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(54) **GENETICALLY MODIFIED GENES AND CELLS, AND METHODS OF USING SAME FOR SILENCING VIRUS GENE EXPRESSION**

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Related U.S. Application Data

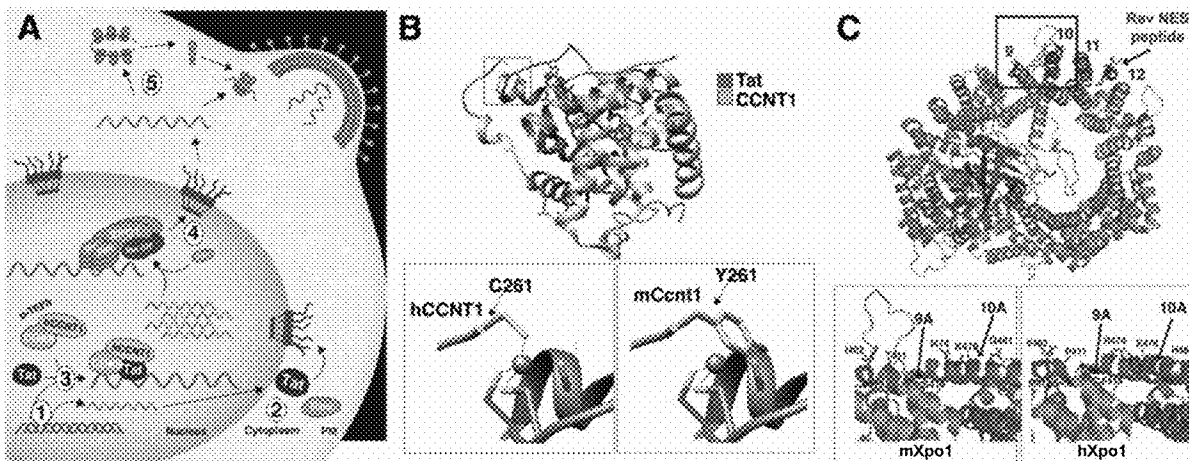
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(57) **ABSTRACT**
Genetically modified CCNT1 and XPO1 genes encoding proteins that inhibit virus infection in cells. The genetically modified CCNT1 gene encodes a protein with a C261Y substitution with respect to the human CCNT1 protein. The genetically modified XPO1 gene encodes a protein with P411T, M412V, and/or F414S substitutions with respect to the human XPO1 protein. The genetically modified CCNT1 and XPO1 genes can be introduced in cells. The cells comprising the genetically modified CCNT1 and XPO1 genes can be introduced in a subject with a virus infection to treat the infection.

Specification includes a Sequence Listing.



CLUSTAL O(1.2.4) multiple sequence alignment

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hCCNT1*   MEGERKNNNKRWYFTREQLENS PSRRFGVDPDKELSYRQQAANLLQDMGQRLNVSQLTIN 60
hCCNT1    MEGERKNNNKRWYFTREQLENS PSRRFGVDPDKELSYRQQAANLLQDMGQRLNVSQLTIN 60
mCCNT1    MEGERKNNNKRWYFTREQLENS PSRRFGVDS DKELSYRQQAANLLQDMGQRLNVSQLTIN 60
*****

hCCNT1*   TAIVYMHRFYMIQSFTQFPGNSVAPAALFLAAKVEEQPKKLEHVIKVAHTCLHPQESLPD 120
hCCNT1    TAIVYMHRFYMIQSFTQFPGNSVAPAALFLAAKVEEQPKKLEHVIKVAHTCLHPQESLPD 120
mCCNT1    TAIVYMHRFYMIQSFTQFHRYSMAPAALFLAAKVEEQPKKLEHVIKVAHTCLHPQESLPD 120
*****

hCCNT1*   TRSEAYLQQVQDLVILESII LQTLGFELTIDHPHTHVVKCTQLVRASKDLAQT SYFMATN 180
hCCNT1    TRSEAYLQQVQDLVILESII LQTLGFELTIDHPHTHVVKCTQLVRASKDLAQT SYFMATN 180
mCCNT1    TRSEAYLQQVQDLVILESII LQTLGFELTIDHPHTHVVKCTQLVRASKDLAQT SYFMATN 180
*****

hCCNT1*   SLHLTTFSLQYTPPVVACVCIHLACKWSNWEI PVSTDGKHWWEYVDATVTLELLDELTHE 240
hCCNT1    SLHLTTFSLQYTPPVVACVCIHLACKWSNWEI PVSTDGKHWWEYVDATVTLELLDELTHE 240
mCCNT1    SLHLTTFSLQYTPPVVACVCIHLACKWSNWEI PVSTDGKHWWEYVDATVTLELLDELTHE 240
*****

hCCNT1*   FLQILEKTPNRLKRIWNWRAYEAAKKT KADDRGTDEKTSEQTILNMI SQSSSDTTIAGLM 300
hCCNT1    FLQILEKTPNRLKRIWNWRACEAAKKT KADDRGTDEKTSEQTILNMI SQSSSDTTIAGLM 300
mCCNT1    FLQILEKTPSRLKRI RNWRAYQAAMKTKPDRGADENTSEQTILNMI SQTSSDTTIAGLM 300
*****

hCCNT1*   SMSTSTTS AVPSLPVSEESSSNLTSVEMLPGKRWLSSQPSFKLEPTQGHRTSEN LALTGV 360
hCCNT1    SMSTSTTS AVPSLPVSEESSSNLTSVEMLPGKRWLSSQPSFKLEPTQGHRTSEN LALTGV 360
mCCNT1    SMSTASTS AVPSLPSEESSSNLTSVDM LQGERWLSSQPPFKLEAAQGHRTSESLALIGV 360
*****

hCCNT1*   DHSLPQDGSNAFISQKQNSKSVPSAKVSLKEYRAKHAEELAAQKRQLENMEANVKSQYAY 420
hCCNT1    DHSLPQDGSNAFISQKQNSKSVPSAKVSLKEYRAKHAEELAAQKRQLENMEANVKSQYAY 420
mCCNT1    DHSLQQDGS S AFGSQASKSVPSAKVSLKEYRAKHAEELAAQKRQLENMEANVKSQYAY 420
*****

hCCNT1*   AAQNLLSHHDSHSSVILKMPIEGSENPERPFLEKADKTALKMRI PVAGGDKAASSKPEEI 480
hCCNT1    AAQNLLSHHDSHSSVILKMPIEGSENPERPFLEKADKTALKMRI PVAGGDKAASSKPEEI 480
mCCNT1    AAQNLLS~HDSHSSVILKMPIESS ENPERPFLDKADKSALKMRLPVASGDKAVSSKPEEI 479
*****

hCCNT1*   KMRIKVHAAADKHNSVEDSVTKSREHKEKHKTHPSNHHHHHHHSHKHSLSQLPVGTKGNK 540
hCCNT1    KMRIKVHAAADKHNSVEDSVTKSREHKEKHKTHPSNHHHHHHHSHKHSLSQLPVGTKGNK 540
mCCNT1    KMRIKVHSAGDKHNSIEDSVTKSREHKEKQ RTHPSNHHHHHHHSHRHSLSQLPAGPVSK 539
*****

hCCNT1*   RPGDPKHSSQTSNLAKHTYLSLSSSFSSSSSTRKRG PSEETGGAVFDHPAKIAKSTKSSSL 600
hCCNT1    RPGDPKHSSQTSNLAKHTYLSLSSSFSSSSSTRKRG PSEETGGAVFDHPAKIAKSTKSSSL 600
mCCNT1    RPSDPKHSSQTSNLAKHTYLSLSSSFSSSSSTRKRG PPEETGAAVFDHPAKIAKSTK~SSL 598
*****

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FIG. 1A

CLUSTAL O(1.2.4) multiple sequence alignment

hXPO1*	MPAIMTMLADHAARQLLDFSQKLDINLLDNVVNCLYHGEGAQQRMAQEVLTHLKEHPDAW	60
hXPO1	MPAIMTMLADHAARQLLDFSQKLDINLLDNVVNCLYHGEGAQQRMAQEVLTHLKEHPDAW	60
mXPO1	MPAIMTMLADHAARQLLDFSQKLDINLLDNVVNCLYHGEGAQQRMAQEVLTHLKEHPDAW	60

hXPO1*	TRVDTILEFSQNMNTKYGLQILENVIKTRWKILPRNQCEGIKKYVVGLI IKTSSDPTCV	120
hXPO1	TRVDTILEFSQNMNTKYGLQILENVIKTRWKILPRNQCEGIKKYVVGLI IKTSSDPTCV	120
mXPO1	TRVDTILEFSQNMNTKYGLQILENVIKTRWKILPRNQCEGIKKYVVGLI IKTSSDPTCV	120

hXPO1*	EKEKVIYIGKLNMI LVQILKQEWPKHWPTFISDIVGASRTSESLCQNNMVIKLLSEEVFD	180
hXPO1	EKEKVIYIGKLNMI LVQILKQEWPKHWPTFISDIVGASRTSESLCQNNMVIKLLSEEVFD	180
mXPO1	EKEKVIYIGKLNMI LVQILKQEWPKHWPTFISDIVGASRTSESLCQNNMVIKLLSEEVFD	180

hXPO1*	FSSGQITQVKSKHLKDSMCNEFSQIFQLCQFVMENSQNAPLVHATLETLLRFLNWIPLGY	240
hXPO1	FSSGQITQVKSKHLKDSMCNEFSQIFQLCQFVMENSQNAPLVHATLETLLRFLNWIPLGY	240
mXPO1	FSSGQITQVKAKHLKDSMCNEFSQIFQLCQFVMENSQNAPLVHATLETLLRFLNWIPLGY	240
	***** ; *****	
hXPO1*	IFETKLISTLIYKFLNVPFRNVSLKCLTEIAGVSVSQYEEQFVTLFTLTMMQLKQMLPL	300
hXPO1	IFETKLISTLIYKFLNVPFRNVSLKCLTEIAGVSVSQYEEQFVTLFTLTMMQLKQMLPL	300
mXPO1	IFETKLISTLIYKFLNVPFRNVSLKCLTEIAGVSVSQYEEQFETLFTLTMMQLKQMLPL	300
	***** *****	
hXPO1*	NTNIRLAYSNGKDDEQNFIQNLSLFLCTFLKEHDQLIEKRLNLRRTLMEALHYMLLVSEV	360
hXPO1	NTNIRLAYSNGKDDEQNFIQNLSLFLCTFLKEHDQLIEKRLNLRRTLMEALHYMLLVSEV	360
mXPO1	NTNIRLAYSNGKDDEQNFIQNLSLFLCTFLKEHGQLEKRLNLRREALMEALHYMLLVSEV	360
	***** , ** , ***** , *****	
hXPO1*	EETEIFKICLEYWNHLAAELYRESPFSTASPLLSGSHFDVPPRRQLYLTVLSKVRLLM	420
hXPO1	EETEIFKICLEYWNHLAAELYRESPFSTASPLLSGSHFDVPPRRQLYLPMLFKVRLLM	420
mXPO1	EETEIFKICLEYWNHLAAELYRESPFSTASPLLSGSHFDI PRRQLYLTVLSKVRLLM	420
	***** , ***** ; *****	
hXPO1*	VSRMAKPEEVLVVENDQGEVVREFMKD TDSINLYKNMRETLVYLTHLDYVDTERIMTEKL	480
hXPO1	VSRMAKPEEVLVVENDQGEVVREFMKD TDSINLYKNMRETLVYLTHLDYVDTERIMTEKL	480
mXPO1	VSRMAKPEEVLVVENDQGEVVREFMKD TDSINLYKNMRETLVYLTHLDYVDTEIIMTKKL	480
	***** ** , **	
hXPO1*	HNQVNGTEWSWKNLNTLCWAIGSISGAMHEEDEKRFLVTVIKDLLGLCEQKRGKDNKAI I	540
hXPO1	HNQVNGTEWSWKNLNTLCWAIGSISGAMHEEDEKRFLVTVIKDLLGLCEQKRGKDNKAI I	540
mXPO1	QNQVNGTEWSWKNLNTLCWAIGSISGAMHEEDEKRFLVTVIKDLLGLCEQKRGKDNKAI I	540

hXPO1*	ASNIMYIVGQYPRFLRAHWKFLKTVVNKLF EFMHETHDGVQDMACDTFIKIAQKRRHFV	600
hXPO1	ASNIMYIVGQYPRFLRAHWKFLKTVVNKLF EFMHETHDGVQDMACDTFIKIAQKRRHFV	600
mXPO1	ASNIMYIVGQYPRFLRAHWKFLKTVVNKLF EFMHETHDGVQDMACDTFIKIAQKRRHFV	600

FIG. 2A

hXPO1* QVQVGEVMPFIDEILNNTIICDLQPQQVHTFYEA VGYMIGAQT DQTVQEHLEIKYMLL 660
 hXPO1 QVQVGEVMPFIDEILNNTIICDLQPQQVHTFYEA VGYMIGAQT DQTVQEHLEIKYMLL 660
 mXPO1 QVQVGEVMPFIDEILNNTIICDLQPQQVHTFYEA VGYMIGAQT DQTVQEHLEIKYMLL 660

hXPO1* PNQVWDSIIQQATKNVDILKDPETVKQLG SILKTNVRACKAVGH PPFVIQLGRIYLDMLNV 720
 hXPO1 PNQVWDSIIQQATKNVDILKDPETVKQLG SILKTNVRACKAVGH PPFVIQLGRIYLDMLNV 720
 mXPO1 PNQVWDSIIQQATKNVDILKDPETVKQLG SILKTNVRACKAVGH PPFVIQLGRIYLDMLNV 720

hXPO1* YKCLSENISAAIQANGEMVTKQPLIRSMRTVKRE TLKLI SGWVSRSN DPQMV AENFV PPL 780
 hXPO1 YKCLSENISAAIQANGEMVTKQPLIRSMRTVKRE TLKLI SGWVSRSN DPQMV AENFV PPL 780
 mXPO1 YKCLSENISAAIQANGEMVTKQPLIRSMRTVKRE TLKLI SGWVSRSN DPQMV AENFV PPL 780

hXPO1* LDAVLIDYQRNVPAAREPEVLSTMAIIVNKLGGHITAEIPQIFDAVFECTLNMINKDFEE 840
 hXPO1 LDAVLIDYQRNVPAAREPEVLSTMAIIVNKLGGHITAEIPQIFDAVFECTLNMINKDFEE 840
 mXPO1 LDAVLIDYQRNVPAAREPEVLSTMAIIVNKLGGHITAEIPQIFDAVFECTLNMINKDFEE 840

hXPO1* YPEHRTNFFLLLQAVNSHCFPAFLAIPPTQFKLV LDSIIWAFKHTMRNVADTGLQILFTL 900
 hXPO1 YPEHRTNFFLLLQAVNSHCFPAFLAIPPTQFKLV LDSIIWAFKHTMRNVADTGLQILFTL 900
 mXPO1 YPEHRTNFFLLLQAVNSHCFPAFLAIPPTQFKLV LDSIIWAFKHTMRNVADTGLQILFTL 900

hXPO1* LQNVAQEEAAAQSFYQTYFC DILQHIFSVVTDTSHTAGLTMHASILAYMFNLVEEGKIST 960
 hXPO1 LQNVAQEEAAAQSFYQTYFC DILQHIFSVVTDTSHTAGLTMHASILAYMFNLVEEGKIST 960
 mXPO1 LQNVAQEEAAAQSFYQTYFC DILQHIFSVVTDTSHTAGLTMHASILAYMFNLVEEGKIST 960

hXPO1* SLNPGNPVNNQIFLQEYVANLLKSAFPHLQDAQVKLFVTGLFSLNQDIPAFKEHLRDFLV 1020
 hXPO1 SLNPGNPVNNQIFLQEYVANLLKSAFPHLQDAQVKLFVTGLFSLNQDIPAFKEHLRDFLV 1020
 mXPO1 PLNPGNPVNNQMFIQDYVANLLKSAFPHLQDAQVKLFVTGLFSLNQDIPAFKEHLRDFLV 1020
 *****;*:*****

hXPO1* QIKEFAGEDTSDLFLEEREIALRQADEEKHKRQMSVPGIFNPHEIPEEMCD 1071 (SEQ ID
 hXPO1 QIKEFAGEDTSDLFLEEREIALRQADEEKHKRQMSVPGIFNPHEIPEEMCD 1071 (SEQ ID
 mXPO1 QIKEFAGEDTSDLFLEERETALRQAQEEKHKLQMSVPGILNPHEIPEEMCD 1071 (SEQ ID
 *****;*****;*****

hXPO1* NO:7)
 hXPO1 NO:9)
 mXPO1 NO:12)

