. In some embodiments, the pharmaceutical composition is used for treating or preventing thromboembolic complications or coagulation disorders in a subject. Preferably, the thromboembolic complications are one or more selected from the group consisting of deep vein thrombosis, pulmonary embolism, myocardial infarction and stroke.

[0297] In a fourth aspect, the present application further provides the use of the nucleic acid according to the first aspect, the second nucleic acid according to the second aspect, the nucleic acid delivery vehicle according to the third aspect, as well as the aforementioned viral particles, the aforementioned cells and the aforementioned pharmaceutical compositions.

[0298] For example, the present application provides the use of the nucleic acid molecule according to the first aspect or a salt thereof, the second nucleic acid molecule according to the second aspect, the aforementioned nucleic acid delivery vehicle, the

aforementioned viral particles or the aforementioned cells in the manufacture of a drug for preventing or treating thromboembolic complications or coagulation disorders in a subject. In some embodiments, the thromboembolic complications are preferably one or more selected from the group consisting of deep vein thrombosis, pulmonary embolism, myocardial infarction and stroke.

[0299] The present application further provides a method for treating or preventing thromboembolic complications or coagulation disorders in a subject, wherein the method comprises administering an effective amount of the nucleic acid molecule according to the first aspect or a salt thereof, the second nucleic acid molecule according to the second aspect, the aforementioned nucleic acid delivery vehicle, the aforementioned viral particles, the aforementioned cells or the aforementioned pharmaceutical composition to a subject in need thereof. In some embodiments, the thromboembolic complications are preferably one or more selected from the group consisting of deep vein thrombosis, pulmonary embolism, myocardial infarction and stroke.

[0300] Also provided is a compound for treating or preventing thromboembolic complications or coagulation disorders in a subject, which comprises the nucleic acid molecule according to the first aspect or a salt thereof, the second nucleic acid molecule according to the second aspect, the aforementioned nucleic acid delivery vehicle, the aforementioned viral particles, the aforementioned cells or the aforementioned pharmaceutical composition. In some embodiments, the thromboembolic complications are preferably one or more selected from the group consisting of deep vein thrombosis, pulmonary embolism, myocardial infarction and stroke.

[0301] The present application further provides the use of the nucleic acid molecule according to the first aspect or a salt thereof or the second nucleic acid molecule according to the second aspect for inhibiting the expression of factor 11.

[0302] The present application further provides the use of the nucleic acid molecule according to the first aspect or a salt thereof or the second nucleic acid molecule according to the second aspect in the manufacture of a drug for inhibiting FXI expression in a subject.

[0303] The present application further provides a compound for inhibiting FXI expression in a subject, wherein the compound comprises the nucleic acid molecule according to the first aspect or a salt thereof, as well as a pharmaceutically acceptable carrier or diluent. The preferred embodiments of the present application have been described in detail above, but the present application is not limited thereto. Various simple modifications may be made to the technical solutions of the present application within the scope of its technical concept,

including combining individual technical features in any other suitable manner. All such simple modifications and combinations shall be deemed part of the disclosure of the present application and fall within its protection scope. The aspects and embodiments described herein include those embodied by the terms “comprising”, “consisting of” and “consisting essentially of”.

BRIEF DESCRIPTION OF THE DRAWINGS

[0304] FIG. 1 shows the high-throughput screening results of naked dsRNA nucleic acid molecules (5 nM) in HepG2 cells.

[0305] FIG. 2 shows the relative levels of FXI mRNA in HepG2 cells upon administration of naked nucleic acid molecules of candidate dsRNA or positive control dsRNA at concentrations of 5 nM and 1 nM, respectively.

[0306] FIG. 3 shows the inhibition rate of FXI mRNA in HepG2 cells by dsRNA modified with modification E at a concentration of 1 nM.

[0307] FIG. 4 shows the inhibition rates of FXI mRNA by GalNAc-conjugated dsRNA at different concentrations.

[0308] FIG. 5 shows the in vivo inhibition results of target protein after administration of different GalNAc-conjugated dsRNAs.

[0309] FIG. 6 shows the comparison results of different GalNAc after in vivo administration.

[0310] FIG. 7 shows the comparison of in vitro cellular IC50 values of dsRNAs with different modifications.

[0311] FIG. 8 shows the comparison results of dsRNAs with different modification motifs after in vivo administration.

[0312] **DETAILED DESCRIPTION**

[0313] The present invention provides a nucleic acid molecule capable of triggering cleavage of RNA transcripts of the coagulation factor FXI (or FX11) gene mediated by the RNA-induced silencing complex (RISC), as well as a second nucleic acid molecule capable of transcribing the nucleic acid molecule. Also provided are delivery vehicles for the nucleic acid molecule and the second nucleic acid molecule, viruses or cells capable of transcribing the nucleic acid molecule, and uses of the nucleic acid molecule and the second nucleic acid molecule.

[0314] Terms

[0315] For the purposes of this specification, the following definitions will apply, and where appropriate, terms used in the singular form will also include the plural form, and vice

versa. Unless otherwise defined, all technical and scientific terms used herein shall have the same meanings as commonly understood by those skilled in the art to which the present invention pertains. All technical and patent publications cited herein are incorporated by reference in their entirety.

[0316] As used herein, "dsRNA" refers to double-stranded RNA. Since siRNA is double-stranded RNA, the term "dsRNA" encompasses siRNA. dsRNA also includes double-stranded RNA longer than siRNA. A dsRNA longer than siRNA means that its sense strand, antisense strand, or both sense and antisense strands are longer than those of siRNA. Generally, after entering cells, double-stranded RNAs longer than the siRNA sequence they contain are cleaved into siRNA by Dicer, a type III endonuclease. In some embodiments, each strand of the dsRNA independently has a length of 15 to 30 nt (as used herein, "nt" stands for nucleotide). After "siRNA" is incorporated into the RNA-induced silencing complex (RISC), one or more helicases within RISC unwind the siRNA duplex. Upon binding to target mRNA complementary to the antisense strand of the siRNA, one or more endonucleases in RISC cleave the target, thereby inducing gene silencing. Typically, most nucleotides of each strand of a dsRNA molecule are ribonucleotides. However, one or both strands may contain one or more non-ribonucleotides, such as deoxyribonucleotides and/or non-natural nucleotides. In some embodiments, the dsRNA molecule comprises no non-natural nucleotides. In some embodiments, every nucleotide of the dsRNA is a ribonucleotide. As used herein, a dsRNA may or may not comprise one or more chemically modified nucleotides.

[0317] The terms "FXI", "coagulation factor FXI" and "factor 11" are used interchangeably, and are also referred to as FXI or PTA in the art. FXI can be an FXI derived from mammals. In some embodiments, the FXI is a primate-derived FXI. In some embodiments, the FXI is a human-derived FXI. In some embodiments, FXI is an FXI derived from cynomolgus monkeys. As used herein, the term "FXI gene mRNA" refers to mRNA that encodes factor 11 protein. It is transcribed from the DNA of the FXI gene, and may be mature mRNA or mRNA precursor (pre-mRNA), and therefore may contain or be free of introns. Since minor nucleotide mutations may exist in the factor 11 gene among different individuals, unless otherwise specified, the factor 11 gene mRNA sequences described herein cover all mRNA sequences transcribed from mutants of the factor 11 gene. The human FXI gene mRNA sequence can be found, for example, in the gene bank accession number GI: 1732746318 (NM_000128.4). The rhesus monkey FXI gene mRNA sequence can be found, for example, in the gene bank accession number GI: 1622942384 (XM_015139652.2). The mRNA sequence of the cynomolgus monkey FXI gene can be found, for example, in the Genebank accession

number GI: 2161917139 (XM_005556483.3). The mouse FXI gene mRNA sequence can be found, for example, in the Genebank accession number GI: 2293430447 (NM_028066.3). The rat FXI gene mRNA sequence can be found, for example, in the Genebank accession number GI: 2293332621 (NM_001411666.1). Additional FXI gene mRNA sequences can be readily retrieved from publicly accessible databases such as GenBANK. Unless otherwise specified, SEQ ID NO: 244 (i.e., NM_000128.4) is adopted as the reference sequence for FXI gene mRNA. That is, the numbering of each nucleotide position in the FXI gene mRNA corresponds to the nucleotide numbering in the 5' to 3' direction of the reference sequence SEQ ID NO: 244. As used herein, the term “reference sequence” means a standard sequence for homologous sequence alignment, which is used to define nucleotide positions within homologous polynucleotides or polynucleotide sequences. For example, the phrase “the base position numbering of the FXI gene mRNA sequence follows that of the corresponding bases in the reference sequence SEQ ID NO: 244” means that gaps are inserted or nucleotides are deleted in the FXI gene mRNA sequence to maximize base matches between the FXI gene mRNA sequence and the reference sequence. The nucleotides of the reference sequence are numbered consecutively starting from the first nucleotide at the 5' end. Nucleotides in the FXI gene mRNA sequence that align with those in the reference sequence are assigned identical position numbers.

[0318] As used herein, the term “complementary” in reference to nucleic acids means that one nucleic acid is capable of forming hydrogen bonds with another nucleic acid via traditional Watson-Crick base pairing. Percent complementarity refers to the percentage of nucleotides in the shorter of two nucleic acid molecules that can form hydrogen bonds (i.e., Watson-Crick base pairs) with the other nucleic acid molecule, relative to the total nucleotides of the shorter nucleic acid molecule. For example, approximately 5, 6, 7, 8, 9 and 10 out of 10 nucleotides correspond to about 50%, 60%, 70%, 80%, 90% and 100% complementarity, respectively. “Fully complementary” means that all contiguous residues of a nucleic acid sequence form hydrogen bonds with an equal number of contiguous residues in a second nucleic acid sequence. As used herein, “substantially complementary” means a complementarity level of at least any one of 70%, 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99% or 100% over a region of about 40, 50, 60, 70, 80, 100, 150, 200, 250 or more nucleotides, or refers to two nucleic acids that hybridize under stringent conditions. For a single base or a single nucleotide, a base pair formed between A and T or U, and between C and G or I in accordance with Watson-Crick base pairing rules is defined as complementary, paired or matched, and vice versa. All other base pairings are deemed non-complementary.

[0319] As used herein, “hybridization” of nucleic acids refers to a reaction in which one or more polynucleotides react to form a complex stabilized by hydrogen bonds between nucleotide residues. Hydrogen bonding can occur through Watson-Crick base pairing, Hoogsteen binding, or any other sequence-specific mechanism. Such complexes may include two strands forming a duplex structure, three or more strands forming a multi-strand complex, a single self-hybridizing strand, or a combination thereof.

[0320] In the present application, the phrase that two polynucleotide sequences or two nucleic acid strands “hybridize at the maximum complementarity” means a hybridization pattern in which the maximum number of nucleotides of the two strands form pairs via hydrogen bonds. “Hybridization at the maximum complementarity” may permit one or more mismatches, as well as one or more bulges in one or both strands. However, in some embodiments, the two strands that “hybridize at the maximum complementarity” contain no mismatches or bulges after hybridization. As used herein, the term “complementary region” refers to all base pairs formed by hydrogen bonding, extending from the first hydrogen-bonded base pair at the 5' end to the last hydrogen-bonded base pair after the two strands hybridize. A complementary region formed by two hybridized strands may be continuous or discontinuous. “Complementary region 1” as used herein specifically refers to the complementary region composed of base pairs formed by complementary base pairing between the antisense sequence and the FXI gene mRNA when the antisense sequence hybridizes with the FXI gene mRNA at the maximum complementarity. “Complementary region 2” refers to the complementary region composed of base pairs formed by complementary base pairing between the antisense sequence and the sense sequence when the antisense sequence hybridizes with the sense sequence at the maximum complementarity. Since both complementary region 1 and complementary region 2 involve the antisense sequence, the antisense sequence portions of the complementary region 1 and complementary region 2 generally share partial or complete nucleotide composition identity. In the present application, the designations of antisense sequence and sense sequence are defined relative to a third sequence that is complementary to one of the two sequences. For example, for “a nucleic acid molecule that inhibits the expression of factor 11 (FXI) gene in cells via RNAi”, its antisense sequence and sense sequence are defined with reference to the third sequence, i.e., the FXI gene mRNA sequence. Specifically, the antisense sequence refers to the sequence of the nucleic acid molecule that forms a complementary region with the FXI gene mRNA sequence; the sense sequence refers to the sequence of the nucleic acid molecule that has at least 10 consecutive identical nucleotides relative to the FXI gene mRNA sequence. In addition, the 5' end of any complementary region herein refers to the position of the

nucleotide or base pair within the complementary region that is closest to the 5' end of the sense strand or the aforesaid third sequence. The 5' end of the complementary region refers to the side or extremity of the complementary region that is relatively proximal to the 5' end of the sense strand or the aforesaid third sequence. Similarly, the 3' end of any complementary region herein refers to the position of the nucleotide or base pair within the complementary region that is closest to the 3' end of the sense strand or the aforesaid third sequence. The 3' end of the complementary region refers to the side or extremity of the complementary region that is relatively proximal to the 3' end of the sense strand or the aforesaid third sequence. In the present application, when describing the terms “proximal”, “close” or “distant” with respect to nucleotide positions on the same sequence or nucleic acid strand, such terms refer to a smaller or larger number of intervening nucleotides between the two positions.