FIG. 2B

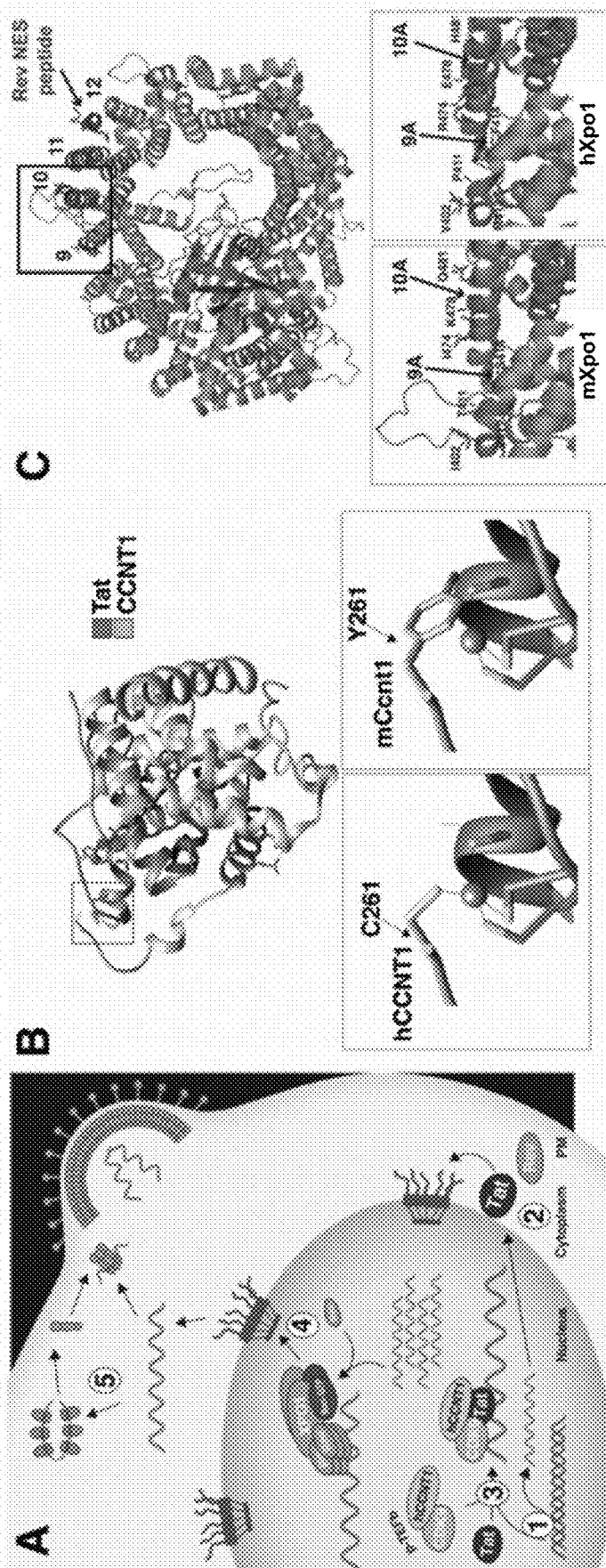


FIG. 3

human CCNT1

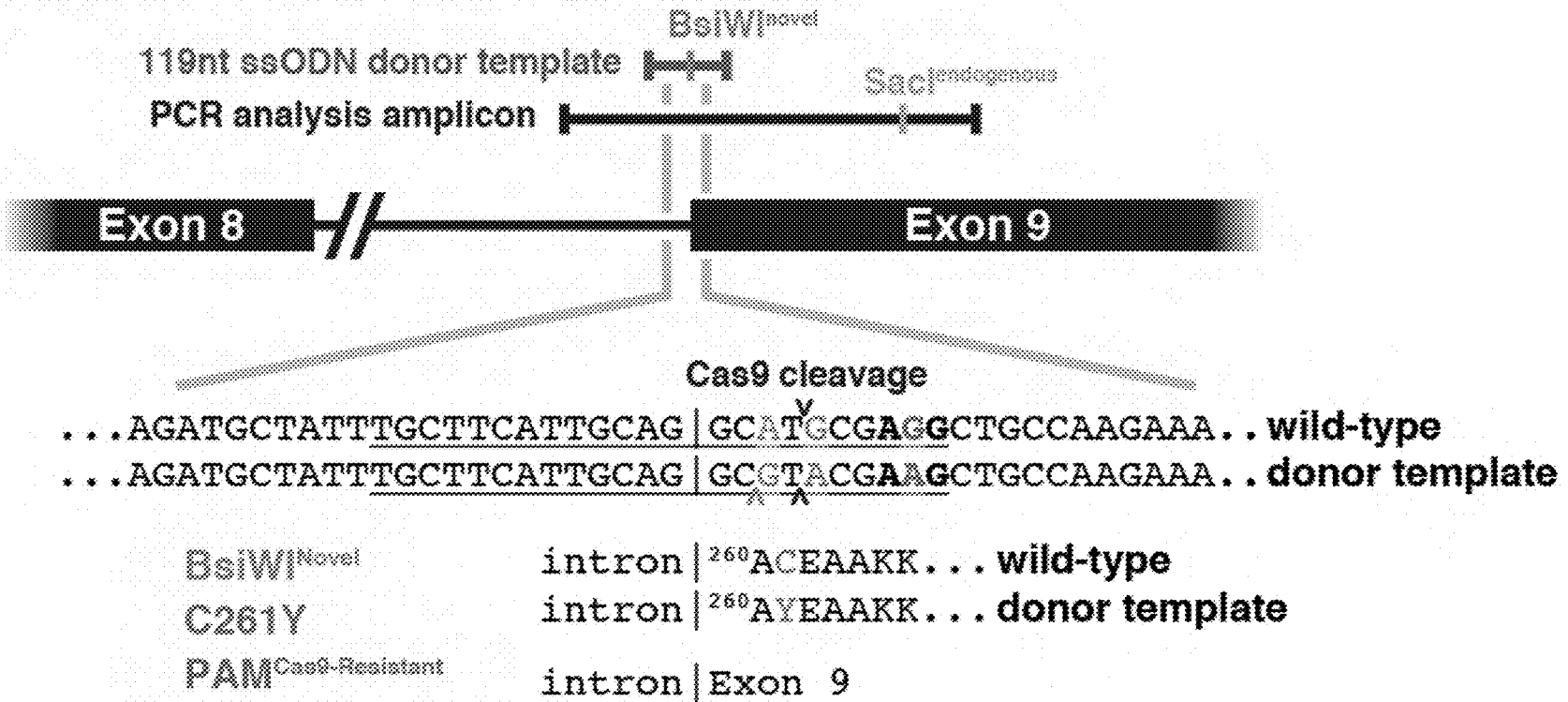


FIG. 4

human XPO1 (V2, Single gRNA)

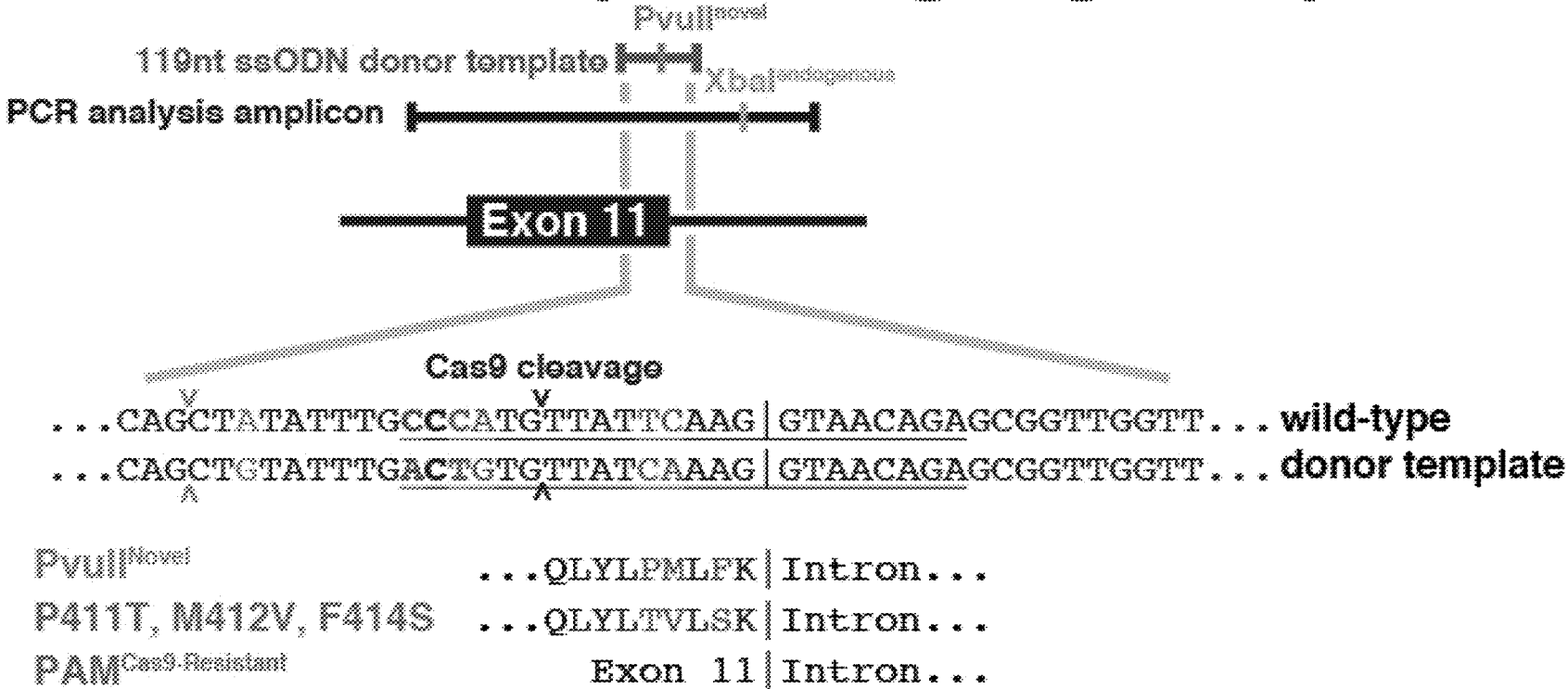


FIG. 5A

human XPO1 (V1, Double gRNA)

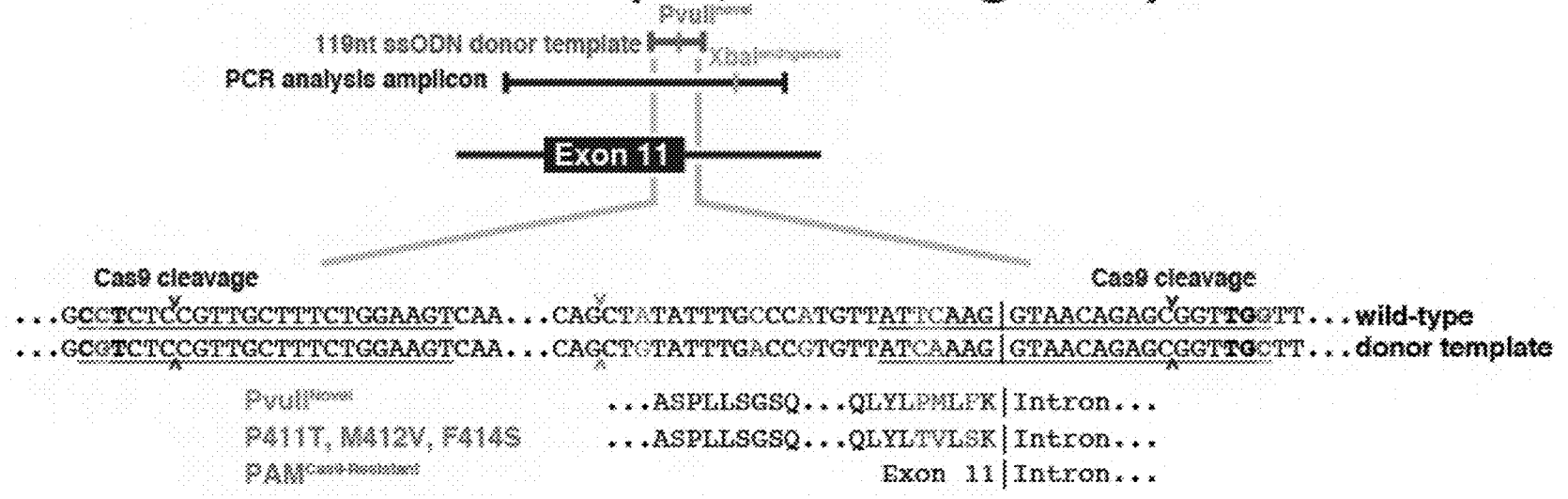


FIG. 5B

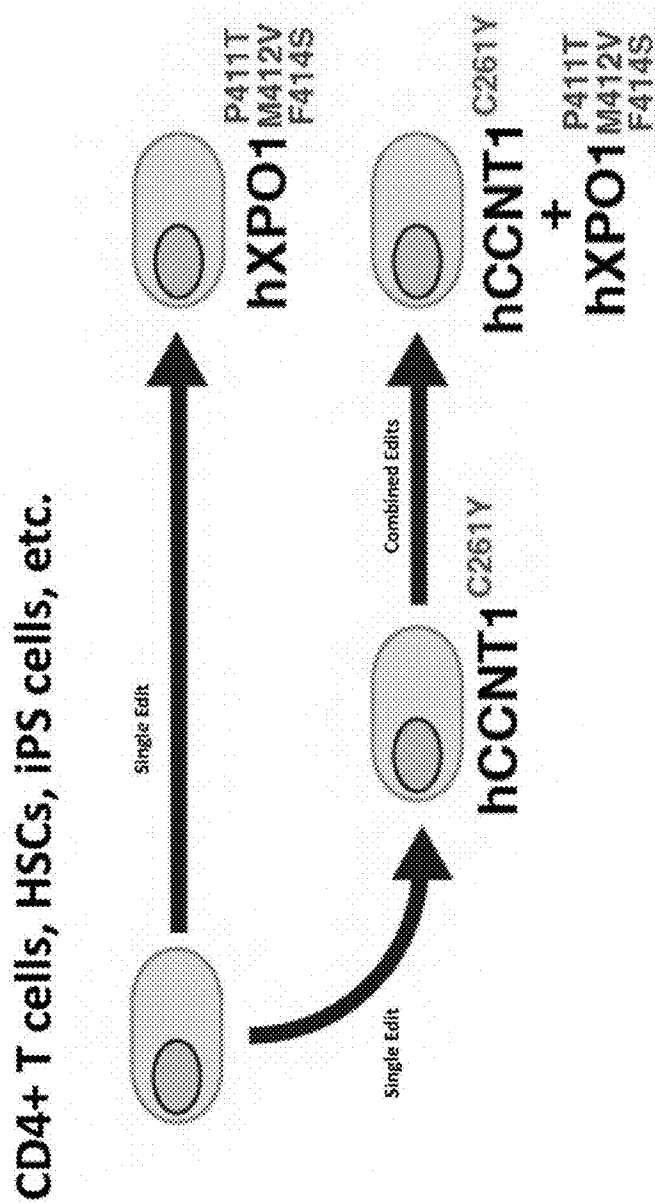


FIG. 6

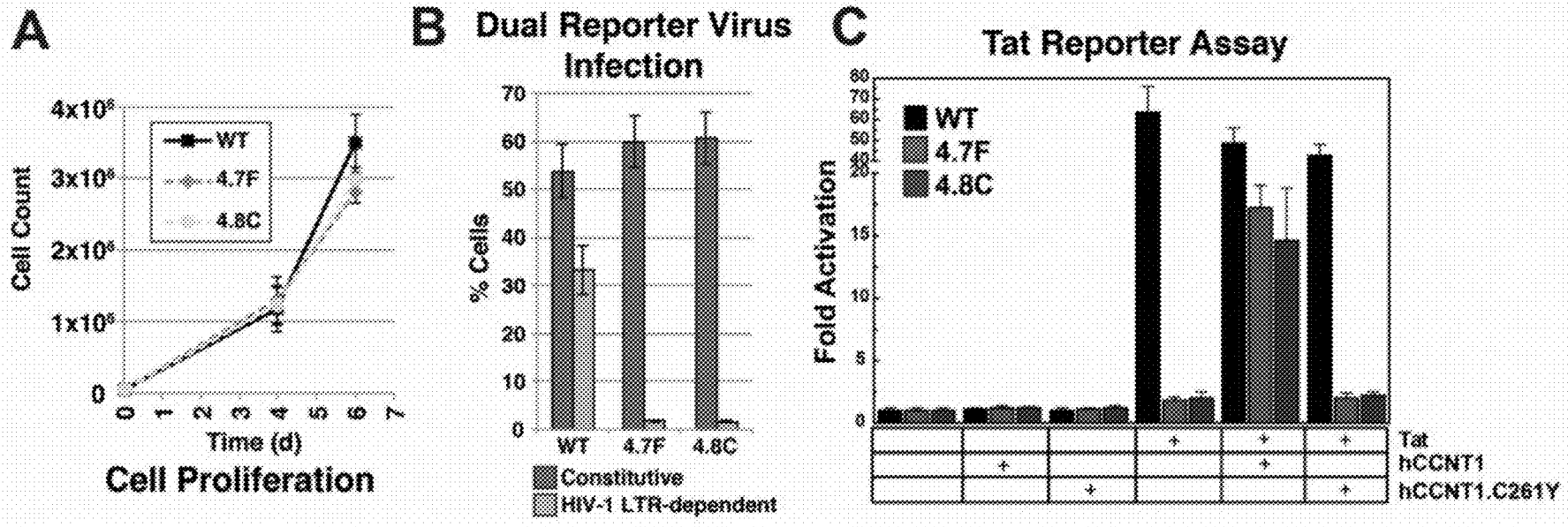


FIG. 7

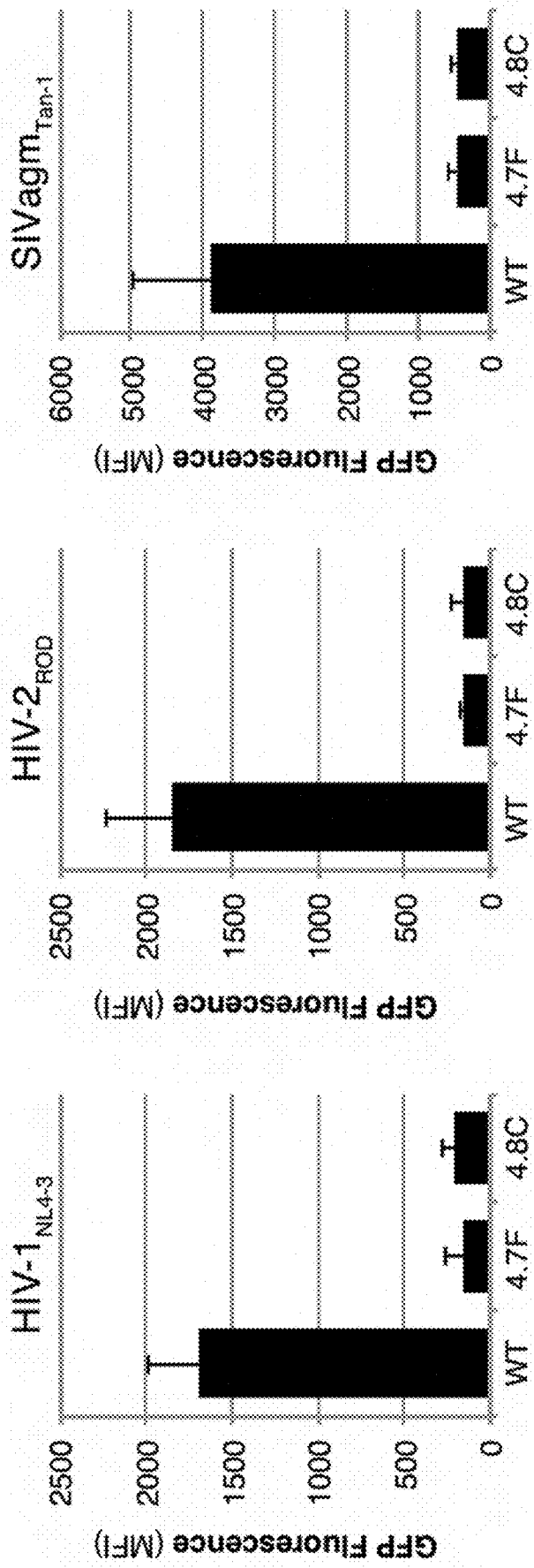


FIG. 8

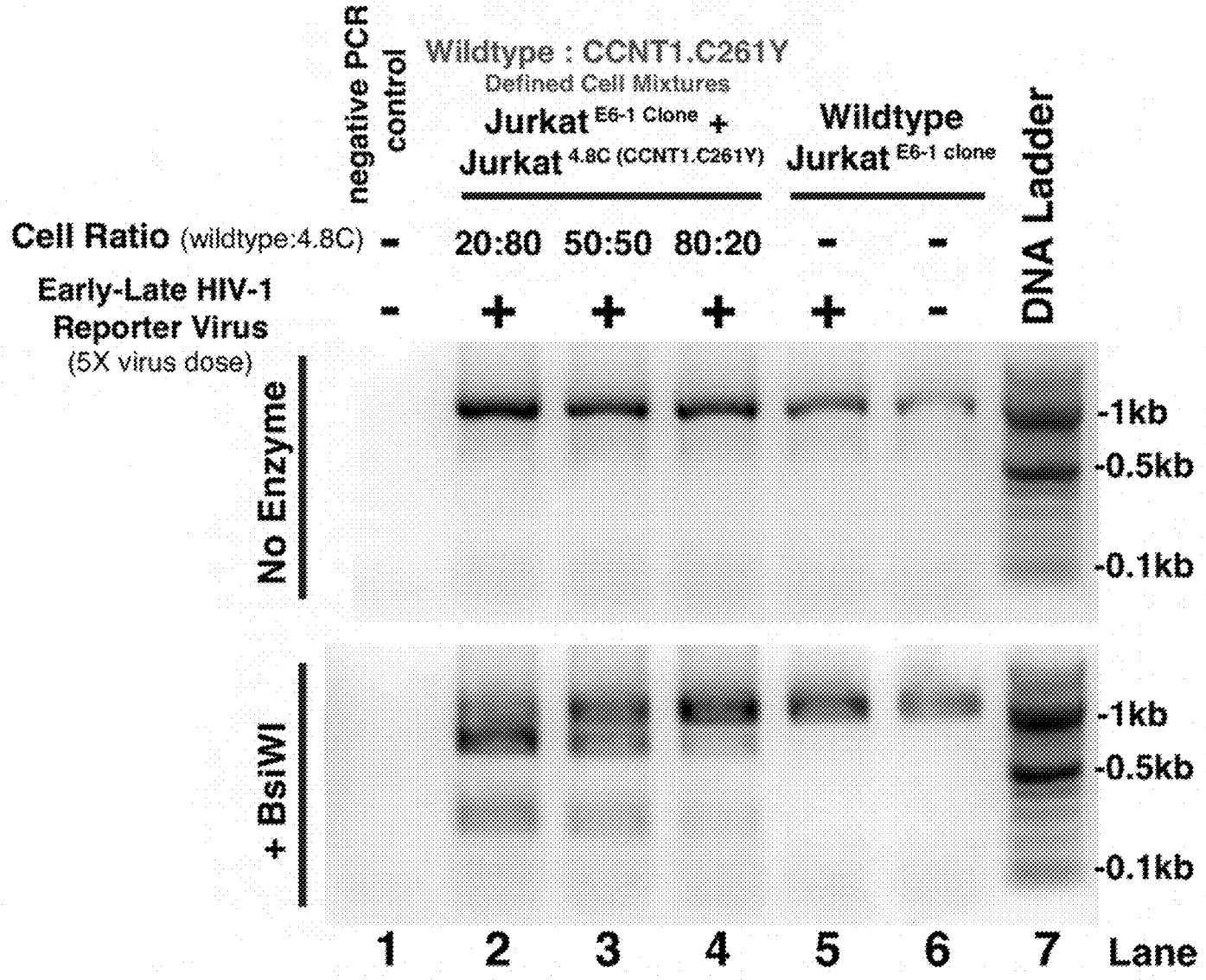


FIG. 9A

Early-Late HIV-1 Reporter Virus Infection Gating Legend

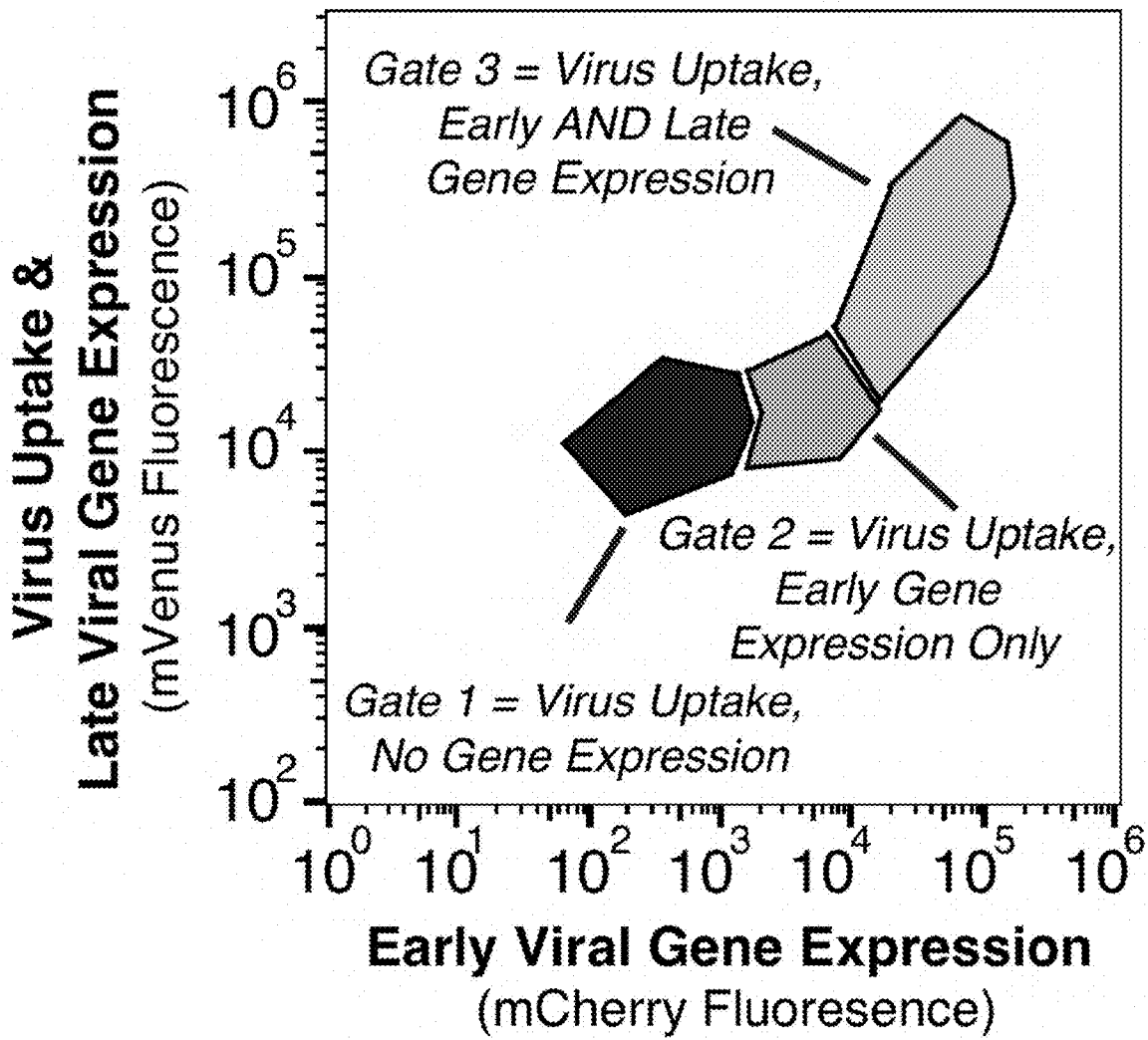


FIG. 9B

Jurkat Cell Mixture Ratio (wildtype : CCNT1-C261Y)