[0321] As used herein, the term “overhang nucleotide” is referred with respect to the complementary region 2, and refers to nucleotides located outside the complementary region 2 in the nucleotides of the sense sequence and/or the antisense sequence after the sense sequence and the antisense sequence are hybridized at maximum complementarity. In some embodiments, the overhang nucleotides are present only on the sense sequence. In some embodiments, the overhang nucleotides are present only on the antisense sequence. In some embodiments, the overhang nucleotides are present on both the sense sequence and the antisense sequence. In some embodiments, the overhang nucleotides are located only on the 5' side of the complementary region 2. In some embodiments, the overhang nucleotides are located only on the 5' side of the complementary region 2 of the sense strand. In some embodiments, the overhang nucleotides are located only on the 5' side of the complementary region 2 of the antisense strand. In some embodiments, the overhang nucleotides are located only on the 3' side of the complementary region 2. In some embodiments, the overhang nucleotides are located only on the 3' side of the complementary region 2 of the sense strand. In some embodiments, the overhang nucleotides are located only on the 3' side of the complementary region 2 of the antisense strand. In some embodiments, the number of overhang nucleotides on the same side (i.e., the 5' end or the 3' end) of the sense sequence or the antisense sequence does not exceed two (namely 1 or 2). In the present application, the expressions “5' side” and “3' side” are used to describe the relative positional relationship between two sequence segments, two nucleotides, or one nucleotide and one sequence segment in the same polynucleotide sequence. The “5' end” refers to the end of the polynucleotide sequence bearing a free 5' phosphate group or a free 5' hydroxyl group. The “3' end” refers to the end of the polynucleotide sequence bearing a free 3' hydroxyl group or a free 3' phosphate group. A

sequence or nucleotide on the 3' side of a given sequence in a nucleic acid strand is closer to the 3' end of the entire nucleic acid strand than the given sequence. For example, the phrase "located on the 5' side of the complementary region 2" with respect to a certain sequence or one or more nucleotides means that "such sequence or nucleotide(s)" is/are closer to the 5' end of the polynucleotide sequence (e.g., the antisense sequence or the sense sequence) where they resides, relative to "the complementary region 2".

[0322] The term "nucleotide", in addition to referring to naturally occurring ribonucleotide or deoxyribonucleotide monomers, shall also be construed herein as including related structural variants thereof, such as derivatives and analogs that are functionally equivalent in the specific context of use, unless the context clearly dictates otherwise. For example, "nucleotide" refers to deoxyribonucleotide or ribonucleotide. A nucleotide may be a canonical nucleotide (i.e., adenosine, guanosine, cytidine, thymidine and uridine), a nucleotide isomer or a nucleotide analog. A nucleotide analog refers to a nucleotide with a modified purine or pyrimidine base or a modified ribose moiety. Nucleotide analogs can be naturally occurring nucleotides (e.g., inosine, pseudouridine, etc.) or non-naturally occurring nucleotides. Non-limiting examples of modifications on the sugar or base moiety of a nucleotide include the addition (or removal) of acetyl, amino, carboxyl, carboxymethyl, hydroxyl, methyl, phosphoryl and thiol groups, as well as the substitution of carbon and nitrogen atoms in bases with other atoms (e.g., 7-deazapurine). Nucleotide analogs further include dideoxynucleotides, 2'-O-methyl nucleotides, locked nucleic acids (LNA), peptide nucleic acids (PNA) and morpholino oligonucleotides (morpholino). In some embodiments, the "nucleotide" as used herein excludes non-naturally occurring nucleotides with modified bases. In some embodiments, the "nucleotide" as used herein excludes any nucleotides with modified bases. As used herein, "G", "C", "A", "T" and "U" generally represent nucleotides with guanine, cytosine, adenine, thymine and uracil as their respective bases. Unless otherwise specified, "G", "C", "A", "T" and "U" indicate nucleotides without limitation on the modifications they contain, that is, they can be used to represent natural nucleotides or non-natural nucleotides. Such non-naturally occurring nucleotides may have modified ribose and/or modified bases, provided that the bases are still capable of forming complementary hydrogen bonds with their natural pairing counterparts in accordance with Watson-Crick base pairing rules. However, in the context of RNA and in RNA sequences, unless otherwise specified, "T" refers to uridine or uracil. It shall be understood that in any context relating to nucleotide sequences herein, "nucleotide", "nucleotide residue" and "base" are used interchangeably. The number of base pairs is

expressed in bp, where one bp equals one base pair. The number of nucleotides is expressed in nt, where one nt equals one nucleotide.

[0323] As used herein, the term “internucleotide linkage” refers to a chemical bond (or linking group) between two adjacent nucleotides, or between a nucleotide and a ligand. Unless otherwise specified, such chemical bonds are phosphoester bonds or phosphodiester bonds. Where the letter “s” is marked in a modification motif to indicate a phosphorothioate modification, only the internucleotide linkage at the position labeled “s” shall be a phosphorothioate bond; all other unmarked internucleotide linkages shall be phosphoester bonds. Examples are as follows:

[0324] (1) sense strand: NmNmNmNmNmNmNfNmNfNdNfNmNmNmNmNmNmNmNmNmNm, antisense strand: NmNfNmNmNdNmNdNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmNmNm;

[0325] all internucleotide linkages in (1) may be either phosphoester bonds or phosphorothioate bonds;

[0326] however, if it is as follows: sense chain: CmsAmsGmAmGmUmUfAmUfCdGfAmGmGmCmAmCmAmUmUmAms-ligand 1,

[0327] antisense strand: vp-UmsAfsAmUmGdUmGdCmCmUmCmGmAmUfAmAfCmUmCmUmGmsGmsCm, since “s” is specifically marked here, except that the internucleotide linkages at the positions specified below are phosphorothioate bonds, and all other internucleotide linkages are phosphoester bonds: the first and second internucleotide linkages starting from the 5' end of the sense strand; the internucleotide linkage between the 3' end nucleotide of the sense strand and ligand 1; the first and second internucleotide linkages starting from the 5' end of the antisense strand; and the first and second internucleotide linkages starting from the 3' end of the antisense strand. The first internucleotide linkage means the chemical bond between the first and second nucleotides starting from an end (e.g., the 5' end or 3' end). The second internucleotide linkage means the chemical bond between the second and third nucleotides starting from an end (e.g., the 5' end or 3' end), and so on. The nth internucleotide linkage means the chemical bond between the nth nucleotide and the (n+1)th nucleotide or ligand starting from an end (e.g., the 5' end or 3' end), and so forth.

[0328] As used herein, “overhang”, “sticky end” and “overhanging sequence” are used interchangeably. They refer to one or more unpaired nucleotides extending beyond the duplex region at the end of a strand. Nucleotide overhangs are typically formed when the 3' end of one strand extends beyond the 5' end of the other strand, or when the 5' end of one strand extends

beyond the 3' end of the other strand. The length of a nucleotide overhang is generally in the range of 1 to 6 nucleotides, 1 to 5 nucleotides, 1 to 4 nucleotides, 1 to 3 nucleotides, 2 to 6 nucleotides, 2 to 5 nucleotides, or 2 to 4 nucleotides. In some embodiments, the nucleotide overhangs comprise 1, 2, 3, 4, 5, or 6 nucleotides. In one particular embodiment, the nucleotide overhang comprises 1 to 4 nucleotides. In some embodiments, the nucleotide overhang comprises 2 nucleotides. In some other embodiments, the nucleotide overhang comprises a single nucleotide.

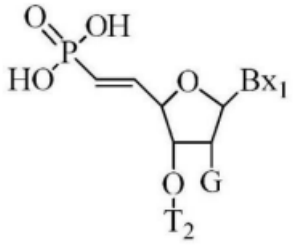
[0329] The nucleotides in an overhang may be ribonucleotides or modified nucleotides as described herein. In some embodiments, the nucleotides in the overhang are 2'-modified nucleotides (e.g., 2'-fluoro modified nucleotides, 2'-O-methyl modified nucleotides), deoxyribonucleotides, inverted nucleotides (e.g., inverted abasic nucleotides, inverted deoxyribonucleotides), or combinations thereof. For example, in one embodiment, the nucleotides in the overhang are deoxyribonucleotides, such as deoxythymidine. In another embodiment, the nucleotides in the overhang are 2'-O-methyl modified nucleotides, 2'-fluoro modified nucleotides, 2'-methoxyethyl modified nucleotides, or combinations thereof. In other embodiments, the overhang comprises 5'-uridine-uridine-3' (5'-UU-3') dinucleotide. In such embodiments, the UU dinucleotide may comprise a ribonucleotide or a modified nucleotide, such as a 2'-modified nucleotide. In other embodiments, the overhang comprises 5'-deoxythymidine-deoxythymidine-3' (5'-dTdT-3') dinucleotide. Where a nucleotide overhang is present on the antisense strand, the nucleotides of the overhang may be complementary to the target gene sequence, form mismatches with the target gene sequence, or contain other sequences (e.g., polypyrimidine or polypurine sequences such as UU, TT, AA, GG and the like).

[0330] A nucleotide overhang may be located at the 5' end or 3' end of one or both strands. For example, in one embodiment, the RNA molecule comprises nucleotide overhangs at both the 5' end and 3' end of the antisense strand. In another embodiment, the RNA molecule has nucleotide overhangs at both the 5' end and 3' end of the sense strand. In some embodiments, the RNA molecule has nucleotide overhangs at the 5' end of the sense strand and the 5' end of the antisense strand. In other embodiments, the RNA molecule has nucleotide overhangs at the 3' end of the sense strand and the 3' end of the antisense strand.

[0331] An RNA molecule may have a nucleotide overhang at one end of a double-stranded RNA molecule and a blunt end at the other end. “Blunt end” means that the sense strand and the antisense strand are fully base-paired at their molecular termini, with no unpaired nucleotides extending beyond the duplex region. In some embodiments, the RNA molecule has

a nucleotide overhang at the 3' end of the sense strand, and blunt ends at the 5' end of the sense strand and the 3' end of the antisense strand. In other embodiments, the RNA molecule has a nucleotide overhang at the 3' end of the antisense strand, and blunt ends at the 5' end of the antisense strand and the 3' end of the sense strand. In some embodiments, the RNA molecule has blunt ends at both ends of the double-stranded RNA molecule. In such embodiments, the sense strand and the antisense strand are of equal length, and the length of the duplex region is identical to that of the sense strand and the antisense strand (i.e., the molecule is double-stranded along its entire length). As used herein, "GalNAc" or "N-acetylgalactosamine" refers to 2-(acetylamino)-2-deoxy-D-galactopyranose. Unless otherwise specified, the terms "GalNAc" and "N-acetylgalactosamine" encompass both the β-form: 2-(acetylamino)-2-deoxy-β-D-galactopyranose and the α-form: 2-(acetylamino)-2-deoxy-α-D-galactopyranose. Preferably, the GalNAc compound used herein is the β-form, namely 2-(acetylamino)-2-deoxy-β-D-galactopyranose.

[0332] As used herein, "modification VP" or "E-VP" refers to a 5'-phosphate mimic modification located on the antisense strand. Its specific structure is as follows:



[0333] wherein:

[0334] Bx1 represents uracil, thymine, cytosine, 5-methylcytosine, adenine or guanine;

[0335] T_2 denotes a phosphodiester or phosphorothioate internucleotide linkage that connects the above compound to the oligonucleotide strand; G represents halogen, OCH_3, OCF_3, OCH_2CH_3, OCH_2CF_3, OCH_2-CH=CH_2, $O(CH_2)_2$-OCH_3, $O(CH_2)_2$-$O(CH_2)_2$-$N(CH_3)_2$, OCH_2C(=O)-$N(H)CH_3$, OCH_2C(=O)-$N(H)$-$(CH_2)_2$-$N(CH_3)_2$ or OCH_2-$N(H)$-C(=$NH)NH_2$.

[0336] As used herein, "3' end" specifically refers to the position of the first nucleotide or the first base pair at the 3' end of a single-stranded nucleotide sequence or a double-stranded polynucleotide. The term "5' end" specifically refers to the position of the first nucleotide or the first base pair at the 5' end of a single-stranded nucleotide sequence or a double-stranded polynucleotide.

[0337] As used herein, the term “nucleic acid molecule” refers to any molecule comprising a nucleotide sequence formed by more than two nucleotides linked via phosphoester bonds or modified phosphoester bonds (such as phosphorothioate bonds).

[0338] As used herein, the term “nucleotide sequence” means a polynucleotide strand consisting of nucleotides arranged in a specific order. Such a polynucleotide strand may constitute an entire nucleic acid molecule or a fragment of one strand of a nucleic acid molecule. Accordingly, a “nucleotide sequence” may be represented as a definite polynucleotide sequence composed of various nucleotides (e.g., ATCG), such as any one of SEQ ID NO: 1 to 100. It may also refer to nucleotides spanning from one position to another in a given sequence, for example, “nucleotides at positions 363 to 382” in the “FXI gene mRNA sequence”. As used herein, unless otherwise specified, for any sequence or nucleic acid molecule comprising “nucleotides from one position to another in a given sequence”, the arrangement order of the nucleotides from one position to another is identical to that of the aforesaid nucleotides in the given sequence. Without limitation, a “nucleotide sequence” may be RNA, DNA, or a hybrid molecule of RNA and DNA, and may also incorporate non-naturally occurring nucleotides or artificially modified nucleotides.

[0339] In addition, REL or relative expression level shown in the drawings of the present application refers to the relative expression level of mRNA. In the context of sequences, N denotes a ribonucleotide, dN denotes a deoxyribonucleotide (DNA), Nm denotes a 2'-O-Me modified nucleotide or a 2'-O-methyl modified nucleotide; Nf denotes a 2'-F modified ribonucleotide or a 2'-fluoro modified ribonucleotide; s denotes a phosphorothioate modification, namely a 5'-thio modified phosphate. When present between two nucleotides or between a nucleotide and a ligand, this modification replaces the phosphoester bond between native nucleotides and is referred to as a phosphorothioate bond. As used herein, unless otherwise specified, “phosphorothioate bond” and “phosphorothioate diester bond” are used interchangeably. Similarly, “phosphoester bond” and “phosphodiester bond” are used interchangeably.

[0340] As used herein, the term “about” refers to the customary margin of error for respective values as commonly known to those skilled in the art. References to a value or parameter preceded by “about” include embodiments directed to the value or parameter per se. As used herein, when the term “about” precedes a numerical value, it means a range within 10% above or below such value. For example, “about 100” encompasses the range from 90 to 110.

[0341] As used herein, the singular forms “a”, “an” and “the” include plural referents unless the context clearly dictates otherwise.

[0342] Unless otherwise defined herein, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which the present invention belongs.

[0343] It is to be understood that the present application covers all aspects, embodiments described herein, as well as combinations of the aforesaid aspects and/or embodiments. The foregoing description and the subsequent examples are intended to illustrate, rather than limit, the scope of the present application. Various simple modifications may be made to the technical solutions of the present application within the scope of its technical concept, including combining individual technical features in any other suitable manner. All such simple modifications and combinations shall be deemed part of the disclosure of the present application and fall within its protection scope.

[0344] Unless otherwise indicated, the practice of the present application employs conventional techniques of cell biology, cell culture, molecular biology, transgenics, microbiology, recombinant DNA technology and immunology. Such conventional techniques are well documented in the prior art.

[0345] It is to be understood that the present application covers all aspects, embodiments described herein, as well as combinations of the aforesaid aspects and/or embodiments. The foregoing description and the subsequent examples are intended to illustrate, rather than limit, the scope of the present application. Other aspects, improvements and modifications falling within the scope of the present application will be apparent to those skilled in the art. Accordingly, a person of ordinary skill in the art will recognize that the scope of the present application also covers such improvements and modifications to the aforesaid aspects and embodiments.

EXAMPLES

[0346] Example 1: Sequence design and synthesis of dsRNA

[0347] In Examples 1 to 7 of the present application, none of the nucleotides comprise modifications not specified herein. That is, all nucleotide sequences used in Examples 1 to 7 of the present application are naked sequences, or only comprise modifications as indicated in their respective modification motifs.

[0348] 1.1 Design of dsRNA

[0349] Based on the human FXI gene (Gene ID, 2160) mRNA sequence (NM_000128.4), multiple FXI dsRNAs were designed at different sites. All designed

individual dsRNA sequences were compared with all other non-target gene sequences using sequence similarity software and showed the lowest homology. Details are listed in the Sequence Listing at the end of this document. For modified dsRNAs: if the first nucleotide at the 3' end of the sense strand is neither A nor U, the original nucleotide is replaced with uracil ribonucleotide. If the first nucleotide at the 5' end of the antisense sequence is neither A nor U, the original nucleotide is replaced with adenine ribonucleotide. The original nucleic acid sequence is changed from n to n' for detection. For instance, the nucleic acid sequences of E3, E4, E9, E14, E16, E24, E26, E51, E52, E69 and E70 correspond to the dsRNAs numbered 3', 4', 9', 14', 16', 24', 26', 51', 52', 69' and 70' respectively in the Sequence Listing at the end of this document.

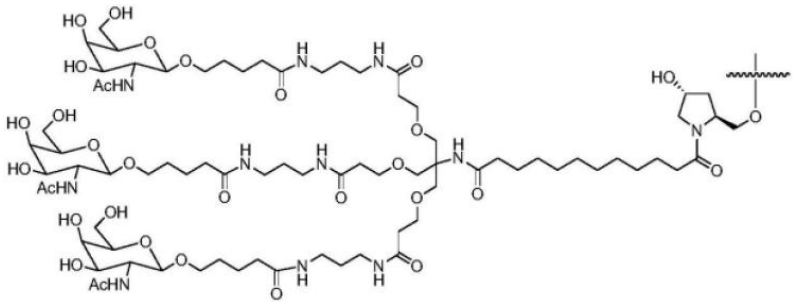
[0350] In the present application, the ASO sequence of ISIS-416858 disclosed in the patent of ionis (Patent No.: CN 109797150 A) is adopted. We also select the top four sequences with the highest inhibition activity against the FXI gene disclosed in Patent No. CN113227376A (naked sequences designated RB2, RB4, RB6 and RB7, and modified sequences designated ERB2, ERB4, ERB6 and ERB7, and these naked and modified sequences correspond to the ligand-free sequences of L10-siFXIa1M1SP, L10-siFXIc1M1SP, L10-siFXIe1M1SP and L10-siFXIg1M1SP as recorded in the aforesaid patent). Furthermore, the top three sequences with the highest inhibition activity against the FXI gene disclosed in Patent No. WO 2022028457A1 are chosen as positive control sequences for the FXI gene (their naked sequences are named TJ632, TJ645 and TJ772, and the modified sequences are named ETJ632, ETJ645 and ETJ772, which correspond to the ligand-free sequences of TRD0632, TRD0645 and TRD0772 specified in the corresponding patent respectively). The naked sequences of the aforementioned dsRNA sequence and positive control sequence are shown in the Sequence Listing at the end of this document.

[0351] 1.2 Synthesis and purification of dsRNA and conjugates thereof

[0352] dsRNA was synthesized using chemically modified (e.g., 2'-methoxy modified, 2'-fluoro modified or thio modified) or unmodified ribonucleotides at a theoretical synthesis scale of 1 μmol. All oligonucleotides were prepared on an LK-192X synthesizer (Jiangsu Lingkun Biotechnology Co., Ltd.) by adopting a common 1 μmol Frit support (1000 Å = 100 nm, Biocomma) or a CPG support loaded with protecting group-bearing GalNAc derivative L96 (Asymchem). In accordance with sequence requirements, all corresponding nucleoside phosphoramidite monomers were diluted with anhydrous acetonitrile at a ratio of 1:40 (g/mL). The coupling reaction was performed twice with a coupling time of 3 min for each run. Deprotection was performed using 3% TCA, activation was performed using 0.3 M

benzylthiotetrazole in acetonitrile, and capping and oxidation were performed using CAPA/CAPB and 50 mM I2 solution, respectively. After the trityl-off synthesis, the solid support was transferred to a 2 mL centrifuge tube, 1.2 mL of ammonia was added, and the tube was heated in an oven at 65 °C for 3 h to remove the protecting group. The mixture was then cooled to room temperature and concentrated under vacuum for 30 min. The resulting solution was filtered through a 0.22 μm membrane into a sample vial, and single-strand purification was performed via semi-preparative reversed-phase chromatography. The elution gradient was set from 7% to 30% (acetonitrile: 100 mM TEAA) over 10 min at a flow rate of 5 mL/min. After purification, the eluate was concentrated under vacuum and dried by rotary evaporation at room temperature. Finally, the sample was dissolved in water, and each solution was desalted on a GE Hi-Trap desalting column to elute the final oligonucleotide product. All properties and purity were confirmed using ESI-MS and IEX HPLC, respectively. The concentration was determined via UV measurement with a microplate reader. Equimolar sense strand and antisense strand were combined in a new shipping tube, heated at 95 °C for 5 min, and then slowly annealed to room temperature. The final dsRNA product was obtained after drying by vacuum concentration at room temperature.

[0353] The structure of L96 used in the example of the present application is shown in formula I below:



(Formula I);

[0354] wherein  denotes linkage to the 3'-end of the sense strand or antisense strand of the RNA molecule via a phosphodiester bond.

[0355] Example 2: High-throughput screening assay for in vitro activity of FXI-dsRNA

[0356] 2.1 Transfection of HepG2 cells with FXI dsRNA

[0357] HepG2 cells were cultured in DMEM medium containing 10% fetal bovine serum in a constant-temperature incubator at 37 °C with 5% CO_2. After cell digestion, the cell density was adjusted to 4×10^5 cells/ml, and 1 ml of cell suspension was seeded into each well of a 12-well plate. Preparation of 100 ul of transfection complex: 45 μL of Opti-MEM was mixed with 5 μL of dsRNA (naked nucleic acid, unmodified) at various concentrations. 48 μL

of Opti-MEM was combined with 2 μL of RNAiMax transfection reagent, and the mixture was allowed to stand for 5 min. The two resultant mixtures were then combined and allowed to stand for 20 min to form the transfection complex. The transfection complex was added to a 12-well plate and further cultured for 24 h in a constant-temperature incubator at 37 °C with 5% CO2. Cells were harvested 24 h after transfection, and RNA was extracted using the TRIZOL method.

[0358] 2.2 Real-time fluorescence quantitative PCR analysis

[0359] Cells were lysed 24 h after transfection, and total RNA was extracted using the Vazyme FastPure Cell/Tissue Total RNA Isolation Kit V2 (refer to the Vazyme RC112-01 instruction manual). Reverse transcription into cDNA was performed using Takara PrimeScript RT Master Mix RR036Q. The qPCR primer sequence information is shown in the primer section of the sequence listing at the end of this document. The human GAPDH gene was used as the internal reference gene, and the PCR reaction was performed using a CFX96 fluorescence quantitative PCR instrument from Bio-Rad, USA. In the experiment, normalization was carried out with the Mock group as the control, and the relative expression level of FXI mRNA in the Mock group was set to 1.

[0360] 2.3 Data analysis

[0361] After the PCR reaction was completed, relative quantitative analysis was performed using the reference gene as a standard and CFX96 software, and statistical analysis was performed using GarphPad software. As shown in Table 1 and FIG. 1, these are the results of high-throughput screening validation of dsRNA (naked nucleic acid, unmodified) molecules in HepG2 cells.

[0362] Table 1: High-throughput screening results at a single dose of 5 nM in HepG2 cells

No.	Relative FXI activity	±SEM
1	0.601	0.037
2	0.448	0.015
3	0.381	0.019
4	0.309	0.022
5	0.313	0.011
6	0.315	0.025
7	0.331	0.027
8	0.227	0.033
9	0.297	0.013
10	0.464	0.033
11	0.399	0.012

12	0.548	0.011
13	0.375	0.023
14	0.327	0.019
15	0.336	0.022
16	0.414	0.012
17	0.364	0.013
18	0.358	0.009
19	0.389	0.020
20	0.273	0.008
21	0.656	0.010
22	0.460	0.010
23	0.461	0.029
24	0.342	0.012
25	0.484	0.025
26	0.334	0.013
27	0.430	0.033
28	0.518	0.039
29	0.390	0.033
30	0.531	0.032
31	0.449	0.019
32	0.557	0.036
33	0.499	0.026
34	0.524	0.008
35	0.602	0.008
36	0.440	0.036
37	0.384	0.016
38	0.626	0.026
39	0.425	0.014
40	0.485	0.108
41	0.525	0.121
42	0.496	0.042
43	0.525	0.008
44	0.592	0.015
45	0.557	0.003
46	0.489	0.014
47	0.513	0.031
48	0.479	0.028
49	0.702	0.045
50	0.344	0.018
51	0.390	0.019
52	0.393	0.022
53	0.419	0.023
54	0.415	0.016

55	0.364	0.047
56	0.384	0.029
57	0.536	0.028
58	0.528	0.024
59	0.423	0.015
60	0.478	0.021
61	0.585	0.008
62	0.479	0.022
63	0.469	0.013
64	0.483	0.017
65	0.429	0.021
66	0.431	0.005
67	0.392	0.014
68	0.221	0.005
69	0.520	0.018
70	0.349	0.017
71	0.441	0.028
72	0.411	0.029
73	0.404	0.058
74	0.385	0.016
75	0.460	0.029
76	0.441	0.023
77	0.486	0.015
78	0.463	0.064
79	0.489	0.036
80	0.533	0.023
81	0.446	0.004
82	0.375	0.017
83	0.469	0.010
83	0.523	0.010
85	0.617	0.069
86	0.518	0.034
87	0.702	0.028
88	0.762	0.084
89	0.634	0.051
90	0.519	0.040
91	0.563	0.016
92	0.533	0.035
93	0.587	0.038
94	0.533	0.037
95	0.514	0.015
96	0.562	0.016
97	0.508	0.028

98	0.581	0.009
99	0.591	0.035
100	0.662	0.038
RB2	0.512	0.016
RB4	0.471	0.008
RB6	0.558	0.032
RB7	0.481	0.061
TJ632	0.484	0.011
TJ645	0.386	0.009
TJ772	0.485	0.017

[0363] Screening of FXI dsRNA in HepG2 cells revealed 28 preferred sequences, and dsRNA molecules with good efficacy and targeting FXI in both humans and cynomolgus monkeys were retained as candidate sequences. Furthermore, multiple positive control reference sequences were compared with our candidate sequences. Table 2 and FIG. 2 present the single-dose screening results of candidate sequences and positive control sequences in HepG2 cells.