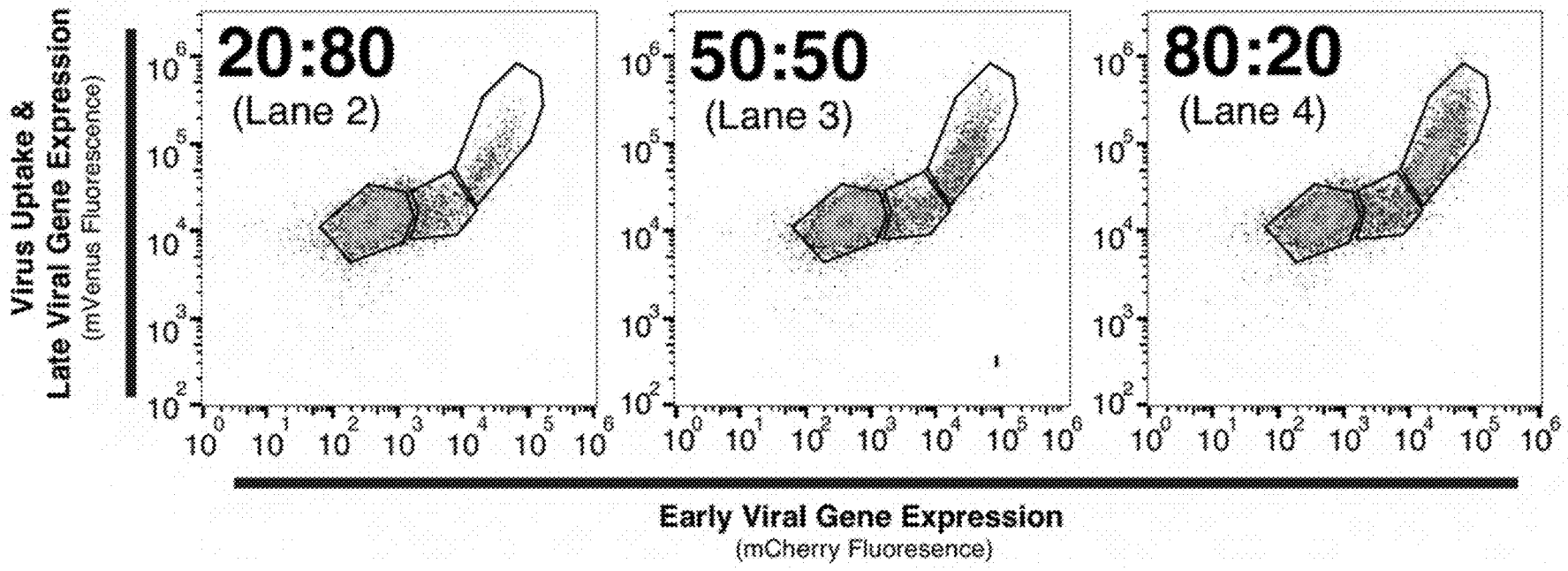


FIG. 9C

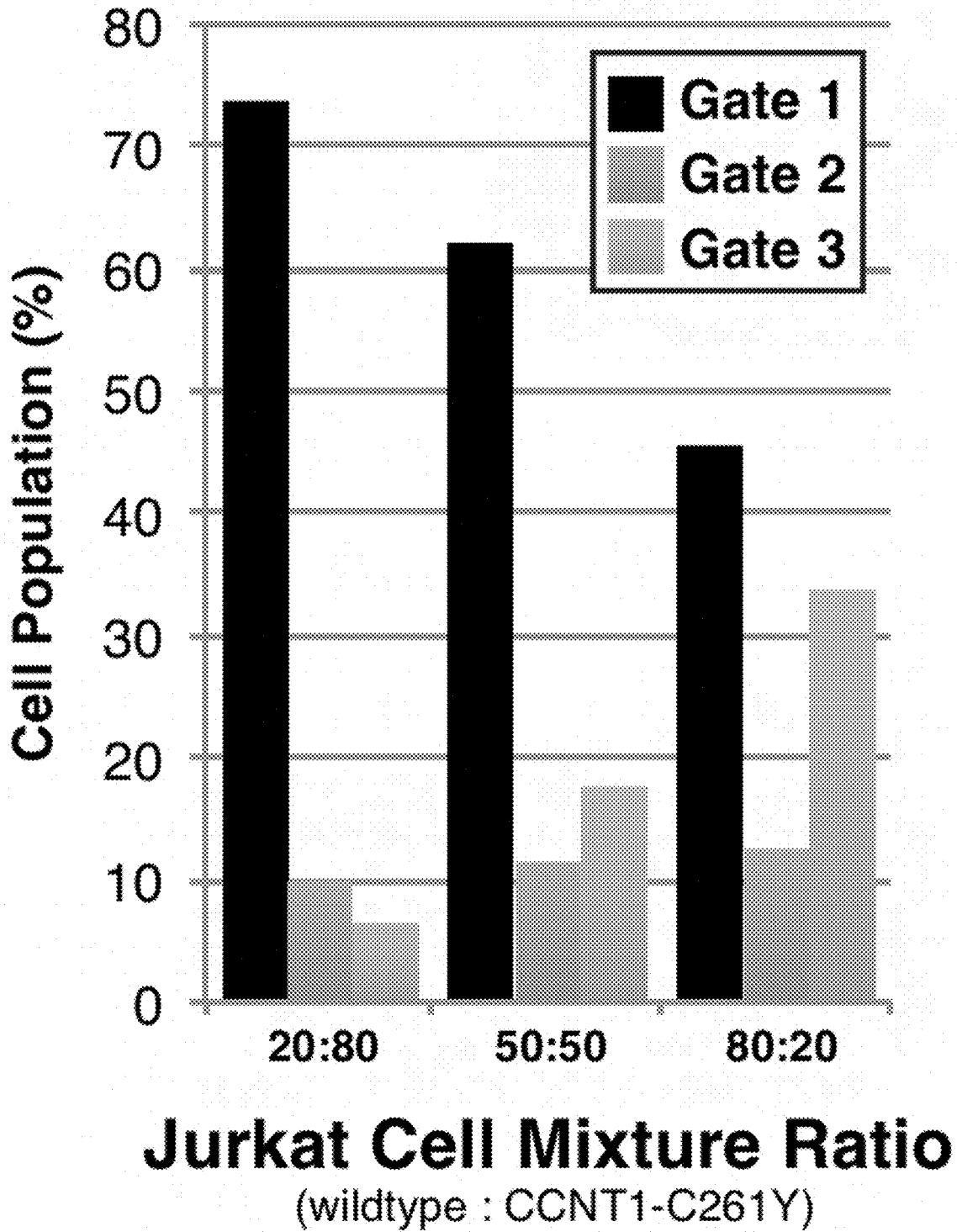


FIG. 9D

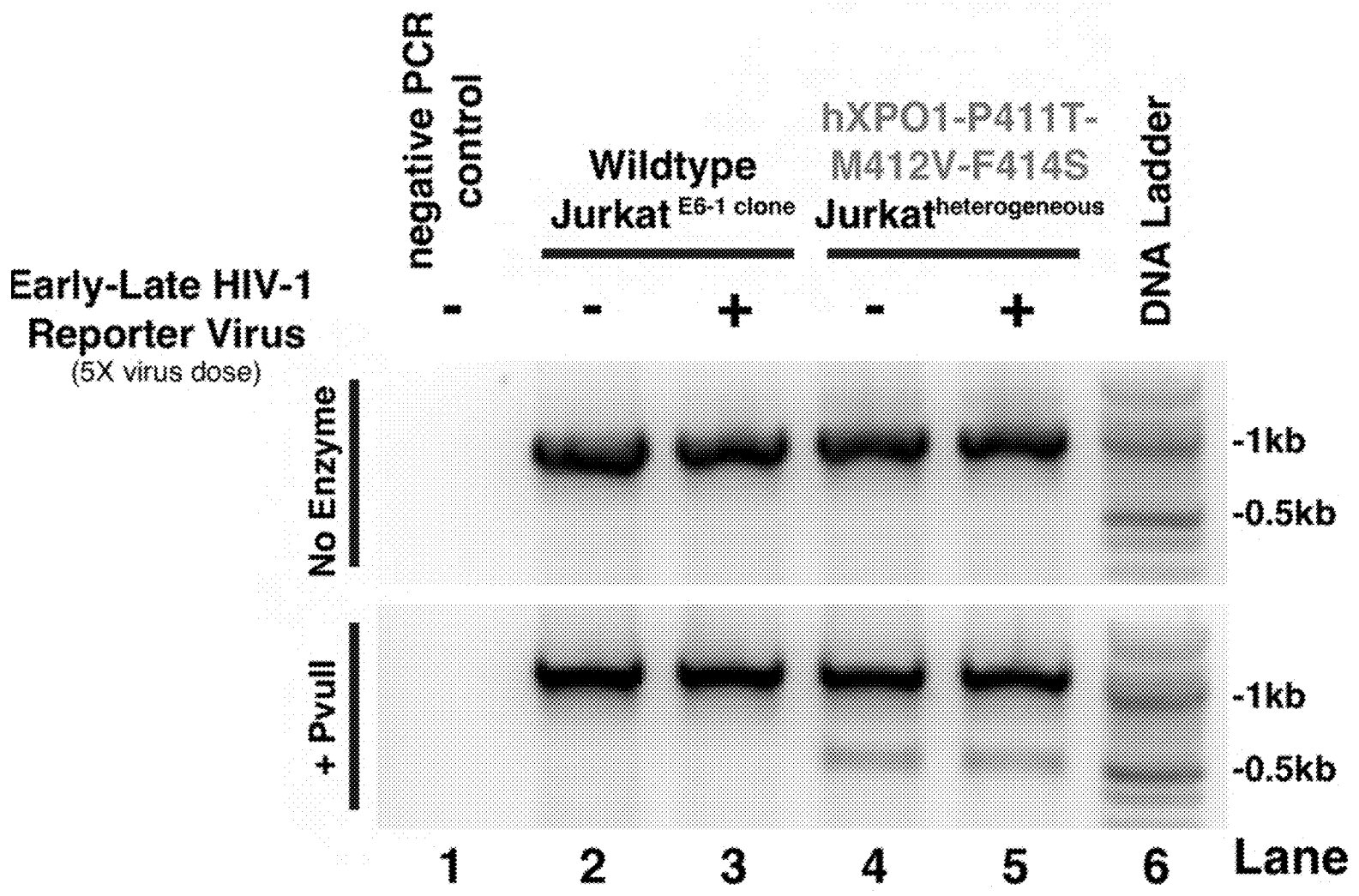


FIG. 10A

Early-Late HIV-1 Reporter Virus Infection Gating Legend

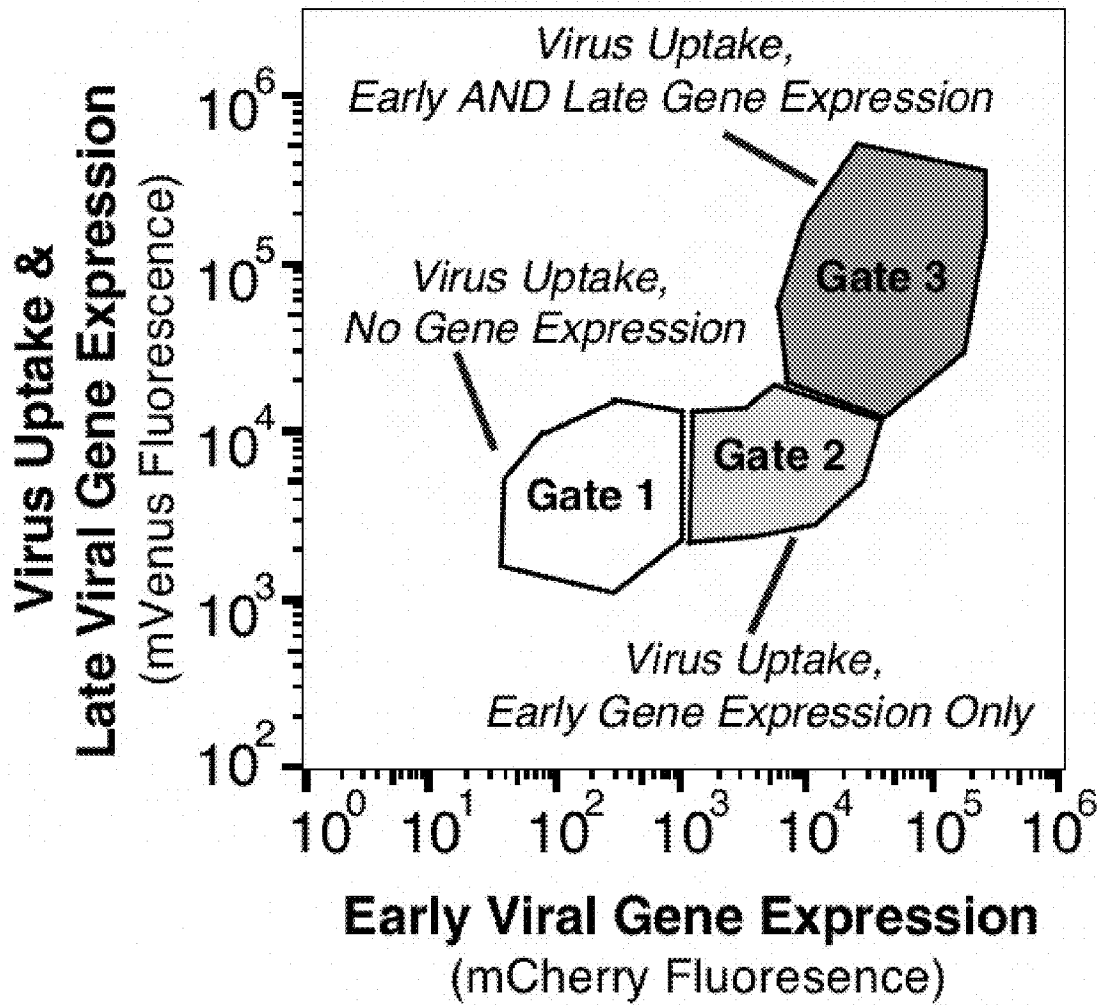


FIG. 10B

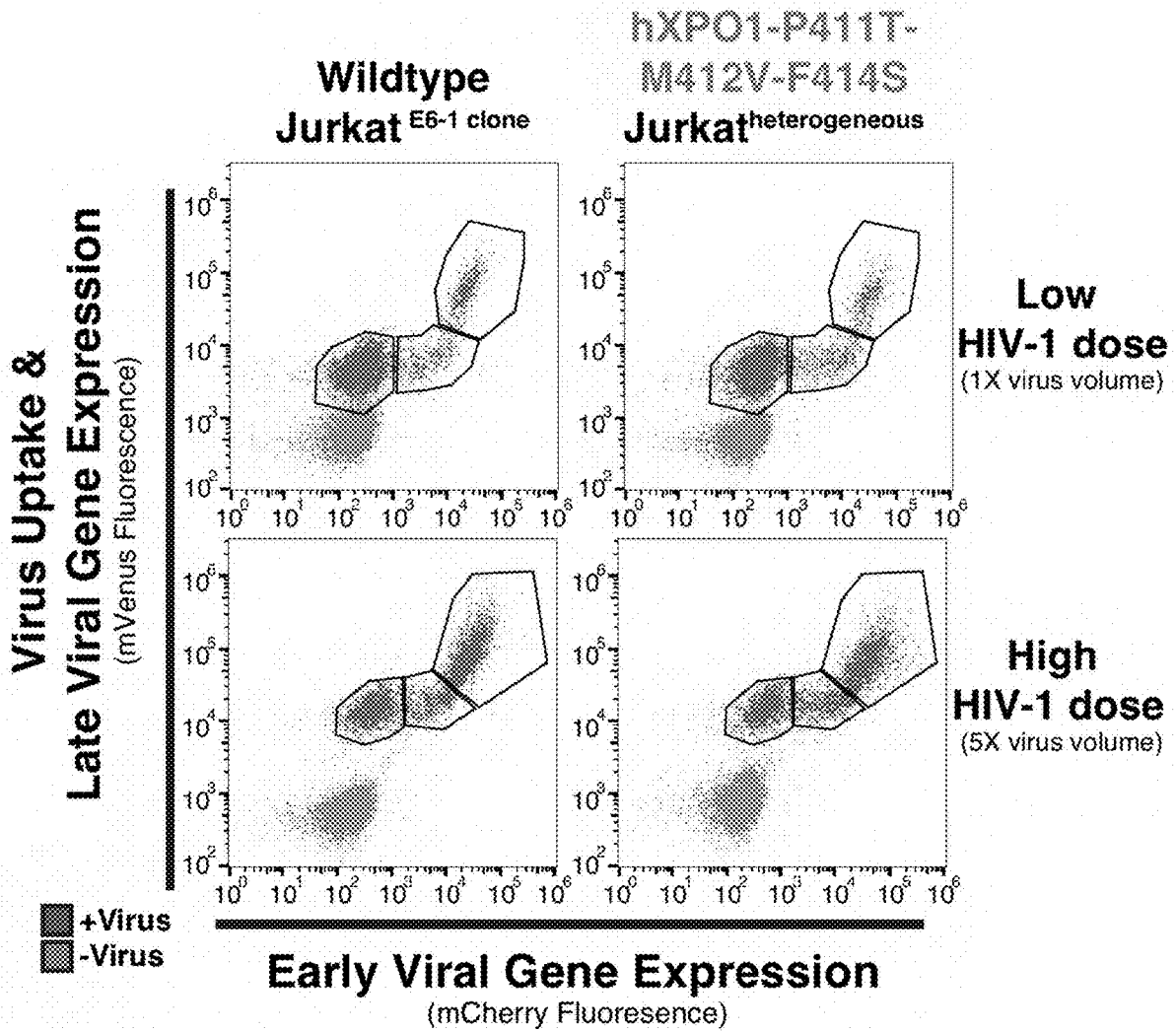


FIG. 10C

Early vs. Late Viral Gene Expression (% Cell Population Per Gate)