[0364] Table 2: Single-dose screening of unmodified dsRNA in HepG2 cells

Sample name	Relative FXI mRNA expression (5nM)	±SEM	Relative FXI mRNA expression (1nM)	±SEM
3	0.201	0.024	0.400	0.027
4	0.244	0.017	0.389	0.032
5	0.236	0.008	0.301	0.049
6	0.234	0.016	0.359	0.011
7	0.266	0.020	0.434	0.034
8	0.153	0.011	0.290	0.026
9	0.167	0.004	0.346	0.017
13	0.282	0.011	0.423	0.019
14	0.225	0.028	0.340	0.037
15	0.250	0.020	0.448	0.027
16	0.253	0.013	0.503	0.005
17	0.746	0.028	0.465	0.012
18	0.224	0.003	0.465	0.010
19	0.276	0.003	0.466	0.004
20	0.320	0.006	0.375	0.006
24	0.194	0.009	0.297	0.102
26	0.190	0.046	0.402	0.031
50	0.229	0.019	0.352	0.031
51	0.239	0.007	0.496	0.009
52	0.312	0.031	0.492	0.032
53	0.251	0.016	0.435	0.018
54	0.268	0.015	0.439	0.026

55	0.327	0.014	0.481	0.016
56	0.335	0.012	0.544	0.026
68	0.251	0.028	0.382	0.020
69	0.398	0.015	0.406	0.024
70	0.226	0.007	0.350	0.005
82	0.246	0.007	0.430	0.008
RB2	0.254	0.008	0.412	0.018
RB4	0.297	0.009	0.433	0.011
RB6	0.475	0.030	0.560	0.035
RB7	0.331	0.008	0.400	0.007
TJ632	0.235	0.004	0.335	0.013
TJ645	0.307	0.012	0.363	0.005
TJ772	0.346	0.022	0.449	0.008

[0365] The results show that among the 7 candidate positive control sequences, TJ632 exhibited the best activity. Our multiple candidate dsRNA sequences demonstrated superior or equivalent efficacy compared with the positive control TJ632, including dsRNA No. 3, 5, 8, 9, 24, 26, 50, 70 and 82.

[0366] Example 3: Optimization of FXI-dsRNA

[0367] 3.1 Assay for inhibitory activity

[0368] To further validate the preferred dsRNA molecules, two distinct modification motifs were applied to the aforementioned preferred sequences. Combinations of fluoro and methoxy modifications were introduced at different positions of the candidate sequences.

[0369] The specific modification is selected from any of the following types:

[0370] (1) Modification E (dsRNA modified with E is designated as En, where n represents the number of the candidate dsRNA molecule, for instance, dsRNA molecule No. 3 modified with E is denoted as E3):

[0371] sense sequence : nucleotides at positions 7, 9, 10 and 11 starting from the 5' end are 2'-fluoro modified, and nucleotides at all other positions are 2'-O-methyl modified, the first and second phosphoester bonds at the 5' end are phosphorothioate bonds via thiation; for example, taking N to represent any ribonucleotide, the modified sense sequence is shown below: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmsNmsNm;

[0372] antisense sequence: nucleotides at positions 2, 6, 14 and 16 starting from the 5' end are 2'-fluoro modified (in other words, nucleotides at positions 2, 6, 14 and 16 starting from the 3' end of the region where the antisense sequence is complementary to the sense strand are 2'-fluoro modified), nucleotides at all other positions are 2'-O-methyl modified, and the first and second phosphoester bonds at both the 5' end and the 3' end are converted into phosphorothioate bonds via thiation; for example, taking N to represent any ribonucleotide, the

modified antisense sequence is shown below: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm.

[0373] (2) Modification E-1 (dsRNA modified with E-1 is designated as En-1, where n represents the number of the candidate dsRNA molecule, for instance, dsRNA molecule No. 3 modified with E-1 is denoted as E3-1),

[0374] the only difference between modification E-1 and modification E1 is that the 6th nucleotide starting from the 5' end of the antisense strand of dsRNA with modification E-1 is a 2'-O-methyl modified nucleotide instead of a 2'-fluoro modified nucleotide, as exemplified below:

[0375] modified sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmsNmsNm;

[0376] modified antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm.

[0377] In the sequence modifications described herein: s stands for phosphorothioate bond, m stands for 2'-O-methyl modification, and f stands for 2'-fluoro modification.

[0378] In addition, the antisense compound ISIS-416858 adopts the structure and modification pattern disclosed in CN109797150A, namely a “5-10-5” structure. Each flanking segment comprises five 2'-O-methoxyethyl modified nucleotides, the middle 10 deoxyribonucleotides and fully phosphorothioated, methylcytosine replacing cytosine.

[0379] HepG2 cells were used for transfection. The procedures for synthesis, transfection and quantitative PCR detection were consistent with those described in Example 2. Table 3 and FIG. 3 present the mean expression levels of the target gene relative to the blank control group (the relative mRNA expression level of the blank control group was set to 1).

[0380] Table 3: Single-dose (1 nM) screening of modified dsRNA in HepG2 cells

Sample name	Relative FXI mRNA expression	±SEM
E3	0.355	0.020
E4	0.454	0.039
E5	0.439	0.025
E6	0.401	0.039
E8	0.329	0.019
E9	0.437	0.018
E13	0.343	0.006
E14	0.302	0.035
E15	0.345	0.021
E16	0.371	0.008
E18	0.420	0.018
E19	0.329	0.013

E20	0.427	0.011
E24	0.326	0.013
E26	0.301	0.016
E50	0.252	0.011
E51	0.369	0.023
E52	0.351	0.021
E53	0.371	0.043
E54	0.368	0.011
E55	0.618	0.053
E56	0.455	0.034
E68	0.450	0.008
E69	0.344	0.009
E70	0.375	0.013
E82	0.323	0.009
ERB2	0.353	0.005
ERB4	0.467	0.015
ERB6	0.514	0.040
ERB7	0.342	0.008
ETJ632	0.318	0.005
ETJ645	0.358	0.021
ETJ772	0.405	0.022
ISIS-416858	0.734	0.016

[0381] It can be seen that compared with the chemically modified positive control sequences, multiple candidate sequences including E8, E14, E19, E24, E26 and E50 exhibited equivalent or better activity than the optimal positive control ETJ632. This result further demonstrates that the selected preferred sequences have the potential to effectively downregulate the nucleic acid level of the FXI gene.

[0382] Example 4: In vitro efficacy assay of GalNAc-dsRNA

[0383] Candidate modified dsRNAs were conjugated with GalNAc to form GAL-dsRNA complexes, i.e., each dsRNA was subjected to modification G.

[0384] Modification G refers to the conjugation of L96 to the 3' end of the dsRNA sense sequence, including modification GE and modification GE-1, wherein:

[0385] modification GE is shown below, where "-L96" indicates that L96 is conjugated to the 3' end of the dsRNA sense sequence:

[0386] modified sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96;

[0387] modified antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0388] dsRNA with modification GE is designated as GEn, where n represents the number of the candidate dsRNA molecule, for example, dsRNA No. 3 molecule modified with GE is denoted as GE3;

[0389] GE-1 is as shown below:

[0390] modified sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm-L96;

[0391] modified antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0392] Similarly, "-L96" means L96 is conjugated to the 3' end of the dsRNA sense sequence via a phosphoester bond, dsRNA with modification GE-1 is designated as GEn-1, where n represents the number of the candidate dsRNA molecule, for instance, dsRNA molecule No. 3 modified with GE-1 is denoted as GE3-1.

[0393] We further validated the activity of the G-modified candidate complex in HepG2 cells.

[0394] 4.1 Detection of mRNA expression levels in HepG2 cells

[0395] The detection steps are the same as in Example 2, with the human GAPDH gene as the internal reference gene, and the detection primers used are shown in the primer section of the sequence listing at the end of this document. Table 4 shows the average mRNA levels in HepG2 cells treated with G-modified dsRNA relative to the target gene expression levels in the untreated group (with the relative mRNA expression level in the untreated group set to 1). The statistical results are shown in FIG. 4.

[0396] Table 4 IC50 data of some GalNAc-conjugated dsRNAs in HepG2 cells

Sample name	IC_{50} (nM)
GE14	0.089
GE19	0.059
GE26	0.066
GE50	0.026
GE70	0.040
GE82	0.030

[0397] It can be seen that the IC50 values of all GalNAc-conjugated dsRNAs in HepG2 cells were below 0.1 nM, which further verifies that the candidate sequences exert potent inhibitory activity against the target gene.

[0398] Example 5: In vivo efficacy evaluation

[0399] SPF-grade male homozygous humanized FXI mice aged 6 to 8 weeks (purchased from Shanghai Model Organisms Center, Inc.) were used in this study. The mice

were randomly divided into groups with 6 animals per group. A single subcutaneous injection was administered to each group. Mice in different groups received distinct GE-modified dsRNAs, while the Blank group was injected with normal saline. Blood samples were collected on Day 7 and Day 13 after administration. Serum was obtained via retro-orbital bleeding for the detection of FXI protein levels.

[0400] The specific dosages are shown in Table 5.

[0401] Table 5 Single-dose regimen at 3 mpk

Drug name	Dosage	Humanized FXI mice
Blank	Normal saline	N=6, male
GE3	3mpk	
GE8		
GE14		
GE19		
GE24		
GE26		
GE50		
GE70		
GE82		

[0402] 5.1 ELISA Detection of FXI Protein

[0403] Serum samples were diluted at a ratio of 1:500, and the residual FXI content in plasma of each group was measured using the ab108834 ELISA kit (Abcam). All operations were performed in accordance with the product manual of ab108834 ELISA kit. The absorbance was read at 450 nm using a microplate reader, and data analysis was conducted with GraphPad software. The percentage inhibition was calculated relative to the untreated group. The results are presented in Table 6. FIG. 5 shows the relative expression levels of FXI protein in plasma of each group after normalization against the blank control group.

[0404] Table 6: Inhibition rate of FXI protein in plasma after a single 3mpk dose

Drug name	Day 7 Inhibition rate (%)	Day 13 Inhibition rate (%)
GE3	53.9	52.3
GE8	44.0	53.7
GE15	53.1	58.3
GE14	55.3	53.4
GE19	69.4	71.6
GE24	50.5	56.2
GE26	70.7	81.8
GE50	87.9	92.8
GE70	78.6	86.7
GE82	61.2	64.7

[0405] It is evident that, after a single administration at 3 mpk, the plasma FXI levels in mice of the groups treated with the candidate dsRNA molecules were significantly lower

than those in the blank control group. On day 13, the inhibition rates of GE26, GE50 and GE70 were all above 80%, with GE50 showing the highest inhibition rate of 92.8%. Meanwhile, on day 13, the inhibition rate was >50% in all treatment groups. These results demonstrate that our multiple dsRNA molecules designed against FXI can effectively inhibit the expression and secretion of FXI protein in the liver, and represent a potential therapeutic drug targeting the FXI gene.

[0406] Example 6: In vivo efficacy evaluation of different ligands

[0407] We further validated the effects of different ligand conjugates using candidate dsRNAs. Table 7 shows the dsRNA sequence information. The dsRNAs were modified with either LN-E05 or GN-E05. Modification LN-E05 is identical to modification GE-1 described in Example 4. The details of modification GN-E05 are as follows:

[0408] modified sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1;

[0409] modified antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

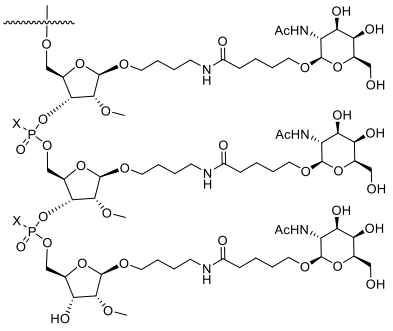
[0410] That is, the only difference between modification GN-E05 and modification LN-E05 is that the L96 ligand conjugated to the 3' end of the sense strand was replaced with ligand 1, and ligand 1 was linked to the 3' end of the sense strand via a phosphorothioate bond.

[0411] Table 7 Synthesized oligonucleotides

dsRNA sequence	Base sequence	Modification motif	Ligand structure
LN50-E05	Sense strand: SEQ ID NO: 58 Antisense strand: SEQ ID NO: 177	LN-E05	L96
GN50-E05		GN-E05	Ligand 1
LN26-E05	Sense strand: SEQ ID NO: 34 Antisense strand: SEQ ID NO: 153	LN-E05	L96
GN26-E05		GN-E05	Ligand 1

[0412] Grouping was performed in accordance with Table 7. All groups received a single subcutaneous injection at a dose of 3 mpk. Blood samples were collected at different time points after administration, and the plasma was used for the detection of FXI protein levels. The percentage residual rate was calculated relative to the pre-administration baseline. The results are shown in FIG. 6. After single administration at 3 mpk, the dsRNA conjugated with ligand 1 exhibited a significantly higher inhibition rate against FXI protein than the dsRNA conjugated with L96 (paired t-test, $p<0.05$).