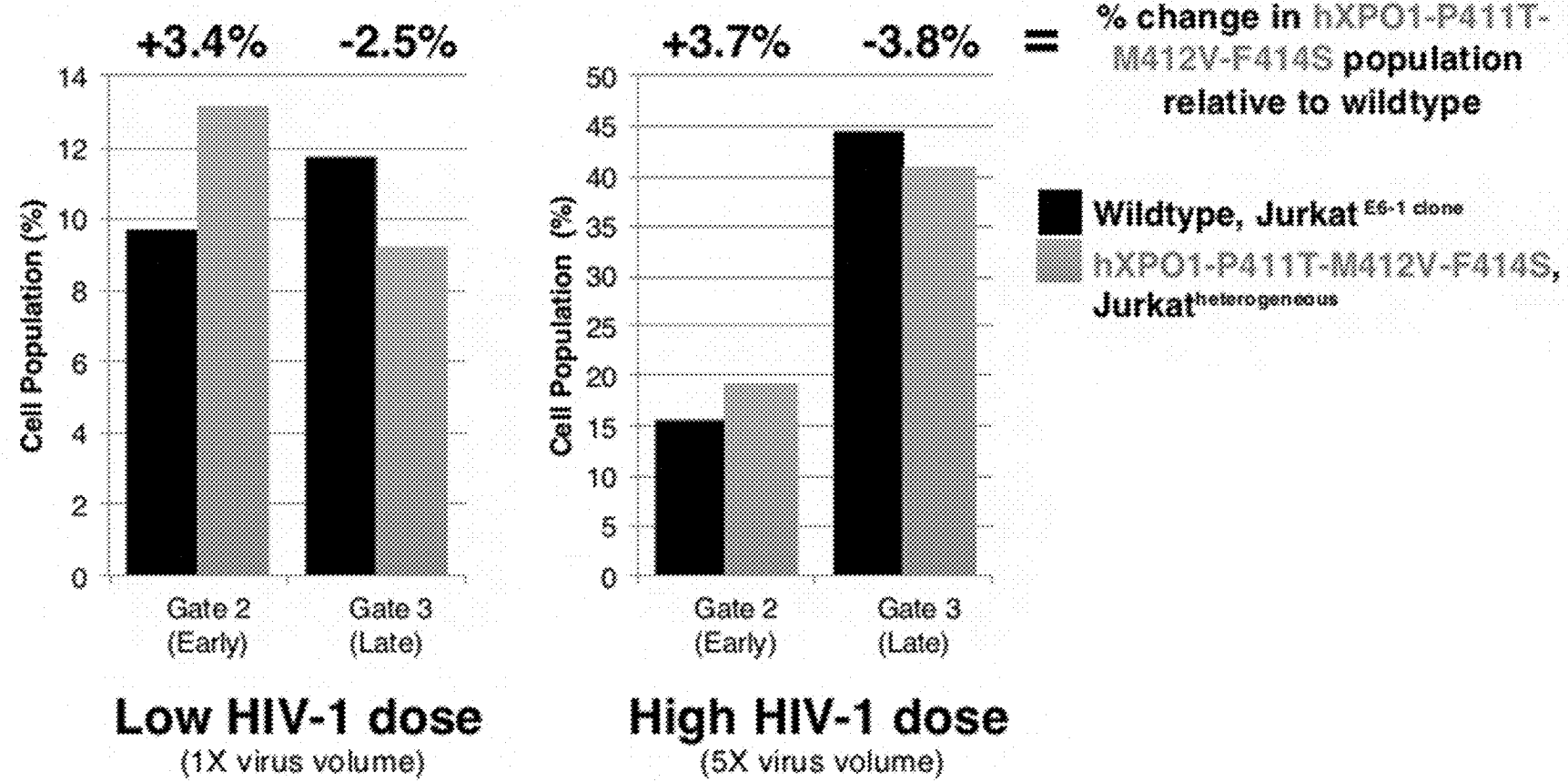


FIG. 10D

**GENETICALLY MODIFIED GENES AND
CELLS, AND METHODS OF USING SAME
FOR SILENCING VIRUS GENE
EXPRESSION**

STATEMENT REGARDING FEDERALLY
SPONSORED RESEARCH

[0001] This invention was made with government support under A1110221 and A1143800 awarded by the National Institutes of Health. The government has certain rights in the invention.

SEQUENCE LISTING

[0002] The instant application contains a Sequence Listing which has been submitted in ASCII format via EFS-Web and is hereby incorporated by reference in its entirety. The ASCII copy, created on Sep. 4, 2019, is named USPTO-190905-Nonpro_Patent_App-P180284US02-SEQ_LIST.txt and is 65,821 bytes in size.

FIELD OF THE INVENTION

[0003] Methods and tools for autologous T cell transplant to introduce amino acid changes in CCNT1 and/or XPO1 that permanently suppress HIV-1 gene expression in patient cells, and other purposes.

BACKGROUND

[0004] The human immunodeficiency virus type 1 (HIV-1) is the causative agent of the acquired immunodeficiency syndromes (AIDS). HIV-1 infects more than 1 million people in the United States and more than 35 million worldwide, causing ~1 million deaths annually. While combined antiretroviral therapy (cART) can reduce viral load and slow progression AIDS, there is no vaccine or cure for life-long, persistent infection.

[0005] Highly active anti-retroviral therapy (HAART) was a major breakthrough in the treatment of human immunodeficiency virus (HIV) infection as it can effectively reduce viral load and support regeneration of cellular immunity, thereby considerably prolonging survival of HIV-infected patients. However, despite the effective suppression of virus replication, HIV persists, integrated into the host genome, and rebounds as soon as treatment is interrupted or drug-resistant virus emerges. Even with the most effective antiviral drug combinations, it has not been possible to “cure” HIV infection, and life-long antiviral therapy is required to prevent progression of immunodeficiency. This vital long-term treatment is expensive and limited by drug toxicity and viral resistance, and the number of patients for whom HAART fails is increasing. Moreover, even prolonged periods of successful HAART have failed to restore HIV-specific immune responses. Thus, novel therapeutic approaches are still urgently required.

[0006] Several therapeutic strategies involving the transfer of antiviral genes have been developed for HIV-1 infection. In clinical trials, T cells and hematopoietic stem cells have been targeted. See Trickett et al. 2002 (Trickett A E, Kwan Y L, Cameron B, Dwyer J M. Ex vivo expansion of functional T lymphocytes from HIV-infected individuals. *J Immunol Methods*. 2002 Apr. 1; 262(1-2):71-83), Lieberman et al. 1997 (Lieberman J, Skolnik P R, Parkerson G R 3rd, Fabry J A, Landry B, Bethel J, Kagan J. Safety of autologous, ex vivo-expanded human immunodeficiency virus (HIV)-spe-

cific cytotoxic T-lymphocyte infusion in HIV-infected patients. *Blood*. 1997 Sep. 15; 90(6):2196-206), van Lunzen et al. 2007 (van Lunzen J, Glaunsinger T, Stahmer I, von Baehr V, Baum C, Schilz A, Kuehlcke K, Naundorf S, Martinius H, Hermann F, Giroglou T, Newrzela S, Muller I, Brauer F, Brandenburg G, Alexandrov A, von Laer D. Transfer of autologous gene-modified T cells in HIV-infected patients with advanced immunodeficiency and drug-resistant virus. *Mol Ther*. 2007 May; 15(5):1024-33), Tebas et al. 2014 (Tebas P, Stein D, Tang W W, Frank I, Wang S Q, Lee G, Spratt S K, Surosky R T, Giedlin M A, Nichol G, Holmes M C, Gregory P D, Ando D G, Kalos M, Collman R G, Binder-Scholl G, Plesa G, Hwang W T, Levine B L, June C H. Gene editing of CCR5 in autologous CD4 T cells of persons infected with HIV. *N Engl J Med*. 2014 Mar. 6; 370(10):901-10), von Laer et al. 2006, (von Laer, D, Hasselmann, S and Hasselmann, K (2006). Gene therapy for HIV infection: what does it need to make it work? *J Gene Med* 8: 658-667), and Levine et al. 2006 (Levine, B L, Humeau, L M, Boyer J, Macgregor, R R, Rebello, T, Lu, X et al. (2006). Additional strategies are needed.

SUMMARY OF THE INVENTION

[0007] The present invention builds on observations that rodents and their cells are refractory to HIV-1 infection, due to structural differences in the rodent proteins that render them incompatible for complexing with HIV-1 regulatory proteins. The human CCNT1 (hCCNT1) transcription factor is recruited by the HIV-1 Tat protein to activate robust viral mRNA transcription in human cells, but mouse CCNT1 (mCCNT1) interacts poorly with Tat due to a single amino acid difference: a tyrosine at mCCNT1 position 261 that is a cysteine in hCCNT1. The human XPO1 (hXPO1, aka CRM1) nuclear export receptor is recruited by the viral Rev protein to intron-retaining viral mRNAs in human cells to activate mRNAs nuclear export, but murine XPO1 (mXPO1) interacts poorly with Rev/RNA complexes, a defect that maps to a cluster of mXPO1 species-specific amino acids: threonine-411, valine-412, and serine-414.

[0008] The present invention relates to tools and methods for permanently suppressing HIV-1 gene expression in cells through surgical editing of cellular genes to express CCNT1 and/or XPO1 with refractory residues. One method is based on autologous cell transplant, in which cells are removed from a patient, modified (edited) in vitro, and returned to the patient, where they can outcompete the infected cells. The method can be performed with patient-derived primary CD4+ T cells, precursors thereof, hematopoietic stem or progenitor cells, or other types of cells. The native hCCNT1 and/or hXPO1 can be edited to express hCCNT1 with a C261Y substitution and/or hXPO1 with P411T, M412V, and/or F414S substitutions, respectively. These edits render the cells resistant to HIV-1 gene expression in vivo, thus providing an HIV-1 cure-targeted strategy. Editing multiple target genes in the same cells or cell lines (e.g., both hCCNT1 and hXPO1) inactivates multiple essential virus-host interactions with even greater suppression of viral replication and reduces the chances of developing resistance.

[0009] The genes can be edited using gene editing tools such as CRISPR/Cas9, TALENs, etc., thereby generating permanent, homozygous edits that are heritable and can be introduced in any cell type, including hematopoietic stem cells or their HIV-susceptible progeny cells (including but

not limited to CD4+ T cells, macrophages, dendritic cells, and astrocytes). The edits have little to no discernible impact on the natural cellular functions of these proteins outside the context of infection. Thus, the strategy yields low to no cytotoxicity.

[0010] The mutations proposed also offer resistance to other viruses (e.g., in humans, primates, and other animals or mammals), since those host factors are relevant for other lentiviral pathogens including HIV-2 and simian immunodeficiency viruses (SIVs) commonly used for AIDS vaccine research in NHP models; and also deltaretroviruses such as human T lymphotropic virus type 1 (HTLV-1).

[0011] To date, there are no approved therapies for targeting HIV-1 following integration of the HIV-1 provirus (i.e., “after” infection). The present approach abolishes viral gene expression, virus particle production, and productive spread among cells, tissues, or people. Moreover, targeting species-specific protein features of CCNT1 or XPO1 is superior (i.e., less toxic) than other antiviral approaches that target virus-host interfaces because these particular protein features inhibit viral replication but do not play other essential roles in cell signaling.

[0012] The objects and advantages of the invention will appear more fully from the following detailed description of the preferred embodiment of the invention made in conjunction with the accompanying drawings.

BRIEF DESCRIPTION OF THE DRAWINGS

[0013] The patent or application file contains at least one drawing executed in color. Copies of this patent or patent application publication with color drawing(s) will be provided by the Office upon request and payment of the necessary fee.

[0014] FIGS. 1A and 1B. An alignment of hCCNT1-C261Y (SEQ ID NO:1, shown as hCCNT1*), hCCNT1 (SEQ ID NO:3), and mCCNT1 (SEQ ID NO:6) as aligned by Clustal Omega (www.ebi.ac.uk) (Sievers F, Wilm A, Dineen D, Gibson T J, Karplus K, Li W, Lopez R, McWilliam H, Remmert M, Söding J, Thompson J D, Higgins D G. Fast, scalable generation of high-quality protein multiple sequence alignments using Clustal Omega. *Mol Syst Biol.* 2011 Oct. 11; 7:539). An “*” (asterisk) indicates positions which have a single, fully conserved residue. A “:” (colon) indicates conservation between groups of strongly similar properties (scoring >0.5 in the Gonnet PAM 250 matrix). A “.” (period) indicates conservation between groups of weakly similar properties (scoring =<0.5 in the Gonnet PAM 250 matrix).

[0015] FIGS. 2A and 2B. An alignment of hXPO1-P411T-M412V-F414S (SEQ ID NO:7, shown as hXPO1*), hXPO1 (SEQ ID NO:9), and mXPO1 (SEQ ID NO:12) as aligned by Clustal Omega.

[0016] FIG. 3. Species-specific differences underpinning defects to HIV-1 Tat-CCNT1 and Rev-XPO1 interactions. (A) Summary of HIV-1's post-integration stages. (1) Host transcription factors activate low level HIV-1 transcription. (2) Early fully spliced viral mRNAs are translated to generate Tat and Rev. (3) Tat and Rev both translocate to the nucleus where Tat recruits pTEFb to upregulate transcriptional elongation and (4) Rev activates the nuclear export of late-stage, intron-retaining viral mRNAs and RNA genomes. (5) Full-length viral mRNAs are translated to generate Gag and Gag-Pol that encapsidate RNA genomes at virion assembly sites at the plasma membrane. B. Depiction of Tat (green)

bound to CCNT1 (gold). In hCCNT1, C261 is thought to promote Tat activity by stabilizing a zinc bridge (purple). Figure based on PDB: 40R5. C. Depiction of XPO1 species-specific “patch” domain located between HEAT repeats 9 and 10. Mutation of hXPO1 T411, V412, and S414 to P411, M412, and F414 (as found in mXpo1) causes a reduction in Rev activity, potentially due to destabilization of a Rev-bound hXPO1 dimer.

[0017] FIG. 4. Editing of hCCNT1 using CRISPR-Cas9 to introduce C261Y substitution. The AGA TGC TAT TTG CTT CAT TGC AGG CAT GCG AGG CTG CCA AGA AA sequence corresponds to bases 688-731 of SEQ ID NO:5. The AGA TGC TAT TTG CTT CAT TGC AGG CgT aCG AaG CTG CCA AGA AA sequence corresponds to bases 35-78 of SEQ ID NO:13. The ACEAAKK sequence corresponds to residues 260-266 of SEQ ID NO:3. The AYEAAKK sequence corresponds to residues 260-266 of SEQ ID NO:1.

[0018] FIGS. 5A and 5B. Editing hXPO1 using CRISPR-Cas9 to introduce P411T, M412V, and F414S substitutions. The CAG CTA TAT TTG CCC ATG TTA TTC AAG GTA ACA GAG CGG TTG GTT sequence in FIG. 5A corresponds to bases 495-539 of SEQ ID NO:11. The CAG CTG TAT TTG ACT GTG TTA TCA AAG GTA ACA GAG CGG TTG GTT sequence in FIG. 5A corresponds to bases 42-86 of SEQ ID NO:14. The QLYLPMLFK sequence in FIG. 5A corresponds to residues 407-415 of SEQ ID NO:9. The QLYLTVLSK sequence in FIG. 5A corresponds to residues 407-415 of SEQ ID NO:7. The GCC TCT CCG TTG CTT TCT GGA AGT CAA sequence in FIG. 5B corresponds to bases 444-470 of SEQ ID NO:11. The CAG CTA TAT TTG CCC ATG TTA TTC AAG GTA ACA GAG CGG TTG GTT sequence in FIG. 5B corresponds to bases 495-539 of SEQ ID NO:11. The CAG CTA TAT TTG CCC ATG TTA TTC AAG GTA ACA GAG CGG TTG GTT sequence in FIG. 5B corresponds to bases 14-40 of SEQ ID NO:15. The CAG CTA TAT TTG CCC ATG TTA TTC AAG GTA ACA GAG CGG TTG GTT sequence in FIG. 5B corresponds to bases 65-109 of SEQ ID NO:15. The ASPLLSGSQ sequences in FIG. 5B correspond to residues 390-398 of each of SEQ ID NO:9 and SEQ ID NO:7. The QLYLPMLFK sequence in FIG. 5B corresponds to residues 407-415 of SEQ ID NO:9. The QLYLTVLSK sequence in FIG. 5B corresponds to residues 407-415 of SEQ ID NO:7.

[0019] FIG. 6. Stacking species-informed gene changes to block HIV-1 and other retroviruses. The approach modifies CCNT1 (C261Y) and XPO1 (edits P411T, M412V, F414S) either separately or in tandem to generate cells resistant to infection by HIV-1 and other retroviruses including HIV-2 and potentially HTLV.

[0020] FIG. 7. Human T cells can be rendered resistant to HIV-1 by modifying a single, species-specific hCCNT1 codon. A and B: Jurkat T cell lines bearing homozygous hCCNT1 alleles encoding the C261Y mutation (cell lines 4.7F and 4.8C) proliferate similarly to the parental cell line (A) but exhibit profound resistance to HIV-1 gene expression after infection with a dual fluorescent reporter virus expressing a constitutive (EF1a promoter-driven) RFP reporter (red, that confirms infection) and a Tat/LTR-driven GFP reporter (green) (B). C: Tat function is lost in hCCNT1-C261Y cells unless wild-type hCCNT1 is co-expressed in trans after transient transfection of these cells with plasmids encoding a Tat/LTR-driven firefly luciferase reporter with or without Tat and the indicated transgenes.

[0021] FIG. 8. Modified T cells exhibit broad-spectrum antiviral properties. Modified hCCNT1-C261Y cells (cell lines 4.7F and 4.8C) are refractory to HIV-2 and SIVagm gene expression.

[0022] FIGS. 9A-9D. Population-level analysis of HIV-1 resistance in heterogeneous cell mixtures. FIG. 9A: Genomic DNA analysis of prescribed mixtures of wild-type and modified hCCNT1-C261Y cells (cell line 4.8C). The relative abundance of hCCNT1-C261Y cells in each defined mixture (lanes 2, 3, and 4) is confirmed by polymerase chain reaction (PCR) targeting hCCNT1 genomic locus and subsequent DNA cleavage using BsiWI enzyme. BsiWI restriction enzyme sites are only present in DNA amplicons from hCCNT1-C261Y cells (compare lanes 5 and 6 to lanes 2, 3, and 4; also see FIG. 4 design scheme). FIG. 9B: Flow cytometric characterization of HIV-1 resistance. Using a HIV-1 reporter virus (encoding genes expressing mVenus and mCherry proteins) and the gating scheme shown, the number of infected cells exhibiting virus uptake (Gate 1, black), virus uptake with early gene expression only (Gate 2, orange), and virus uptake with early and late gene expression (Gate 3, gray) are quantified. FIGS. 9C and 9D: Flow cytometric analysis of defined mixtures of wild-type and modified hCCNT1-C261Y cells infected with a HIV-1 reporter virus. Example flow cytometry dot plots (FIG. 9C) and the percentage of infected cells present within each gate (FIG. 9D) are shown. Consistent with the previous data showing that hCCNT1-C261Y cells are resistant to both early and late HIV-1 gene expression (FIGS. 7B and 7C), cell mixtures containing a high abundance of hCCNT1-C261Y cells (e.g., 20:80, left panel) have a higher relative proportion of infected cells in Gate 1 (FIG. 9D, black bars). Conversely, cell mixtures containing a high abundance of wild-type cells (e.g., 80:20, right panel) have a higher relative proportion of infected cells in Gates 2 and 3 (FIG. 9D, orange and gray bars).

[0023] FIGS. 10A-10D. Human T cells treated to express hXPO1-P411T-M412V-F414S are refractory to viral late gene expression. FIG. 10A: Genomic DNA analysis of wild-type and heterogeneous, modified hXPO1-P411T-M412V-F414S cells. CRISPR-treated T cells exhibit detectable editing at XPO1 genomic locus in a subset of cells (0.97 kb DNA amplicons are digested by PvuII restriction enzyme and yield smaller ~0.49kb bands, lanes 4 and 5) but not in untreated, wild-type cells (lanes 2 and 3) (also see FIGS. 5A and/or 5B for design scheme). FIG. 10B: Flow cytometric characterization of HIV-1 resistance (as previously described in FIG. 9B). FIGS. 10C and 10D: Flow cytometric analysis of infected, heterogeneous human T cell populations treated to produce the hXPO1-P411T-M412V-F414S modification. Example flow cytometry dot plots (FIG. 10C) are shown, with uninfected control cell populations in blue and infected cell populations in red for both low (1x) and high (5x) HIV-1 reporter virus doses. The percentage of infected cells present within the early (Gate 2) and late (Gate 3) viral gene expression gates are shown (FIG. 10D) for each HIV-1 reporter virus dose. CRISPR-treated or wild-type control cells exhibiting early gene expression only or early and late gene expression were quantified (FIG. 10D, orange and black bars, respectively). At both infectious doses, treated cell populations had fewer cells expressing both early and late genes (late phase, gate 3) compared to the number of cells expressing only early genes (early phase, gate 2), consistent with a block to HIV-1 Rev function (i.e.,

the XPO1-mediated transition from early gene expression to early and late gene expression).

DETAILED DESCRIPTION OF THE INVENTION

Genetically Modified Genes

[0024] One aspect of the invention is a genetically modified CCNT1 gene. The genetically modified CCNT1 gene of the invention encodes a protein comprising a sequence with a sequence identity of at least about 80% with respect to SEQ ID NO:1 and includes a tyrosine at a position corresponding to position 261 of SEQ ID NO:1.

[0025] SEQ ID NO:1 represents hCCNT1-C261Y, which is a modified version of the human CCNT1 protein (hCCNT1, CCNT1, Cyclin-T1) comprising a substitution of a cysteine to a tyrosine at position 261 of hCCNT1 (C261Y). The genetically modified CCNT1 gene encoding SEQ ID NO:1 can be generated from the human CCNT1 gene encoding hCCNT1 by modifying the codon encoding the cysteine at position 261 in hCCNT1 to a codon encoding a tyrosine.

[0026] SEQ ID NO:1 is:

```
(SEQ ID NO: 1)
MEGERKNNKRWYFTREQLNSPSRRFGVDPDKELSYRQQAANLLQDMGQR
LNVSQLTINTAIVYMHRFYMIQSFTQFPNGSVAPAAFLAAKVEEQPKKLE
HVIKVAHTCLHPQESLPDTRSEAYLQQVQDLVILESIILQTLGPFELTIDHP
HTHVVKCTQLVRASKDLAQTSYFMATNSLHLTTFSLQYTPPVVACVCIHLA
CKWSNWEIPVSTDGKHWWEYVDATVTLELDELDELTHEFLQILEKTPNRLKRI
WNWRAYEAAKKTADRRGTDEKTSEQTILNMIQSQSSDPTIAGLMSMSTST
TSAVPSLPVSESSSNLTSVEMLPGKRWLSSQPSFKLEPTQGHRTSENLLAL
TGVDSHSLPDGGSNAFISQKQNSKSVPSAKVSLKEYRAKHAEEAQAQKQKLE
NMEANVKSQYAYAAQNLLSHHDSHSSVILKMPIEGSENPERPFLEKADKTA
LKMRIPVAGGDKAASKPEEIKMRIKVHAAADKHNSVEDSVTKSREHKEKH
KTHPSNNHHHHHSHKSHSLSQLPVGTGNKRPDGPKHSSQTSNLAHKTYSL
SSSFSSSSSTRKRGPSSEETGGAVFDHPAKIAKSTKSSLNFSFPLPTMGQ
MPGHSSDTSGLSFSQPSCKTRVPHSLDKGPTGANGHNTTQTIDYQDVTVM
LHSLLSAQGVQPTQPTAFEFVRPYSYDYLNPRSGGISRRSGNTDKRPPPLP
SEPPPPPLPLPK
```