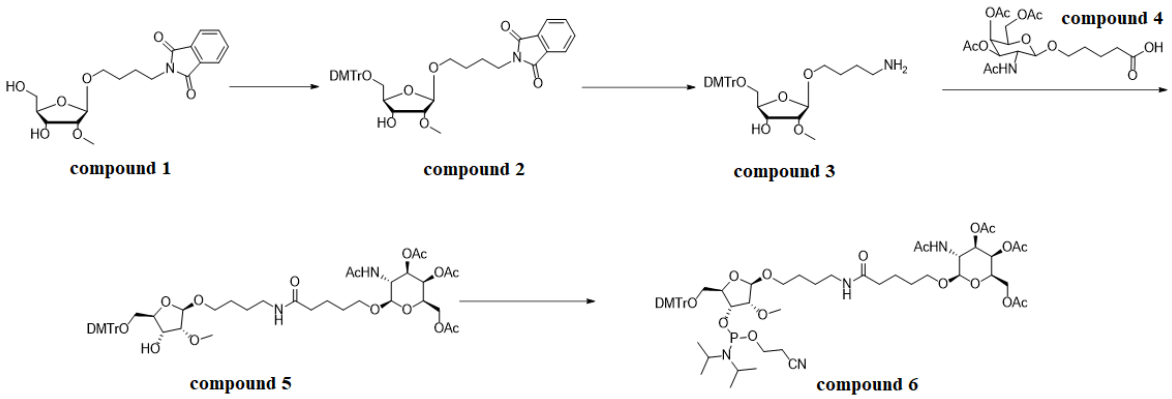
[0413] The structure of ligand 1 is shown in Formula II:



(X=S$^-$) (Formula II);

[0414] wherein the wavy line indicates the linkage to the 3' end of the dsRNA sense strand via a phosphorothioate bond. The molecule of formula II is a trivalent GalNAc conjugate synthesized from two equivalents of compound 6 and one equivalent of compound 8 using conventional raw materials and synthetic methods known in the art. The synthetic procedures for compound 6 and compound 8 are described below.

[0415] 1. Synthesis of compound 6



[0416] (1) Synthesis of compound 2:

[0417] To a three-necked flask were added 1.2 g of compound 1 and 12 mL of pyridine. After stirring to achieve dissolution, 1.2 g of DMTrCl was added. The mixture was stirred at room temperature for 1 h under nitrogen protection. Methanol (0.6 mL) was slowly added to the reaction mixture to quench excess DMTrCl. After stirring at room temperature for 15 min, 303.5 mg of NaHCO3 was supplemented. The mixture was concentrated to afford a crude product. DCM/H2O were added, followed by stirring for dissolution. The mixture was washed with water, and the organic phase was dried over anhydrous sodium sulfate and then filtered. The collected organic phase was concentrated under vacuum to give crude compound 2, which was used directly in the next reaction step.

[0418] (2) Synthesis of compound 3:

[0419] Ethanol (24 mL) was added to the crude compound 2. The mixture was gradually heated to 50 °C and stirred until fully dissolved. Hydrazine hydrate (0.75 mL) was

then added, and the reaction was stirred overnight at 50 °C. Afterwards, the system was cooled to room temperature and stirred for another half an hour, during which a large amount of white solid gradually precipitated. The mixture was filtered, and the filter cake was washed with ethanol. The resulting filtrate was concentrated under reduced pressure, redissolved in DCM/H2O, and then separated. The organic phase was collected, washed with saturated brine, dried over anhydrous sodium sulfate and filtered. The collected organic phase was concentrated under vacuum to yield crude compound 3, which was carried forward to the subsequent reaction directly.

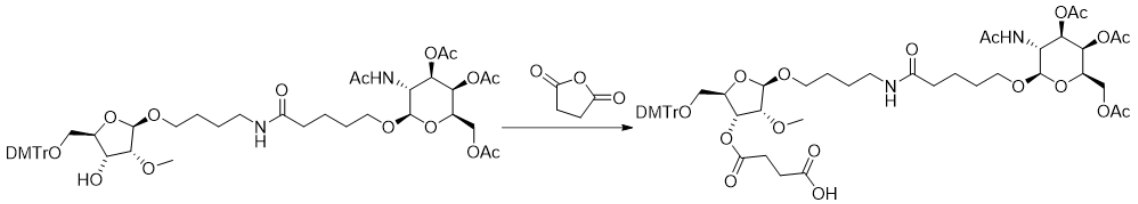
[0420] (3) Synthesis of compound 5:

[0421] To the crude compound 3 were sequentially added 0.9 g of compound 4, DCM (24 mL), 0.84 mL of Et3N and 2.3 g of HBTU. The reaction mixture was stirred at room temperature for 2 h under nitrogen protection. The reaction was quenched with 9 mL of water. The organic phase was separated, washed with saturated brine, dried over anhydrous sodium sulfate and filtered. The collected organic phase was concentrated under vacuum, and the residue was purified by reversed-phase preparative chromatography (75%ACN-H2O) to give 1.4 g of compound 5. The overall yield over three steps was 44.2%. ESI-MS: m/z 1025.6 $[M^{+}OAc^{-}]^{-}$.

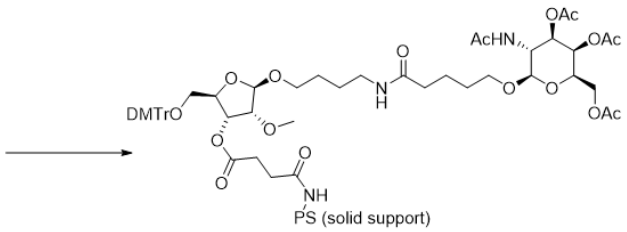
[0422] (4) Synthesis of compound 6:

[0423] 1.4 g of compound 5 was dissolved in DCM (14 mL) and cooled to 0±2 °C. 524.7 mg of 2-cyanoethyl-N,N,N',N'-tetraisopropylphosphorodiamidite was added, and then 81.3 mg of 1H-tetrazole was added. The reaction mixture was first stirred at 0 ± 2 °C for 15 min, then warmed to room temperature and stirred for an additional 2 h. The reaction mixture was cooled to 0 ± 2 °C and quenched with 14 mL of 5% NaHCO3. The organic phase was separated, washed with saturated brine (1x14mL) at 0 ± 2 °C, dried over anhydrous sodium sulfate and filtered. The collected organic phase was concentrated under vacuum. The resulting crude product was redissolved in DCM/MTBE, and the solution was slowly added dropwise into vigorously stirred n-heptane (hept.). A large amount of viscous oily substance gradually deposited on the wall and bottom of the flask. After standing for 10 min, the supernatant was decanted. The crude product was purified by column chromatography using EA:hept.=10:1-2:1 as eluent. After concentration and drying, 560 mg of white solid powder was obtained with a yield of 32% and a purity of 98.19% determined by phosphorus NMR. ESI-MS: m/z 1225.4 $[M^{+}OAc^{-}]^{-}$.

[0424] 2. Synthesis of compound 8



compound 5 **compound 7**



compound 8

[0425] (1) Synthesis of compound 7:

[0426] 2 g of compound 5 was dissolved in DCM (20 mL), and 248.4 mg of succinic anhydride and 0.69 mL of Et3N were added sequentially. Finally, 25.3 mg of DMAP was added. The mixture was stirred at room temperature for 16 h. HPLC monitoring indicated that a large amount of starting material remained, so additional 310.4 mg of succinic anhydride and 0.69 mL of Et3N were supplemented. Finally, 25.3 mg of DMAP was added and the mixture was further stirred at room temperature for 36 h. The reaction mixture was cooled to 0 ± 2 °C, then ice water and DCM were added successively, followed by 40 mL of 1% aqueous HOAc solution. After stirring for 10 min, the organic phase was separated. It was washed sequentially with 1% aqueous HOAc solution and water, dried over anhydrous sodium sulfate and filtered. The collected organic phase was concentrated under vacuum to give a crude product. The crude product was purified by column chromatography using DCM:MeOH=70:1-20:1 as eluent. After concentration and drying, 0.9 g of white solid powder was obtained with a yield of 40.7%. No residual succinic acid was detected, and the residual content of succinic anhydride was 0.26%. ESI-MS: m/z 1065.7$[M^{-}H]^{-}$.

[0427] (2) Synthesis of compound 8:

[0428] To a three-necked flask were added 300 mg of compound 7, 72.7 mg of DIPEA, 106 mg of HBTU and 10 mL of acetonitrile. The mixture was stirred at 25 °C for 10 min, then 650 mg of solid support PS was added, and stirring was continued at 25 °C for 24 h. Upon completion of the reaction, the mixture was filtered. The filter cake was rinsed with acetonitrile and concentrated under vacuum to remove the solvent, affording 850 mg of solid. A 700 mg portion of the solid was transferred to a three-necked flask, and 1.74 g of acetic anhydride, 4.15

mg of DMAP, 103 mg of triethylamine and 10 mL of pyridine were added. The mixture was stirred at 25 °C for 4 h. Upon completion of the reaction, the mixture was filtered. The filter cake was washed successively with acetonitrile, methanol and acetonitrile, then the filter cake was collected and concentrated under vacuum to remove the solvent, yielding 750 mg of compound 8. The measured loading capacity was 254.82 μmol/g.

[0429] 3. Preparation of oligonucleotide sequence conjugated with ligand 1

[0430] Compound 8 reacted sequentially with 2 equivalents of compound 6, or with 3 equivalents of compound 6 in sequence. Conventional solid-phase synthesis cycles (deprotection, coupling, oxidation and capping) were performed to give the precursor (or intermediate) of ligand 1 immobilized on the solid support. Subsequent solid-phase synthesis cycles were carried out using the corresponding 2'-modified monomers. After the synthesis was completed, the entire synthesized molecule was dissociated from the solid support via ammonolysis, to give the oligonucleotide sequence conjugated with ligand 1.

[0431] Example 7: In vitro and in vivo efficacy evaluation of different modification motifs

[0432] We further evaluated the effects of different modification motifs using dsRNAs sharing the base sequences of Duplex No. 50 and Duplex No. 26. The specific molecules used are shown in Table 8. It can be seen that all molecules in Table 8 were conjugated with Ligand 1 at the 3' end of the sense strand via a phosphorothioate bond, and the nucleotide at the 5' end of the antisense strand was modified with 5'-vp.

[0433] Table 8: Synthesized oligonucleotides

dsRNA No.	Base sequence	Chemical modification
GN50-E20VP	Sense strand: SEQ ID NO: 58 Antisense strand: SEQ ID NO: 177	GN-E20VP
GN50-E04VP		GN-E04VP
GN50-E05VP		GN-E05VP
GN50-E05		GN-E05
GN26-E20VP	Sense strand: SEQ ID NO: 34 Antisense strand: SEQ ID NO: 153	GN-E20VP
GN26-E04VP		GN-E04VP
GN26-E05VP		GN-E05VP
GN26-E05		GN-E05

[0434] The modification motifs in Table 8 are shown below:

[0435] GN-E20VP:

[0436] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1

[0437] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0438] GN-E04VP:

[0439] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1

[0440] antisense sequence: vp-NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

[0441] GN-E05VP:

[0442] sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1

[0443] antisense sequence: vp-NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm.

[0444] Modification GN-E05 is the same as in Example 6, except that the only difference from modification GN-E05VP is that the 5' end of the antisense strand of GN-E05 has no vinyl phosphate modification.

[0445] 7.1 In vitro efficacy evaluation of different modification motifs

[0446] First, the effects of various modifications listed in Table 8 on efficacy were evaluated in HepG2 cells in vitro. The experimental methods were performed in accordance with Example 4. Table 4 presents the average mRNA relative expression levels of the target gene in each treatment group relative to the untreated group (with the relative mRNA expression level of the untreated group set as 1). The in vitro assays were performed as independent replicate experiments in different batches, and the statistical results are shown in Table 9 and Table 10.

[0447] Table 9 IC50 data of dsRNA with modification 5VP in HepG2 cells

Sample name	IC_{50} (nM)
GN26-E05	0.117
GN26-E05VP	0.041
GE50-E05	0.034
GN50-E05VP	0.026

[0448] Table 10 IC50 data of dsRNA with different motif modifications in HepG2 cells

Sample name	IC_{50} (nM)
GN26-E04VP	0.167
GN26-E05VP	0.099
GN26-E20VP	0.044

[0449] The in vitro results are shown in Table 9 and FIG. 7, indicating that modification 5-VP helps to further improve the efficacy. Meanwhile, with the same ligand and base sequence, the modification motifs of GN-E05VP and GN-E20VP were significantly superior to those of GN-E04VP (as shown in Table 10). Furthermore, the modification motif of GN-E20VP was also significantly better than that of GN-E05VP.

[0450] 7.2 In vivo efficacy evaluation of different modification motifs

[0451] The experimental protocol was referred to Example 5. Groups specified in Table 8 received a single subcutaneous injection of No. 50 dsRNA with different modifications at a dose of 1 mpk. Blood samples were collected on Day 7, 14, 21 and 28 after administration, and plasma was used for the detection of FXI protein levels. The percentage residual rate was calculated relative to the pre-administration baseline. The results are presented in Table 11 and FIG. 8.