[0027] An exemplary coding sequence encoding SEQ ID NO:1 is represented by SEQ ID NO:2:

```
(SEQ ID NO: 2)
atggaggagagaggaagaacaacacacacgggtggtatttctactcgagaa
cagctggaaaatagccatcccgtcggttttggcgtggaccagataaagaa
ctttcttatcgccagcaggcggccaatctgcttcaggacatggggcagcgt
cttaacgtctcacaattgactatcaacactgctatagtatacatgcatcga
ttctacatgattcagtccttcacacagttccctggaaattctgtggctcca
gcagccttgtttctagcagctaaagtggaggagcagcccaaaaattggaa
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catgtcatcaaggtagcacatacttgtctccatcctcaggaatcccttctc
gatactagaagtgaggcttatttgcaacaagttcaagatctggtcatttta
gaaagcataattttgagactttaggctttgaactaacaattgatcaccca
catactcatgtagtaagtgactcaacttgttcgagcaagcaaggactta
gcacagacttcttacttcatggcaaccaacagcctgcatttgaccacatt
agcctgcagtagcacacctcctgtggtggcctgtgtctgcattcactggct
tgcaagtggccaattgggagatcccagcttcaactgacgggaagcactgg
tgggagtagttgacgcccactgtgacctggaaactttatgatgaactgaca
catgagtttctacagattttgagaaaaactcccaacagcctcaaacgcatt
tggaaatggagggcgtacgaagctccaagaaaaaaaagcagatgaccga
ggaacagatgaaaagacttcagagcagacaatcctcaatgatattccag
agctctcagacacaaccattgcaggtttaatgagcagtgcaactcttacc
acaagtgcagtgcccttccctgccagcttccgaagagtcacccagcaactta
accagtgtagagatgttgccgggaagcgttggctgtctcccaaccttct
ttcaaacagaacctactcagggtcatcggactagtgagaatttagcactt
acaggagtgatcattccttaccacaggatggttcaaagcatttatttcc
cagaagcagaatagtaagagtggtccatcagctaaagtgcactgaaagaa
taccgcgcaagcagcagaagaattggctgccagaagaggcaactggag
aacatggaagccaatgtgaagtcacaatagcattgctgccagaatctc
ctttctcatcatgatagccattcttctcagtcattctaaaaatgccatagag
ggttcagaaaaaccgagcggcctttcttgaaaaggctgacaaaacagct
ctcaaaatgagaatcccagtgccaggtggagataaagctgcttctcaaaa
ccagaggagataaaaaatgcgcataaaaagtcctgctgcagctgataagcac
aattctgtagaggacagtggtacaagagccgagagcacaagaaaaagcac
aagactcaccatctaatcatcatcatcatcataatcaccactcacacaag
cactctcattcccaacttccagttggtagctgggaacaaacgctcctggtag
ccaaaacatagtagccagacaagcaacttagcacataaaacctatagcttg
tctagttcttttctcttccagttctactcgtaaaaggggaccctctgaa
gagactggaggggctgtgttgatcatccagccaagattgccaagagtact
aaatcctcttccctaaatcttctcttcccttcaacttctacaatgggtcag
atgctcgggcatagctcagacacaagtgcccttctctttcacagcccagc
tgtaaaactcgtgtccctcattcgaaactggataaagggcccactggggcc
aatggtcacaacacgaccagacaatagactatcaagacactgtgaatatg
cttactcctcctcagtgccaggggtgtcagcccactcagcctactgca
tttgaattgttctcctttagtgactatctgaatcctcggctcgggtgga
atctcctcagatctggcaatcacagacaacccccggccaccacctctgcca
tcagaacctcctccaccacttccacccttctcaagtaa

[0028] The amino acid sequence of an exemplary hCCNT1 is represented by SEQ ID NO:3:

(SEQ ID NO: 3)
MEGERKNNKRWFPTREQLENSFRRFGVDPDKELSYRQQAANLLQDMGQR
LNVSQLTINTAIVMHRFYMIQSFTQFPGNSVAPAALFLAAKVEEQPKKLE
HVIKVAHTCLHPQESLPDTRSEAYLQQVQDLVILESIIILQTLGFELTIDHP
HTHVVKCTQLVRASKDLAQTSYFMATNSLHLTTFSLQYTPPVVACVCIH
CKWSNWEI PVSTDGKHWWEYVDATVTLLELDELHFLQILEKTPNRLKRI
WNWRACEAAKTKADDRGTDEKTSQTI LNMI SQSSSDTTIAGLMSMSTST
TSAVPLPVSSESSNLTSVEMLPGKRWLSQPSFKLEPTQGHRTSENAL
TGVDSHSLPQDGSNAFISQKQNSKSVPSAKVSLKEYRAKHAEEELAAQKQLE
NMEANVKSQYAYAAQNLLSHHDSHSSVILKMP IEGSENPERPFLEKADKTA
LKMRIPVAGDKAASKPEEIKMRIKVHAAADKHNSVEDSVTSKREHKEKH
KTHPSNHHHHHHHSHKSHS QLPVGTGNKRPGPKHSSQTSNLAKHTYSL
SSSFSSSSSTRKRGPSSEETGGAVFDHPAKIAKSTKSSSLNFSFPLPTMGQ
MPGHSSDTSGLSFSQPSCKTRVPHSKLDKPGTANGHNTTQTIIDYQDVTNM
LHLSLLSAQGVQPTQPTAFEFVRPYSYDLNPRSGGISRSNGNTDKPRPPPLP
SEPPPPPLPLPK

Various isoforms or variants of hCCNT1 include modifications to SEQ ID NO:3 in which positions 181-184 include a sequence or arginine-threonine-aspartic acid-threonine (RTDT) in place of serine-leucine-histidine-leucine (SLHL), position 77 includes arginine (R) in place of glutamine (Q), position 362 includes arginine (R) in place of histidine (H), and/or position 541 includes cysteine (C) in place of arginine (R). Any of these modifications can be included in the protein encoded by the genetically modified CCNT1 gene of the invention.

[0029] A coding sequence of the exemplary hCCNT1 is represented by SEQ ID NO:4:

(SEQ ID NO: 4)
atggaggagagaggaagaacaacaacaacgggtggtatttctcgcagaa
cagctggaaaatagccatcccgtcggtttggcgtggaccagataaagaa
ctttcttatcgccagcaggcggccaatctgctcaggacatggggcagcgt
cttaacgtctcacaattgactatcaacactgctatagtatacatgcatcga
ttctacatgattcagtccttcacacagttccctggaaattctgtggctcca
gcagccttgttcttagcagctaaagtggaggagcagccaaaaaattggaa
catgtcatcaaggtagcacatactgtctccatcctcaggaatcccttctc
gatactagaagtgaggcttatttgcaacaagttcaagatctggtcatttta
gaaagcataattttgagactttaggctttgaactaacaattgatcaccca
catactcatgtagtaagtgactcaacttgttcgagcaagcaaggactta
gcacagacttcttacttcatggcaaccaacagcctgcatttgaccacattt
agcctgcagtagcacacctcctgtggcctgtgtctgcattcactggct
tgcaagtggccaatgggagatcccagcttcaactgacgggaagcactgg

-continued

tgggagatggttgacgccactgtgaccttggaaacttttagatgaactgaca
catgagtttctacagattttgagaaaaactcccaacaggctcaaacgcatt
tggaaatggagggcatgagggctgccaagaaaaaaaagcagatgaccga
ggaacagatgaaaagacttcagagcagacaatcctcaatgatattccag
agctcttcagacacaaccattgcaggtttaatgagcatgtcaacttctacc
acaagtgcagtgacctccctgccagtctccgaagagtcaccagcaactta
accagtgtggagatgttgcgggcaagcgttggtgtcctcccaaccttct
ttcaaaactagaacctactcagggtcatcggactagtgagaatttagcactt
acaggagtgatcattccttaccacaggatggttcaaatgcatttatttcc
cagaagcagaatagtaagagtggtccatcagctaaagtgtcactgaaagaa
taccgcggaagcatgcagaagaatgggtgccagaagggcaactggag
aacatggaagccaatgtgaagtcacaatagcatatgctgccagaatctc
ctttctcatcatgatagccattctctcagtcattctaaaaatgccatagag
ggttcagaaaaccgagcggcctttctggaaaaggctgacaaaacagct
ctcaaaatgagaatcccagtgccaggtggagataaagctgcgtcttcaaaa
ccagaggagataaaaaatgcgcataaaaagtccatgctgcagctgataagcac
aattctgtagaggacagtggtacaagagccgagagcacaagaaaagcac
aagactcaccatctaatcatcatcatcataatcaccactcacacaag
cactctcattcccaactccagttggtactgggaacaacagctcctggtgat
ccaaaaatagtagccagacaagcaacttagcacataaaacctatagcttg
tctagtcttttctctcttccagttctactcgtaaaagggaccctctgaa
gagactggaggggctgtgttgatcatccagccaagattgccaagagtaact
aaatcctcttccctaaatttctcttcccttcaacttctacaatgggtcag
atgctcgggatagctcagacacaagtgcccttctcttccacagcccagc
tgtaaaactcgtgtccctcatcgaaaactggataaagggcccaactggggcc
aatggtcacaacacgaccagacaatagactatcaagacactgtgaatatg
cttcaactcctgctcagtgccagggtgtcagcccactcagcctactgca
tttgaattgttcgctccttatagtgactatctgaatcctcggtctggtgga
atctcctcgagatctggcaatcacagacaacccccggccaccacctctgcca
tcagaacctcctccaccacttccacccttccctaagtaa

[0030] The sequence of a portion of an exemplary human CCNT1 gene that can be edited to generate an exemplary modified CCNT1 gene is represented by SEQ ID NO:5:

(SEQ ID NO: 5)

TGAGATTAGAAGTAGGCTTGAGAGGCCGGGCATGGTGGCTCATGCCGTGAG
TCCCAGCACTTTGGGAGGCCAAGGCAGGCGGATCAACTGAGGTCAGGAGTT
CGAGACCAGCCTGGCCAACATGGTGAAACCTCGTCTCTACTAAAAATACAA
AAATAGCCAGGCATGGTGATGCACACCTGTAGTCCAGCTACTTGGGAGG
CTGAGACAGGAGAATCGCTTGAACCTCGGACGTTAGGTTGCAGTGAGCCGA

-continued

GATTGTGCCACTGCCTCCAGCCTGGATGACAAAAGTGAGACTCTGTCTCAA
ACAAAACAAACAAACAAAAACACAGTAACAAACAAAAAGAGTAGGCTTG
AGAGCACATCTTTTACTTTAGCATAAAAACCTCACAAAATTTCTAGAATC
AGTTATGGACTAACTATAATCATAAGCGAAGGCATGGATGTTTATGTATGA
ATTTTAGATAAGCATAGATCTTTTGTGTTATTATTGCTTTGTAACGTTTG
GATAGATTGCTGTGACTCTTAATTGAAGGTTTTAAAATCTTCTCTTGATGG
TAATATTTATTGGATTACATGTTAGGATAGCCTCCTGCCTGTGGCCTATCC
AGAACTTCCAGTGTGCTGCAAGTACAATCTACTCATCTCAGTGTTTTTTT
ATTTAGTAAATTACCTAAGTAAAGAGATGCTATTGCTTCATTGCAGGCAT
GCGAGGCTGCCAAGAAAACAAAAGCAGATGACCGAGGAACAGATGAAAAGA
CTTCAGAGCAGACAATCCTCAATATGATTTCCAGAGCTCTTCAGACACAA
CCATGCAGGTTTAAATGAGCATGTCAACTTCTACCACAAGTGCAGTGCCTT
CCCTGCCAGTCTCCGAAGAGTCATCCAGCAACTTAACCAGTGTGGAGATGT
TGCCGGGCAAGCGTTGGTGTCTCCCAACCTTCTTCAAACCTAGAACCTA
CTCAGGGTCATCGGACTAGTGAGAATTTAGC

Exemplary methods for performing the editing are described in the following examples.

[0031] The tyrosine at position 261 of the protein encoded by the genetically modified CCNT1 gene of the invention is modeled after the tyrosine at position 261 of the mouse CCNT1 protein (mCCNT1, Ccnt1), which is represented by SEQ ID NO:6:

(SEQ ID NO: 6)

MEGERKMNKRWFYFTREQLNSPSRRFVSDKELSYRQQAANLLQDMGQR
LNVSQLTINTAIVMHRFYMIQSFTQPHRYSMAPALFLAAKVEEQPKKLE
HVIKVAHTCLHPQESLPDRSEAYLQQVQDLVILESIIILQTLGPFELTIDHP
HTHVVKCTQLVRASKDLAQTSYFMATNSLHLTTFSLQYTPPVVACVCIHILA
CKWSNWEI PVSTDGKHWWEYVDATVTLBLELDELTHEFLQILEKTPSRLKRI
RNWRAYQAAMKTKPDRGADENTSEQTILNMI SQTSSDTTIAGLMSMSTAS
TSAVPSLPSSSESSSLTSVDMLQGERWLSQPPFKLEAAQGHRTSESLAL
IGVDHSLQDQSSAFGSQKQASKSVPSAKVSLKEYRAKHAEELAAQKRQLE
NMEANVKSQYAYAAQNLLSHDSHSSVILKMPIESSENPERPFLDKADKLSAL
KMRLPVASGDKAVSSKPEEIKMRIKVHSAGDKHNSIEDSVTKSREHKEKQR
THPSNHHHHHHNHSRHSLSLQLPAGVPSKRPSDPKHSSQTS TLAHKTYLSL
STLSSSSSTRKRGPEETGAAVFDHPAKIAKSTKSSLPFPFPPLPTMTQLP
GHSSDTSGLPFSQPSCKTRVPHMKLDKGGPPGANGHNATQSIDYQD TVNMLH
SLLSAQGVQPTQAPAFEFVHSYGEYMNPRAGAISSRSRGTDKPRPPPLPSE
PPPPPLPLPK

[0032] An alignment of hCCNT1-C261Y (SEQ ID NO:1, shown as hCCNT1*), hCCNT1 (SEQ ID NO:3), and mCCNT1 (SEQ ID NO:6) as aligned by Clustal Omega using default parameters is shown in FIGS. 1A and 1B.

[0033] With the exception of Y261, the genetically modified CCNT1 gene may encode a number of differences with

amino acid other than serine at a position corresponding to position 686 of SEQ ID NO:1; an amino acid other than glycine at a position corresponding to position 688 of SEQ ID NO:1; an amino acid other than glutamic acid at a position corresponding to position 689 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 691 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 695 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 697 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 698 of SEQ ID NO:1; an amino acid other than threonine at a position corresponding to position 704 of SEQ ID NO:1; and an amino acid other than leucine at a position corresponding to position 710 of SEQ ID NO:1.

[0034] In some versions, the differences encoded by the genetically modified CCNT1 gene with respect to mCCNT1 or other native CCNT1 proteins may comprise at least one, some, or all of: proline at a position corresponding to position 31 of SEQ ID NO:1; tyrosine at a position corresponding to position 37 of SEQ ID NO:1; proline at a position corresponding to position 79 of SEQ ID NO:1; glycine at a position corresponding to position 80 of SEQ ID NO:1; asparagine at a position corresponding to position 81 of SEQ ID NO:1; valine at a position corresponding to position 83 of SEQ ID NO:1; threonine at a position corresponding to position 110 of SEQ ID NO:1; asparagine at a position corresponding to position 250 of SEQ ID NO:1; tryptophan at a position corresponding to position 256 of SEQ ID NO:1; glutamic acid at a position corresponding to position 262 of SEQ ID NO:1; lysine at a position corresponding to position 265 of SEQ ID NO:1; alanine at a position corresponding to position 269 of SEQ ID NO:1; threonine at a position corresponding to position 274 of SEQ ID NO:1; lysine at a position corresponding to position 277 of SEQ ID NO:1; serine at a position corresponding to position 290 of SEQ ID NO:1; serine at a position corresponding to position 305 of SEQ ID NO:1; threonine at a position corresponding to position 306 of SEQ ID NO:1; threonine at a position corresponding to position 307 of SEQ ID NO:1; leucine at a position corresponding to position 313 of SEQ ID NO:1; valine at a position corresponding to position 315 of SEQ ID NO:1; serine at a position corresponding to position 316 of SEQ ID NO:1; asparagine at a position corresponding to position 322 of SEQ ID NO:1; serine at a position corresponding to position 325 of SEQ ID NO:1; glutamic acid at a position corresponding to position 327 of SEQ ID NO:1; proline at a position corresponding to position 330 of SEQ ID NO:1; lysine at a position corresponding to position 332 of SEQ ID NO:1; serine at a position corresponding to position 340 of SEQ ID NO:1; proline at a position corresponding to position 345 of SEQ ID NO:1; threonine at a position corresponding to position 346 of SEQ ID NO:1; asparagine at a position corresponding to position 354 of SEQ ID NO:1; threonine at a position corresponding to position 358 of SEQ ID NO:1; proline at a position corresponding to position 365 of SEQ ID NO:1; asparagine at a position corresponding to position 370 of SEQ ID NO:1; isoleucine at a position corresponding to position 373 of SEQ ID NO:1; asparagine at a position corresponding to position 378 of SEQ ID NO:1; histidine at a position corresponding to position 429 of SEQ ID NO:1; glycine at a position corresponding to position 443 of SEQ ID NO:1; glutamic acid at a position corresponding to