[0452] Table 11: Residual rate of plasma FXI protein after single administration at 1 mpk

Drug name	Day 7 Residual rate (%)	Day 14 Residual rate (%)	Day 28 Residual rate (%)
GN50-E05	16.3	11.9	20.4
GN50-E04VP	14.0	10.1	14.9
GN50-E05VP	10.7	7.7	12.2
GN50-E20VP	9.4	6.6	9.1

[0453] As indicated by the results of the modified molecule No. 50, following single administration at 1 mpk, with the same dsRNA base sequence, the modification motifs E05VP and E20VP were superior to E04VP ($P<0.05$), and E20VP was further superior to E05VP ($P < 0.05$). Meanwhile, the results show that modification 5'-VP helps to further improve the efficacy.

[0454] Furthermore, we compared the efficacy of different modification motifs of molecule No. 26 in vivo (as shown in Table 8). Blood samples were collected on days 7 and 14 after administration, and plasma was used for the detection of FXI protein levels. The percentage residual rate was calculated relative to the pre-administration baseline. The results are shown in Table 12.

[0455] Table 12: Residual rate of plasma FXI protein after single administration at 1 mpk

Drug name	Day 7 Residual rate (%)	Day 14 Residual rate (%)
GN26-E04VP	20.6	24.7
GN26-E05VP	17.2	18.0

GN26-E20VP	12.4	14.0

[0456] The superior efficacy of E05VP and E20VP over E04VP was further confirmed with molecule No. 26, and the E20VP motif exhibited the best efficacy overall. In addition, modification 5'-VP contributed to further improved efficacy.

[0457] The sequences used in the above examples of the present application are listed in the following Sequence Listing. It should be understood that the following sequences are merely exemplary sequences for the embodiments of this application and shall not be construed as any limitation thereto. The nucleic acid sequences in the following Sequence Listing can represent DNA or RNA sequences. When they represent RNA sequences, "T" represents uridine. Furthermore, in the context of RNA, unless otherwise specified, both "T" and "U" refer to uracil or uridine.

[0458] Sequence Listing:

dsRNA No.	Sense sequence (5'->3')	Sense sequence ID	Antisense sequence (5'->3')	Antisense sequence ID
1	CCGATGGTTTACTTGTGTCCT	SEQ ID NO: 1	AGGACACAAGTAAACCATCGGGT	SEQ ID NO: 120
2	CTTTCAAGCAATGCTCACACC	SEQ ID NO: 2	GGTGTGAGCATTGCTTGAAAGAA	SEQ ID NO: 121
3	GCAATGCTCACACCAAATAAG	SEQ ID NO: 3	CTTATTTGGTGTGAGCATTGCTT	SEQ ID NO: 122
3'	GCAATGCTCACACCAAATAAT	SEQ ID NO: 4	ATTATTTGGTGTGAGCATTGCTT	SEQ ID NO: 123
4	CAATGCTCACACCAAATAAGC	SEQ ID NO: 5	GCTTATTTGGTGTGAGCATTGCT	SEQ ID NO: 124
4'	CAATGCTCACACCAAATAAGT	SEQ ID NO: 6	ACTTATTTGGTGTGAGCATTGCT	SEQ ID NO: 125
5	ACCAAATAAGCGCTTGCAACA	SEQ ID NO: 7	TGTTGCAAGCGCTTATTTGGTGT	SEQ ID NO: 126
6	CCAAATAAGCGCTTGCAACAA	SEQ ID NO: 8	TTGTTGCAAGCGCTTATTTGGTG	SEQ ID NO: 127
7	CAAAGACATTTATGTGGACCT	SEQ ID NO: 9	AGGTCCACATAAATGTCTTTGTT	SEQ ID NO: 128
8	AGGGCATAAACTATAACAGCT	SEQ ID NO: 10	AGCTGTTATAGTTTATGCCCTTC	SEQ ID NO: 129
9	GGGCATAAACTATAACAGCTC	SEQ ID NO: 11	GAGCTGTTATAGTTTATGCCCTT	SEQ ID NO: 130
9'	GGGCATAAACTATAACAGCTT	SEQ ID NO: 12	AAGCTGTTATAGTTTATGCCCTT	SEQ ID NO: 131

10	ATGCCAAGAAAGATGCACGGA	SEQ ID NO: 13	TCCGTGCATCTTTCTTGGCATTC	SEQ ID NO: 132
11	ACTTTTTCACGTACGCCACAA	SEQ ID NO: 14	TTGTGGCGTACGTGAAAAAGTGG	SEQ ID NO: 133
12	ATTTCCTACACAAGCTCATTC	SEQ ID NO: 15	GAATGAGCTTGTGTAGGAAATGG	SEQ ID NO: 134
13	TGCTCACACCAAATAAGCGCT	SEQ ID NO: 16	AGCGCTTATTTGGTGTGAGCATT	SEQ ID NO: 135
14	AAATAAGCGCTTGCAACAAAG	SEQ ID NO: 17	CTTTGTTGCAAGCGCTTATTTGG	SEQ ID NO: 136
14’	AAATAAGCGCTTGCAACAAAT	SEQ ID NO: 18	ATTTGTTGCAAGCGCTTATTTGG	SEQ ID NO: 137
15	CATGAAGGGCATAAACTATAA	SEQ ID NO: 19	TTATAGTTTATGCCCTTCATGTC	SEQ ID NO: 138
16	GAAGGGCATAAACTATAACAG	SEQ ID NO: 20	CTGTTATAGTTTATGCCCTTCAT	SEQ ID NO: 139
16’	GAAGGGCATAAACTATAACAT	SEQ ID NO: 21	ATGTTATAGTTTATGCCCTTCAT	SEQ ID NO: 140
17	AAGGGCATAAACTATAACAGC	SEQ ID NO: 22	GCTGTTATAGTTTATGCCCTTCA	SEQ ID NO: 141
18	GGCATAAACTATAACAGCTCA	SEQ ID NO: 23	TGAGCTGTTATAGTTTATGCCCT	SEQ ID NO: 142
19	AAGAAAGATGCACGGATGACG	SEQ ID NO: 24	CGTCATCCGTGCATCTTTCTTGG	SEQ ID NO: 143
20	CTGCCACTTTTTCACGTACGC	SEQ ID NO: 25	GCGTACGTGAAAAAGTGGCAGTG	SEQ ID NO: 144
20’	CTGCCACTTTTTCACGTACGT	SEQ ID NO: 26	ACGTACGTGAAAAAGTGGCAGTG	SEQ ID NO: 145
21	AGCACCCTTATTAAGAATTGC	SEQ ID NO: 27	GCAATTCTTAATAAGGGTGCTTT	SEQ ID NO: 146
22	GTTTCTGGTGAATGTGTGACT	SEQ ID NO: 28	AGTCACACATTCACCAGAAACTG	SEQ ID NO: 147
23	GGAGGGGACATTACTACGGTC	SEQ ID NO: 29	GACCGTAGTAATGTCCCCTCCTT	SEQ ID NO: 148
24	ACTGCCAAGAGTGAATAGGAC	SEQ ID NO: 30	GTCCTATTCACTCTTGGCAGTGT	SEQ ID NO: 149
24’	ACTGCCAAGAGTGAATAGGAT	SEQ ID NO: 31	ATCCTATTCACTCTTGGCAGTGT	SEQ ID NO: 150
25	TTTCAAGCAATGCTCACACCA	SEQ ID NO: 32	TGGTGTGAGCATTGCTTGAAAGA	SEQ ID NO: 151
26	TCACACCAAATAAGCGCTTGC	SEQ ID NO: 33	GCAAGCGCTTATTTGGTGTGAGC	SEQ ID NO: 152

26’	TCACACCAAATAAGCGCTTGT	SEQ ID NO: 34	ACAAGCGCTTATTTGGTGTGAGC	SEQ ID NO: 153
27	ACATGAAGGGCATAAACTATA	SEQ ID NO: 35	TATAGTTTATGCCCTTCATGTCT	SEQ ID NO: 154
28	TGAAGGGCATAAACTATAACA	SEQ ID NO: 36	TGTTATAGTTTATGCCCTTCATG	SEQ ID NO: 155
29	GTTGCCAAGAGTGCTCAAGAA	SEQ ID NO: 37	TTCTTGAGCACTCTTGGCAACTG	SEQ ID NO: 156
30	GCACCCTTATTAAGAATTGCA	SEQ ID NO: 38	TGCAATTCTTAATAAGGGTGCTT	SEQ ID NO: 157
31	ACAAGGTCTTTTCAGGATGAT	SEQ ID NO: 39	ATCATCCTGAAAAGACCTTGTTG	SEQ ID NO: 158
32	ACACAAGCTCATTCAGAGGAG	SEQ ID NO: 40	CTCCTCTGAATGAGCTTGTGTAG	SEQ ID NO: 159
33	GTCCACTGCCACTTTTTCACG	SEQ ID NO: 41	CGTGAAAAAGTGGCAGTGGACGT	SEQ ID NO: 160
34	GAAAAGCACCCTTATTAAGAA	SEQ ID NO: 42	TTCTTAATAAGGGTGCTTTTCTT	SEQ ID NO: 161
35	AAAAGCACCCTTATTAAGAAT	SEQ ID NO: 43	ATTCTTAATAAGGGTGCTTTTCT	SEQ ID NO: 162
36	TCCACTGCCACTTTTTCACGT	SEQ ID NO: 44	ACGTGAAAAAGTGGCAGTGGACG	SEQ ID NO: 163
37	CACTGCCACTTTTTCACGTAC	SEQ ID NO: 45	GTACGTGAAAAAGTGGCAGTGGA	SEQ ID NO: 164
38	TTTCCTACACAAGCTCATTCA	SEQ ID NO: 46	TGAATGAGCTTGTGTAGGAAATG	SEQ ID NO: 165
39	CCACTGCCACTTTTTCACGTA	SEQ ID NO: 47	TACGTGAAAAAGTGGCAGTGGAC	SEQ ID NO: 166
40	GATGCACGGATGACGTCCACT	SEQ ID NO: 48	AGTGGACGTCATCCGTGCATCTT	SEQ ID NO: 167
41	CTGAGAGTGGATTGCCCAGTA	SEQ ID NO: 49	TACTGGGCAATCCACTCTCAGAT	SEQ ID NO: 168
42	AGAAAGATGCACGGATGACGT	SEQ ID NO: 50	ACGTCATCCGTGCATCTTTCTTG	SEQ ID NO: 169
43	GAAAGATGCACGGATGACGTC	SEQ ID NO: 51	GACGTCATCCGTGCATCTTTCTT	SEQ ID NO: 170
44	AGATGCACGGATGACGTCCAC	SEQ ID NO: 52	GTGGACGTCATCCGTGCATCTTT	SEQ ID NO: 171
45	GAAACTGTGCACCAATGCCGT	SEQ ID NO: 53	ACGGCATTGGTGCACAGTTTCTG	SEQ ID NO: 172
46	TCCGCTGCCAGTTTTTTACCT	SEQ ID NO: 54	AGGTAAAAAACTGGCAGCGGACG	SEQ ID NO: 173

47	AGTTTTTTACCTATACCCCAG	SEQ ID NO: 55	CTGGGGTATAGGTAAAAAACTGG	SEQ ID NO: 174
48	GTTTTTTACCTATACCCCAGC	SEQ ID NO: 56	GCTGGGGTATAGGTAAAAAACTG	SEQ ID NO: 175
49	TTTTTTACCTATACCCCAGCC	SEQ ID NO: 57	GGCTGGGGTATAGGTAAAAAACT	SEQ ID NO: 176
50	AAGTGTTACTTAAAGCTTTCT	SEQ ID NO: 58	AGAAAGCTTTAAGTAACACTTGC	SEQ ID NO: 177
51	AGCTTTCTTCAAACGGATCTC	SEQ ID NO: 59	GAGATCCGTTTGAAGAAAGCTTT	SEQ ID NO: 178
51’	AGCTTTCTTCAAACGGATCTT	SEQ ID NO: 60	AAGATCCGTTTGAAGAAAGCTTT	SEQ ID NO: 179
52	ACGGATCTCCAACTAAAATAC	SEQ ID NO: 61	GTATTTTAGTTGGAGATCCGTTT	SEQ ID NO: 180
52’	ACGGATCTCCAACTAAAATAT	SEQ ID NO: 62	ATATTTTAGTTGGAGATCCGTTT	SEQ ID NO: 181
53	AACTAAAATACTTCACGGGAG	SEQ ID NO: 63	CTCCCGTGAAGTATTTTAGTTGG	SEQ ID NO: 182
54	CTTTGTCTCCTTAAAACATCT	SEQ ID NO: 64	AGATGTTTTAAGGAGACAAAGAT	SEQ ID NO: 183
55	TTGCCCAGTACACGCATTAAA	SEQ ID NO: 65	TTTAATGCGTGTACTGGGCAATC	SEQ ID NO: 184
56	CCAGTACACGCATTAAAAAGA	SEQ ID NO: 66	TCTTTTTAATGCGTGTACTGGGC	SEQ ID NO: 185
57	CAGTACACGCATTAAAAAGAG	SEQ ID NO: 67	CTCTTTTTAATGCGTGTACTGGG	SEQ ID NO: 186
58	TCAGTCTACAAAGCTGCAGGC	SEQ ID NO: 68	GCCTGCAGCTTTGTAGACTGAAA	SEQ ID NO: 187
59	TAAGCCAACAAGGTCTTTTCA	SEQ ID NO: 69	TGAAAAGACCTTGTTGGCTTACT	SEQ ID NO: 188
60	AAGCCAACAAGGTCTTTTCAG	SEQ ID NO: 70	CTGAAAAGACCTTGTTGGCTTAC	SEQ ID NO: 189
61	GAGGGGACATTACTACGGTCT	SEQ ID NO: 71	AGACCGTAGTAATGTCCCCTCCT	SEQ ID NO: 190
62	GGGGACATTACTACGGTCTTC	SEQ ID NO: 72	GAAGACCGTAGTAATGTCCCCTC	SEQ ID NO: 191
63	GACATTACTACGGTCTTCACA	SEQ ID NO: 73	TGTGAAGACCGTAGTAATGTCCC	SEQ ID NO: 192
64	TCTTCACTTTCACGGCGGAAT	SEQ ID NO: 74	ATTCCGCCGTGAAAGTGAAGAGT	SEQ ID NO: 193
65	CTTCACTTTCACGGCGGAATC	SEQ ID NO: 75	GATTCCGCCGTGAAAGTGAAGAG	SEQ ID NO: 194

66	TTCACTTTCACGGCGGAATCA	SEQ ID NO: 76	TGATTCCGCCGTGAAAGTGAAGA	SEQ ID NO: 195
67	GATCCCACCCGATGGTTTACT	SEQ ID NO: 77	AGTAAACCATCGGGTGGGATCCT	SEQ ID NO: 196
68	CCAAGAGTGAATAGGACAGCA	SEQ ID NO: 78	TGCTGTCCTATTCACTCTTGGCA	SEQ ID NO: 197
69	ATAGGACAGCAGCGATTTCTG	SEQ ID NO: 79	CAGAAATCGCTGCTGTCCTATTC	SEQ ID NO: 198
69’	ATAGGACAGCAGCGATTTCTT	SEQ ID NO: 80	AAGAAATCGCTGCTGTCCTATTC	SEQ ID NO: 199
70	CACCAAATAAGCGCTTGCAAC	SEQ ID NO: 81	GTTGCAAGCGCTTATTTGGTGTG	SEQ ID NO: 200
70’	CACCAAATAAGCGCTTGCAAT	SEQ ID NO: 82	ATTGCAAGCGCTTATTTGGTGTG	SEQ ID NO: 201
71	AAAGATGCACGGATGACGTCC	SEQ ID NO: 83	GGACGTCATCCGTGCATCTTTCT	SEQ ID NO: 202
72	GATACACATTAAGGTTGTGTA	SEQ ID NO: 84	TACACAACCTTAATGTGTATCCA	SEQ ID NO: 203
73	ATTGCCCAGTACACGCATTAA	SEQ ID NO: 85	TTAATGCGTGTACTGGGCAATCC	SEQ ID NO: 204
74	AAAGCTCTTTCTGGTTTCAGT	SEQ ID NO: 86	ACTGAAACCAGAAAGAGCTTTGC	SEQ ID NO: 205
75	TGAAGGAGGGGACATTACTAC	SEQ ID NO: 87	GTAGTAATGTCCCCTCCTTCAAA	SEQ ID NO: 206
76	CATTACTACGGTCTTCACACC	SEQ ID NO: 88	GGTGTGAAGACCGTAGTAATGTC	SEQ ID NO: 207
77	CACACCAAGCGCCAAGTACTG	SEQ ID NO: 89	CAGTACTTGGCGCTTGGTGTGAA	SEQ ID NO: 208
78	CTCTTCACTTTCACGGCGGAA	SEQ ID NO: 90	TTCCGCCGTGAAAGTGAAGAGTA	SEQ ID NO: 209
79	TCACTTTCACGGCGGAATCAC	SEQ ID NO: 91	GTGATTCCGCCGTGAAAGTGAAG	SEQ ID NO: 210
80	TCCCACCCGATGGTTTACTTG	SEQ ID NO: 92	CAAGTAAACCATCGGGTGGGATC	SEQ ID NO: 211
81	AGAGTGAATAGGACAGCAGCG	SEQ ID NO: 93	CGCTGCTGTCCTATTCACTCTTG	SEQ ID NO: 212
82	AGTGAATAGGACAGCAGCGAT	SEQ ID NO: 94	ATCGCTGCTGTCCTATTCACTCT	SEQ ID NO: 213
84	AATAGGACAGCAGCGATTTCT	SEQ ID NO: 95	AGAAATCGCTGCTGTCCTATTCA	SEQ ID NO: 214
83	GCATAAACTATAACAGCTCAG	SEQ ID NO: 96	CTGAGCTGTTATAGTTTATGCCC	SEQ ID NO: 215

85	GGAAGAAAAGCACCCTTATTA	SEQ ID NO: 97	TAATAAGGGTGCTTTTCTTCCTC	SEQ ID NO: 216
86	GAAGAAAAGCACCCTTATTAA	SEQ ID NO: 98	TTAATAAGGGTGCTTTTCTTCCT	SEQ ID NO: 217
87	TTTTTCACGTACGCCACAAGG	SEQ ID NO: 99	CCTTGTGGCGTACGTGAAAAGT	SEQ ID NO: 218
88	TTTTCACGTACGCCACAAGGC	SEQ ID NO: 100	GCCTTGTGGCGTACGTGAAAAG	SEQ ID NO: 219
89	AGAAAAGCACCCTTATTAAGA	SEQ ID NO: 101	TCTTAATAAGGGTGCTTTTCTTC	SEQ ID NO: 220
90	ATTGCAGCAAGTAAGCCAACA	SEQ ID NO: 102	TGTTGGCTTACTTGCTGCAATTC	SEQ ID NO: 221
91	TCTCCTTAAAACATCTGAGAG	SEQ ID NO: 103	CTCTCAGATGTTTTAAGGAGACA	SEQ ID NO: 222
92	GACTCAGTTGTTGAAGGACAC	SEQ ID NO: 104	GTGTCCTTCAACAACTGAGTCAC	SEQ ID NO: 223
93	TTGAAGGAGGGGACATTACTA	SEQ ID NO: 105	TAGTAATGTCCCCTCCTTCAAAG	SEQ ID NO: 224
94	ACTCTTCACTTTCACGGCGGA	SEQ ID NO: 106	TCCGCCGTGAAAGTGAAGAGTAA	SEQ ID NO: 225
95	GGACAGCAGCGATTTCTGGGT	SEQ ID NO: 107	ACCCAGAAATCGCTGCTGTCCTA	SEQ ID NO: 226
96	CATAAACTATAACAGCTCAGT	SEQ ID NO: 108	ACTGAGCTGTTATAGTTTATGCC	SEQ ID NO: 227
97	AACTATAACAGCTCAGTTGCC	SEQ ID NO: 109	GGCAACTGAGCTGTTATAGTTTA	SEQ ID NO: 228
98	AAGAAAAGCACCCTTATTAAG	SEQ ID NO: 110	CTTAATAAGGGTGCTTTTCTTCC	SEQ ID NO: 229
99	AGCAAGTAAGCCAACAAGGTC	SEQ ID NO: 111	GACCTTGTTGGCTTACTTGCTGC	SEQ ID NO: 230
100	GCAAGTAAGCCAACAAGGTCT	SEQ ID NO: 112	AGACCTTGTTGGCTTACTTGCTG	SEQ ID NO: 231
RB2	GGGTATTCTTTCAAGCAAT	SEQ ID NO: 113	ATTGCTTGAAAGAATACCCAG	SEQ ID NO: 232
RB4	GCTCAAGAATGCCAAGAAA	SEQ ID NO: 114	TTTCTTGGCATTCTTGAGCAC	SEQ ID NO: 233
RB6	GAATCTCAAAGAAATCTTT	SEQ ID NO: 115	AAAGATTTCTTTGAGATTCTT	SEQ ID NO: 234
RB7	ATTTCTGGGTATTCTTTCA	SEQ ID NO: 116	TGAAAGAATACCCAGAAATCG	SEQ ID NO: 235
TJ632	AAGCAATGCTCACACCAAA	SEQ ID NO: 117	TTTGGTGTGAGCATTGCTTGA	SEQ ID NO: 236

TJ645	CTTGCAACAAAGACATTTA	SEQ ID NO: 118	TAAATGTCTTTGTTGCAAGCG	SEQ ID NO: 237
TJ772	GGATATTGTTGCTGCAAAA	SEQ ID NO: 119	TTTTGCAGCAACAATATCCAG	SEQ ID NO: 238
ISIS-416858	-	-	ACGGCATTGGTGCACAGTTT	SEQ ID NO: 239
qPCR primers				
Primer name	Upstream primer sequence	Upstream primer sequence ID	Downstream primer sequence	Downstream primer sequence ID
Human-GAPDH	GATTTGGTCGTATTGGGCGC	SEQ ID NO: 240	TTCCCGTTCTCAGCCTTGAC	SEQ ID NO: 241
Human-FXI	TCACTTTCACGGCGGAATCA	SEQ ID NO: 242	TCCGTGCATCTTTCTTGGCA	SEQ ID NO: 243
Reference sequence: FXI mRNA sequence				
NM_000128.4	gaccatttcctacacaagctcattcagaggaggatgaagaccattttggaggaagaaaagcacccttattaagaattgcagcaagtaagccaacaaggtcttttcaggatgattttcttatatcaagtggtacatttcattttatttacttcagtttctggtgaatgtgtgactcagttgttgaaggacacctgctttgaaggaggggacattactacggtcttcacaccaagcgccaagtactgccaggtagtctgcacttaccacccaagatgtttactcttcactttcacggcggaatccaccatctgaggatcccacccgatggtttacttgtgtcctgaaagacagtgttacagaaacactgccaagagtgaataggacagcagcgatttctgggtattctttcaagcaatgctcacaccaaataagcgcttgcaacaaagacatttatgtggacctagacatgaagggcataaactataacagctcagttgccaagagtgctcaagaatgccaagaaagatgcacggatgacgtccactgccacttttcacgtacgccacaaggcagtttcccagcctggagcatcgtaacatttgtctactgaagcacacccaaacagggacaccaaccagaataacgaagctcgataaagtggtgtctggattttcactgaaatcctgtgcactttctaatctggcttgtattagggacattttccctaatacggtgtttgcagacagcaacatcgacagtgtcatggctcccgatgcttttgtctgtggccgaatctgcactcatcatcccggttgcttgttttttaccttcttttcccaggaatggcccaaagaatctcaaagaaatctttgtctccttaaaacatctgagagtggattgcccagtacacgcattaaaaagagcaaagctctttctggtttcagtctacaaagctgcaggcacagcatcccagtgttctgccattcttcattttaccatgacactgatttcttgggagaagaactggatattgttgctgcaaaaagtcacgaggcctgccagaaactgtgcaccaatgccgtccgctgccagttttttacctataccccagcccaagcatcctgcaacgaagggaagggcaagtgttacttaaagctttcttcaaacggatctccaactaaaatacttcacgggagaggaggcatctctggatacacattaaggttgtgtaaaatggataatgagtgtaccaccaaaatcaagcccaggatcgttggaggaactgcgtctgttcgtggtgagtggccgtggcaggtgaccctgcacacaacctcacccactcagagacacctgtgtggaggctccatca	SEQ ID NO: 244

	ttggaaaccagtggatattaacagccgctcactgtttctatggggtagagtcacctaag attttgcgtgtctacagtggcattttaaatcaatctgaaataaaagaggacacatctttctt tggggttcaagaaataataatccatgatcagtataaaatggcagaaagcgggtatgat attgccttgttgaaactggaaaccacagtgaattacacagattctcaacgacccatatgc ctgccttccaaaggagatagaaatgtaatatacactgattgctgggtgactggatggg ggtacagaaaactaagagacaaaatacaaaatactctccagaaagccaagataccctt agtgaccaacgaagagtgccagaagagatacagaggacataaaataacccataaaga tgatctgtgccggctacagggaaggagggaaggacgcttgcaagggagattcggg aggccctctgtcctgcaaacacaatgaggtctggcatctggtaggcatcacgagctgg ggcgaaggctgtgctcaaagggagcggccaggtgtttacaccaacgtggtcgagta cgtggactggattctggagaaaactcaagcagtgtgaatgggttcccaggggccatt ggagtccctgaaggacccaggatttgctgggagagggtgttgagttcactgtgccag catgcttcctccacagtaacacgctgaaggggcttggtgtttgtaagaaaatgctagaa gaaaacaaactgtcacaagttgttatgtccaaaactcccgttctatgatcgttgtagtttgt ttgagcattcagtctctttgtttttgatcacgcttctatggagtccaagaattaccataagg caatatttctgaagattactatataggcagatatagcagaaaataaccaagtagtggca gtggggatcaggcagaagaactggtaaaagaagccaccataaatagatttgttcgat gaaagatgaaaactggaagaaaggagaacaaagacagtcttcaccattttgcagga atctacactctgcctatgtgaacacatttcttttgtaaagaaagaaattgattgcatttaat ggcagattttcagaatagtcaggaattcttgtcatttccattttaaaatatatattaaaaaa aatcagttcgagtagacacgagctaagagtgaatgtgaagataacagaatttctgtgt ggaagaggattacaagcagcaatttacctggaagtgataccttaggggcaatcttgaa gatacactttcctgaaaaatgatttgtgatggattgtatatttatttaaaatatcttgggag gggaggctgatggagatagggagcatgctcaaacctccctaagacaagctgctgct gtgactatgggctcccaaagagctagatcgtatatttatttgacaaaaatcaccatagac tgcatccatactacagagaaaaaacaattagggcgcaaatggatagttacagtaaagt cttcagcaagcagctgcctgtattctaagcactgggattttctgtttcgtgcaaatatttat ctcattattgttgtgatctagttcaataacctagaatttgaattgtcaccacatagctttcaa tctgtgccaacaactatacaattcatcaagtgtg	

CLAIMS

1. A nucleic acid molecule for inhibiting the expression of factor 11 (FXI) gene in cells via RNAi, comprising or consisting of a sense sequence and an antisense sequence complementary to each other, with the antisense sequence comprising a polynucleotide sequence complementary to FXI gene mRNA, wherein

when the antisense sequence hybridizes with the FXI gene mRNA at the maximum complementarity, the base pairs formed by complementary pairing between the antisense sequence and the FXI gene mRNA sequence constitute complementary region 1;

when the antisense sequence hybridizes with the sense sequence at the maximum complementarity, the base pairs formed by complementary pairing between the antisense sequence and the sense sequence constitute complementary region 2;

the complementary region 1 and the complementary region 2 share at least 18, 19 or 20 identical base pairs,

the number of base pairs in the complementary region 1 is 18 to 30 bp, and the complementary region 1 comprises the following nucleotides in the FXI gene mRNA sequence:

nucleotides at positions 418 to 437, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 1141 to 1160, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 363 to 382, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 372 to 391, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 411 to 430, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 421 to 440, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 422 to 441, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 425 to 444, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 465 to 484, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 470 to 489, or 18 or 19 consecutive nucleotides therein,

nucleotides at positions 471 to 490, or 18 or 19 consecutive nucleotides therein, or

nucleotides at positions 518 to 537, or 18 or 19 consecutive nucleotides therein; and

the nucleotide positions of the FXI gene mRNA sequence are numbered in accordance with the corresponding nucleotides in the reference sequence of SEQ ID NO: 244.

2. The nucleic acid molecule according to claim 1, wherein the number of base pairs in the complementary region 1 is 20, 21, 22 or 23 bp.

3. The nucleic acid molecule according to claim 1 or 2, wherein the number of base pairs in the complementary region 2 is 21 bp.

4. The nucleic acid molecule according to any one of claims 1 to 3, wherein the complementary region 1 and the complementary region 2 have 20 or 21 identical base pairs.

5. The nucleic acid molecule according to any one of claims 1 to 4, wherein the length of the sense sequence is 21 nt and the length of the antisense sequence is 23 nt.

6. The nucleic acid molecule according to any one of claims 1 to 5, wherein the sense sequence and/or antisense sequence further comprises 1 to 2 overhang nucleotides outside the complementary region 2.

7. The nucleic acid molecule according to claim 6, wherein there are 2 overhang nucleotides located in the antisense sequence and adjacent to the 5' end of the complementary region 2, and the sense sequence comprises no overhang nucleotides.

8. The nucleic acid molecule according to any one of claims 1 to 7, wherein the antisense sequence comprises only 0, 1 or 2 nucleotides outside the complementary region 1, and the sense sequence comprises only 0, 1 or 2 nucleotides outside the complementary region 2.

9. The nucleic acid molecule according to any one of claims 1 to 8, wherein the 3' end of the complementary region 2 is an AU base pair.

10. The nucleic acid molecule according to any one of claims 1 to 9, wherein the FXI gene is a human FXI gene or a cynomolgus monkey FXI gene.

11. The nucleic acid molecule according to claim 10, wherein the mRNA comprises the polynucleotide sequence set forth in SEQ ID NO: 244.

12. The nucleic acid molecule according to any one of claims 1 to 11, wherein the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 153, 177, 152, 122, 123, 126, 129, 130, 131, 136, 137, 138, 143, 149, 150, 200, 201 or 213.

13. The nucleic acid molecule according to any one of claims 1 to 12, wherein:

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 34, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 153;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 58, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 177; the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 33, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 152;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 3, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 122;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 4, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 123;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 7, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 126;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 10, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 129;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 11, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 130;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 12, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 131;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 17, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 136;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 18, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 137;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 19, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 138;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 24, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 143;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 30, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 149;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 31, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 150;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 81, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 200;

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 82, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 201; or

the sense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 94, and the antisense sequence comprises or is the polynucleotide sequence set forth in SEQ ID NO: 213.

14. The nucleic acid molecule according to any one of claims 1 to 13, wherein the nucleic acid molecule is a dsRNA or shRNA.

15. The nucleic acid molecule according to claim 14, wherein the nucleic acid molecule is an siRNA.

16. The nucleic acid molecule according to any one of claims 1 to 15, wherein one or more nucleotides are chemically modified.

17. The nucleic acid molecule according to any one of claims 1 to 16, wherein the chemical modification is one or more selected from the group consisting of:

locked nucleic acid modification, unlocked nucleic acid modification, 2'-methoxyethyl modification, 2'-O-methyl modification, 2'-O-allyl modification, 2'-C-alkyl modification, 2'-C-allyl modification, 2'-fluoro modification, 2'-deoxy modification, phosphorothioate bond modification, 2'-amino modification, morpholino modification, phosphoramidate modification, methylphosphonate modification, tetrahydropyran modification, 1,5-anhydrohexitol modification, 5'-vinyl phosphate modification, and cyclohexenyl modification;

preferably, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 12 of the sense strand comprise 2'-fluoro modifications and/or the nucleotides at positions 2, 14 and 16 of the antisense strand comprise 2'-fluoro modifications, and further preferably, the remaining nucleotides comprise 2'-O-methyl modifications;

preferably, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 11 of the sense strand comprise 2'-fluoro modifications and/or the nucleotides at positions 2, 14 and 16 of the antisense strand comprise 2'-fluoro modifications; and further preferably, the remaining nucleotides comprise 2'-O-methyl modifications;

preferably, in the nucleic acid molecule, the nucleotides at positions 7, 9 to 11 of the sense strand comprise 2'-fluoro modifications, and the nucleotides at positions 2, 6, 14 and 16 of the

antisense strand comprise 2'-fluoro modifications; and further preferably, the remaining nucleotides comprise 2'-O-methyl modifications;

preferably, the nucleic acid molecule comprises a motif selected from the group consisting of:

(1) sense sequence: NmNmNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmNfNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmNmNm,

(2) sense sequence: NmNmNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmNfNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmNmNm, and

(3) sense sequence: NmNmNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmNfNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmNmNm,

wherein Nm represents a ribonucleotide modified with 2'-O-methyl, and Nf represents a ribonucleotide modified with 2'-fluoro;

further preferably, numbered from the 5' end of the sense strand, at least one of the following positions has a phosphorothioate modification: between the first and second nucleotides, between the second and third nucleotides, between the last and penultimate nucleotides, and between the penultimate and antepenultimate nucleotides; numbered from the 5' end of the antisense strand, at least one of the following positions has a phosphorothioate modification: between the first and second nucleotides, between the second and third nucleotides, between the last and penultimate nucleotides, and between the penultimate and antepenultimate nucleotides.

18. The nucleic acid molecule according to claim 17, wherein the nucleic acid molecule comprises a motif selected from the following group consisting of:

(1) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(2) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(3) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNm, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(4) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmsNmsNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(5) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmsNmsNm, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm; and

(6) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmsNmsNm, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(7) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(8) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms, antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(9) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNms, antisense sequence: NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

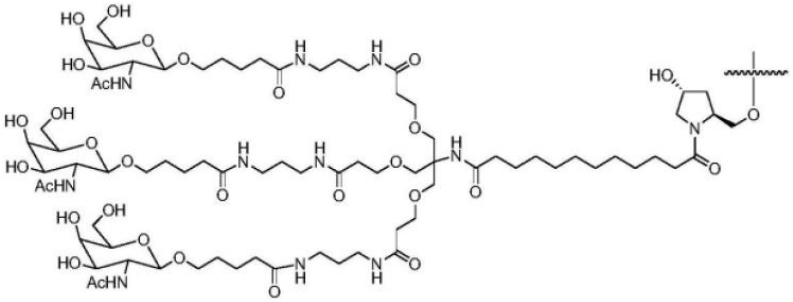
wherein Nm represents a ribonucleotide modified with 2'-O-methyl, Nf represents a ribonucleotide modified with 2'-fluoro, and s represents a phosphorothioate modification.

19. The nucleic acid molecule according to any one of claims 1 to 18, wherein the nucleic acid molecule is linked to at least one asialoglycoprotein receptor (ASGPR) ligand.

20. The nucleic acid molecule according to claim 19, wherein the ASGPR ligand is linked to the 5' end or 3' end of the sense sequence.

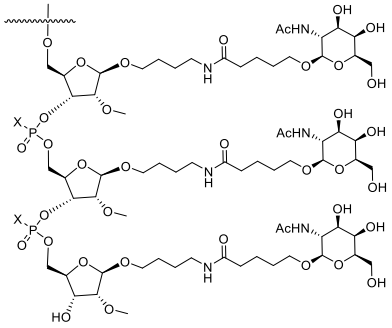
21. The nucleic acid molecule according to claim 19 or 20, wherein the ASGPR ligand is one or more GalNAc derivatives linked via a divalent or trivalent branched structure.

22. The nucleic acid molecule according to claim 21, wherein the GalNAc derivative is L96 or ligand 1, and the structure of the L96 is shown below:



(Formula I);

the structure of the ligand 1 is shown below:



(X=S^- or O^-) (Formula II);

wherein 〰 in each of formula I and formula II represents linkage to the 3' end of the sense sequence or antisense sequence of the nucleic acid molecule; preferably, the linkage is formed via a phosphodiester bond or a phosphorothioate diester bond to 3'-hydroxy;

further preferably, the nucleic acid molecule has any one of the following modification motifs:

(1) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96; antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(2) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNms-ligand 1; antisense sequence: NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(3) sense sequence: NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNm-L96; antisense

sequence:
NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(4) sense sequence:
NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1;
antisense sequence:
NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(5) sense sequence:
NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNm-L96; antisense sequence:
NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

(6) sense sequence:
NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1;
antisense sequence:
NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm.

23. The nucleic acid molecule according to any one of claims 1 to 22, wherein the nucleic acid molecule comprises a phosphate or phosphate mimetic modification at the 5' end of the antisense strand; preferably, the 5' phosphate mimetic modification is 5'-VP;

further preferably, the nucleic acid molecule has any one of the following modification motifs:

GN-E20VP:
sense sequence:
NmsNmsNmNmNmNmNfNmNfNfNfNfNmNmNmNmNmNmNmNmNmNms-ligand 1;
antisense sequence: vp-
NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

GN-E04VP:
sense sequence:
NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1;
antisense sequence: vp-
NmsNfsNmNmNmNfNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm;

GN-E05VP:
sense sequence:
NmsNmsNmNmNmNmNfNmNfNfNfNmNmNmNmNmNmNmNmNmNmNms-ligand 1;
antisense sequence: vp-
NmsNfsNmNmNmNmNmNmNmNmNmNmNmNfNmNfNmNmNmNmNmsNmsNm.

24. A second nucleic acid molecule, wherein the second nucleic acid molecule can be transcribed into a dsRNA or shRNA precursor in a cell, and the dsRNA or shRNA is the nucleic acid molecule according to any one of claims 1 to 23.

25. A viral particle, comprising the polynucleotide sequence of the second nucleic acid molecule according to claim 24.

26. A cell, comprising the polynucleotide sequence of the second nucleic acid molecule according to claim 24.

27. Use of the nucleic acid molecule according to any one of claims 1 to 23 or a salt thereof for inhibiting the expression of factor 11.

28. Use of the nucleic acid molecule according to any one of claims 1 to 23 or a salt thereof in the manufacture of a drug for inhibiting the expression of FXI in a subject.

29. A pharmaceutical composition, comprising the nucleic acid molecule according to any one of claims 1 to 23 or a salt thereof, the second nucleic acid molecule according to claim 24, the viral particle according to claim 25, or the cell according to claim 26.

30. Use of the nucleic acid molecule according to any one of claims 1 to 23 or a salt thereof, the second nucleic acid molecule according to claim 24, the viral particle according to claim 25, the cell according to claim 26, or the pharmaceutical composition according to claim 29 in the manufacture of a drug for preventing or treating thromboembolic complications or coagulation disorders in a subject; preferably, the thromboembolic complications are one or more selected from the group consisting of deep vein thrombosis, pulmonary embolism, myocardial infarction and stroke.

31. A method for preventing or treating thromboembolic complications or coagulation disorders in a subject, wherein the thromboembolic complications are preferably one or more selected from the group consisting of deep vein thrombosis, pulmonary embolism, myocardial infarction and stroke, and the method comprises administering an effective amount of the nucleic acid molecule according to any one of claims 1 to 23 or a salt thereof, the second nucleic acid molecule according to claim 24, the viral particle according to claim 25, the cell according to claim 26, or the pharmaceutical composition according to claim 29 to a subject in need thereof.