position 453 of SEQ ID NO:1; threonine at a position corresponding to position 458 of SEQ ID NO:1; isoleucine at a position corresponding to position 464 of SEQ ID NO:1; glycine at a position corresponding to position 468 of SEQ ID NO:1; alanine at a position corresponding to position 473 of SEQ ID NO:1; alanine at a position corresponding to position 488 of SEQ ID NO:1; alanine at a position corresponding to position 490 of SEQ ID NO:1; valine at a position corresponding to position 496 of SEQ ID NO:1; histidine at a position corresponding to position 510 of SEQ ID NO:1; lysine at a position corresponding to position 511 of SEQ ID NO:1; lysine at a position corresponding to position 527 of SEQ ID NO:1; serine at a position corresponding to position 531 of SEQ ID NO:1; valine at a position corresponding to position 535 of SEQ ID NO:1; threonine at a position corresponding to position 537 of SEQ ID NO:1; glycine at a position corresponding to position 538 of SEQ ID NO:1; asparagine at a position corresponding to position 539 of SEQ ID NO:1; glycine at a position corresponding to position 543 of SEQ ID NO:1; asparagine at a position corresponding to position 553 of SEQ ID NO:1; serine at a position corresponding to position 564 of SEQ ID NO:1; phenylalanine at a position corresponding to position 565 of SEQ ID NO:1; serine at a position corresponding to position 577 of SEQ ID NO:1; glycine at a position corresponding to position 582 of SEQ ID NO:1; serine at a position corresponding to position 599 of SEQ ID NO:1; serine at a position corresponding to position 603 of SEQ ID NO:1; serine at a position corresponding to position 606 of SEQ ID NO:1; glycine at a position corresponding to position 611 of SEQ ID NO:1; methionine at a position corresponding to position 613 of SEQ ID NO:1; serine at a position corresponding to position 624 of SEQ ID NO:1; serine at a position corresponding to position 637 of SEQ ID NO:1; threonine at a position corresponding to position 644 of SEQ ID NO:1; threonine at a position corresponding to position 651 of SEQ ID NO:1; threonine at a position corresponding to position 654 of SEQ ID NO:1; proline at a position corresponding to position 678 of SEQ ID NO:1; threonine at a position corresponding to position 679 of SEQ ID NO:1; glutamic acid at a position corresponding to position 682 of SEQ ID NO:1; arginine at a position corresponding to position 685 of SEQ ID NO:1; proline at a position corresponding to position 686 of SEQ ID NO:1; serine at a position corresponding to position 688 of SEQ ID NO:1; aspartic acid at a position corresponding to position 689 of SEQ ID NO:1; leucine at a position corresponding to position 691 of SEQ ID NO:1; serine at a position corresponding to position 695 of SEQ ID NO:1; glycine at a position corresponding to position 697 of SEQ ID NO:1; isoleucine at a position corresponding to position 698 of SEQ ID NO:1; asparagine at a position corresponding to position 704 of SEQ ID NO:1; and proline at a position corresponding to position 710 of SEQ ID NO:1.

[0035] In some versions, the genetically modified CCNT1 gene encodes a protein comprising a sequence with a sequence identity of at least about 85%, at least about 90%, at least about 91%, at least about 92%, at least about 93%, at least about 94%, at least about 95.0%, at least about 95.1%, at least about 95.2%, at least about 95.3%, at least about 95.4%, at least about 95.5%, at least about 95.6%, at least about 95.7%, at least about 95.8%, at least about 95.9%, 96.0%, at least about 96.1%, at least about 96.2%, at least about 96.3%, at least about 96.4%, at least about

96.5%, at least about 96.6%, at least about 96.7%, at least about 96.8%, at least about 96.9%, 97.0%, at least about 97.1%, at least about 97.2%, at least about 97.3%, at least about 97.4%, at least about 97.5%, at least about 97.6%, at least about 97.7%, at least about 97.8%, at least about 97.9%, 98.0%, at least about 98.1%, at least about 98.2%, at least about 98.3%, at least about 98.4%, at least about 98.5%, at least about 98.6%, at least about 98.7%, at least about 98.8%, at least about 98.9%, 99.0%, at least about 99.1%, at least about 99.2%, at least about 99.3%, at least about 99.4%, at least about 99.5%, at least about 99.6%, at least about 99.7%, at least about 99.8%, at least about 99.9% or more with respect to SEQ ID NO:1.

[0036] Another aspect of the invention is a genetically modified XPO1 gene. The genetically modified XPO1 gene of the invention encodes a protein comprising a sequence with a sequence identity of at least about 80% with respect to SEQ ID NO:7 and includes at least one of a threonine at a position corresponding to position 411 of SEQ ID NO:7, a valine at a position corresponding to position 412 of SEQ ID NO:7, and a serine at a position corresponding to position 414 of SEQ ID NO:7.

[0037] SEQ ID NO:7 represents hXPO1-P411T-M412V-F414S, which is a modified version of the human XPO1 protein (hXPO1, XPO1, Exportin-1) comprising a substitution of a proline to a threonine at position 411 of hXPO1 (P411T), a substitution of a methionine to a valine at position 412 of hXPO1 (M412V), and a substitution of a phenylalanine to a serine at position 414 of hXPO1 (F414S). The genetically modified XPO1 gene encoding SEQ ID NO:7 can be generated from the human XPO1 gene encoding hXPO1 by modifying the codon encoding the proline at position 411 in hXPO1 to a codon encoding a threonine, modifying the codon encoding the methionine at position 412 in hXPO1 to a codon encoding a valine, and modifying the codon encoding the phenylalanine at position 414 in hXPO1 to a codon encoding a serine.

[0038] SEQ ID NO:7 is:

(SEQ ID NO: 7)

MPAIMTMLADHAARQLLDFSQKLDINLLDNVNVNCLYHGEGAQQRMAQEVLT
HLKEHPDAWTRVDTILEFSQNMNTKYIQLILENVIKTRWKILPRNQCEGI
KKYVVGILIKTSSDPTCEVEKEKVYIGKLNMLLVQILKQEWPKHWPTFISDI
VGSARTSESLCQNNMVIKLLSEEVDFPSSGQITQVKS KHLKDMSCNEFSQ
IFQLCQFVMENSQNAFLVHATLETLRLNWIPLGYIFETKLISTLIYKFL
NVPMPFRNVSLKCLTEIAGVSVSQYEEQFVTLFTLTMMLKQMLPLNTNIRL
AYSNGKDDQNF IQNLSLFLCTFLKEHDQLIEKRLNLRRETLMEALHYMLLV
SEVEETEIPKICLEYWNHLAAELYRESFPFSTSASPLLSGSQHFDVPPRRQL
YLTVLSKVRLLMVSMAKPEEVLVVENDDQGEVVREFMKDTSINLYKNMRE
TLVYLTHLDYVDTERIMTEKLNHNQVNGTEWSWKNLNTLCWAIGSISGAMHE
EDEKRFLVTVIKDLLGLCEQKRGKDNKAI IASNIMYIVGQYPRFLRAHWKF
LKTVVNKLFEPMHETHDGVQDMACDTFIKIAQKCRRHVQVQVGVMPFID
EILNNINTIICDLQPQQVHTFYEAVGYMIGAQTDTVQEHLEIKYMLLPNQ
VWDSIIQQATKNVDILKDPETVKQLGSIKINVRACKAVGHFFVIQLGRIY

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LDMLNVYKCLSENI SAAIQANGEMVTKQPLIRSMRTVKRETLKLSGWVSR
SNDPQMVAENFVPPLLDAVLIDYQRNVPAAREPEVLSTMAI IVNKLGGHIT
AEIPQIFDAVFECTLNMINKDFEEYPEHRTNFFLLQLQAVNSHCFFAFLAIP
PTQPKLVLDSSI IWAFKHTMRNVADTGLQILFTLLQNVAQEEAAAQSFYQTY
FCDILQHIFSVVTDTSHTAGLTMHASILAYMFNLVVEEGKISTSLNPGNPVN
NQIFLQEYVANLLKSAFFHLQDAQVKLFVTVGLFSLNQDIPAFKEHLRDFLV
QIKEFAGEDTSDLFLEEREIALRQADEEKHKRQMSVPGIFNPHEIPEEMCD

[0039] An exemplary coding sequence encoding SEQ ID NO:7 is represented by SEQ ID NO:8:

(SEQ ID NO: 8)

ATGCCAGCAATTATGACAATGTTAGCAGACCATGCAGCTCGTCAGCTGCTT
GATTTACAGCAAAAACCTGGATATCAACTATTAGATAATGTGGTGAATTGC
TTATACCATGGAGAAGGAGCCAGCAAGAATGGCTCAAGAAGTACTGACA
CATTTAAAGGAGCATCCTGATGCTTGGACAAGAGTCGACACAATTTGGAA
TTTTCTCAGAATATGAATACGAAATACTATGGACTACAAATTTGGAAAAAT
GTGATAAAAACAAGGTGAAGATTCTTCCAGGAACCCAGTGCAGGAAGTA
AAAAAATACGTTGTTGGCCTCATTATCAAGACGTCATCTGACCCAACTGT
GTAGAGAAAAGAAAGGTGTATATCGGAAATTAATATGATCCTTGTTCAG
ATACTGAACAAGAATGGCCCAACATTTGGCCAACTTTTATCAGTGATATT
GTTGGAGCAAGTAGGACCAGCGAAAGTCTCTGTCAAATAATATGGTGATT
CTTAAACTCTTGAGTGAAGAAGTATTGTATTTCTCTAGTGGACAGATAACC
CAAGTCAAATCTAAGCATT TAAAAGACAGCATGTGCAATGAATCTCACAG
ATATTTCAACTGTGTGAGTTTGTAAATGAAAAATTCTCAAAATGCTCCACTT
GTACATGCAACCTTGAAAAATGCTCAGATTCTGAACTGGATTCCCTTG
GGATATATTTTTGAGACCAATTAATCAGCACATTGATTTATAAGTTCCTG
AATGTTCCAATGTTTCGAAATGTCTCTCTGAAGTGCCTCAGTGGATTGCT
GGTGTGAGTGAAGCCAATATGAAGAACAATTTGTAACACTATTTACTCTG
ACAATGATGCAACTAAAGCAGATGCTTCTTTAAATACCAATATTCGACTT
CGCTACTCAAATGAAAAAGATGATGAACAGAAGTCTCATTCAAATCTCAGT
TTGTTTCTCTGCACCTTTCTTAAAGAACATGATCAACTTATAGAAAAAGA
TTAAATCTCAGGAAACTCTTATGGAGGCCCTTCAATTATATGTTGTTGGTA
TCTGAAGTAGAAGAACTGAAATCTTTAAATTTGTCTTGAATACTGGAAAT
CATTGCGCTGCTGAACTCTATAGAGAGTCCATTCTCTACATCTGCGTCT
CCGTTGCTTTCTGGAAGTCAACATTTTGTATGTTCTCCAGGAGACAGCTG
TATTTGACCGTGTATCAAAGGTCGTTTATTAATGGTTAGTCGAATGGCT
AAACCAGAGGAAGTATTGGTTGTAGAGAAATGATCAAGGAGAAGTTGTGAGA
GAATTCATGAAGGATACAGATCCATAAATTTGTATAAGAATATGAGGGAA

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ACATTGGTTTATCTTACTCATCTGGATTATGTAGATACAGAAAGAATAATG
 ACAGAGAAGCTTCACAATCAAGTGAATGGTACAGAGTGGTCATGGAAAAAT
 TTGAATACATTGTGTTGGGCAATAGGCTCCATTAGTGGAGCAATGCATGAA
 GAGGACGAAAAACGATTTCTTGTACTGTTATAAAGGATCTATTAGGATTA
 TGTGAACAGAAAAGAGGCAAGATAATAAAGCTATTATTGCATCAAATATC
 ATGTACATAGTAGGTCATACCACGTTTTTTGAGAGCTCACTGGAAATTT
 CTGAAGACTGTAGTTAACAAGCTGTTCAATTCATGCATGAGACCCATGAT
 GGAGTCCAGGATATGGCTTGTGATACTTTCATATAAATAGCCAAAAATGC
 CGCAGGCATTTCTGTTCAAGTTCAGGTTGAGGAAAGTATGCCATTTATTGAT
 GAAATTTTGAACAACATTAACACTATTATTTGTGATCTTCAGCCTCAACAG
 GTTCATACGTTTTTGAAGCTGTGGGTACATGATTGGTGCAAAACAGAT
 CAAACAGTACAAGAACACTGTATAGAAAAGTACATGTTACTCCCTAATCAA
 GTGTGGGATAGTATAATCCAGCAGGCAACCAAAAATGTGGATATACTGAAA
 GATCCTGAAACAGTCAAGCAGCTTGGTAGCATTTTGAAAACAAATGTGAGA
 GCCTGCAAGCTGTTGGACACCCCTTTGTAATTCAGCTTGAAGAATTTAT
 TTAGATATGCTTAATGTATACAAGTGCCTCAGTAAAAATATTTCTGCAGCT
 ATCCAAGCTAATGGTGAATGGTTACAAGCAACCATGATTAGAAAGTATG
 CGAACTGTAAGGAAAGGAACTTTAAAGTTAATATCTGGTGGGTGAGCCGA
 TCCAATGATCCACAGATGGTCGCTGAAAATTTGTTCCCCCTCTGTTGGAT
 GCAGTCTCATTGATTATCAGAGAAATGTCCAGCTGCTAGAGAACCAGAA
 GTGCTTAGTACTATGGCCATAATGTCAACAAGTTAGGGGACATATAACA
 GCTGAAATACCTCAAATATTTGATGCTGTTTTTGAATGCACATTGAATATG
 ATAAATAAGGACTTTGAAGAATATCCTGAACATAGAAGCAACTTTTTCTTA
 CTACTTCAGGCTGTCAATTCTCATTGTTTCCAGCATTCCTTGCTATTCCA
 CCTACACAGTTTAACTTGTGTTTGGATCCATCATTGGGCTTCAAACAT
 ACTATGAGGAATGTCGAGATACGGGCTTACAGATACTTTTACACTCTTA
 CAAAATGTGCAACAAGAAGCTGCAGCTCAGAGTTTTATCAAACCTTAT
 TTTTGTGATATCTCCAGCATATCTTTTCTGTTGTGACAGACACTTCACAT
 ACTGTGGTTTTAACATGCATGCATCAATTCTTGATATATGTTTAAATTTG
 GTTGAAGAAGGAAAAATAAGTACATCATTAAATCCTGGAAATCCAGTTAAC
 AACCAATCTTTCTCAGGAATATGTGGCTAATCTCCTTAAGTCGGCCTTC
 CCTCACCTACAAGATGCTCAAGTAAAGCTCTTTGTGACAGGGCTTTTCAGC
 TTAAATCAAGATATCTCGCTTTCAGGAACATTTAAGAGATTTCTAGTT
 CAAAATAAGGAATTTGAGGTGAAGACACTTCTGATTTGTTTTGGAAGAG
 AGAGAAATAGCCCTACGGCAGGCTGATGAAGAGAAACATAAACGTCAAATG
 TCTGTCCCTGGCATCTTAAATCCACATGAGATTCAGAAAGAAATGTGTGAT
 TAA

[0040] The amino acid sequence of an exemplary hXPO1 is represented by SEQ ID NO:9:

(SEQ ID NO: 9)
 MPAINTMLADHAARQLLDFSQKLDINLLDNVNCVLYHGEGAQQORMAQEVLV
 HLKEHPDAWTRVDTILEFSQNMNTKYYGLQILENVIKTRWKILPRNQCEGI
 KKYVVGLIIKTSDDPTCVEKEKVYIGKLNMLVQILKQEWPKHWPTFISDI
 VGASRTSESLCQNNMVIKLLSEEVDFSSGQITQVKSJKLKDSDMCNEFSQ
 IFQLCQFVMENSQNAPLVHATLETLLRFLNWIPLGYIFETKLISLTIYKFL
 NVPMFRNVSLKCLTEIAGVSVSQYEEQFVTLFTLTMMLKQMLPLNTNIRL
 AYSNGKDEQNFIQNLSLFLCTFLKEHDQLIEKRLNLRRETLMEALHYMLLV
 SEVEETEIFKICLEYWNHLAAELYRESPFSTASPLLSGSQHFDVPPRRQL
 YLPMLFKVRLMLVSRMAKPEEVLVVENDQGEVREVMKDTDSINLYKNMRE
 TLVYLTHLDYVDTERIMTEKLNQVNGTEWSKLNLTLCWAIGSISGAMHE
 EDEKRFVTVIKDLLGLCEQKRGKDNKAIIASNIMYIVGQYPRFLRAHWKF
 LKTVVKNLFEFMHETHDGVQDMACDTFIKIAQKCRHFVQVQVGEVMPFID
 EILNNINTIICDLQPPQVHTFYEAVGYMIGAQTDTQTVQEHLEIKYMLLPNQ
 VWDSIIQQATKNVDILKDPETVKQLGSLKTNVRACKAVGHFFVQLGRIY
 LDMLNVYKCLSENI SAAIQANGEMVTKQPLIRSMRTVKRETLKLSGWVSR
 SNDPQMVAENFVPLLDVAVLIDYQRNVPAAREPEVLSTMAIIVNKLGGHIT
 AEIPQIFDAVFECTLMNINKDFEYEPHRTNFFLLQAVNSHCFFPAFLAIP
 PTQFKLVLDSIIWAFKHTMRNVADTGLQILFTLLQNVAQEEAAQSFYQTY
 FCDILQHIFSVVTDTSHTAGLTMHASILAYMFNLVEEGKISTSLNPNPNVN
 NQIFLQEVYVANLLKSAPFHLQDAQVKLQVTLGLFSLNQDIPAFKEHLRDLV
 QIKEFAGEDTSDLFLEEREIALRQADEEKHKRQMSVPGIFNPHEIPEEMCD

[0041] Various isoforms or variants of hXPO1 include modifications to SEQ ID NO:9 in which position 406 includes glycine (G) in place of arginine (R), position 953 includes glycine (G) in place of valine (V), and/or position 989 includes isoleucine (I) in place of leucine (L). Any of these modifications can be included in the protein encoded by the genetically modified XPO1 gene of the invention.

[0042] A coding sequence of the exemplary hXPO1 is represented by SEQ ID NO:10:

(SEQ ID NO: 10)
 ATGCCAGCAATTATGACAATGTTAGCAGACCATGCAGCTCGTCAGCTGCTT
 GATTTACAGCAAAAACCTGGATATCAACTATTAGATAAATGTGGTGAATTGC
 TTATACCATGGAGAAGGAGCCAGCAAGAATGGCTCAAGAAGTACTGACA
 CATTAAAGGAGCATCCTGATGCTTGGACAAGAGTCGACACAATTTGGAA
 TTTTCTCAGAATATGAATACGAAATACTATGGACTACAAATTTGGAAAAT
 GTGATAAAAACAAGGTGGAAGATTCTTCCAAGGAACAGTGCAGGAAGAA
 AAAAAATACGTTGTTGGCCTCATTATCAAGACGTCATGTGCCAACCTGT
 GTAGAGAAAAGAAAGGTGTATATCGGAAAATTAATATGATCCTTGTTCAG
 ATACTGAACAAGAATGGCCAAACATTTGGCCAACTTTTATCAGTGATATT

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GTTGGAGCAAGTAGGACCAGCGAAAAGTCTCTGTCAAATAATATGGTGATT
CTTAAACTCTTGAGTGAAGAAGTATTTGATTTCTCTAGTGGACAGATAACC
CAAGTCAAATCTAAGCATTAAAGACAGCATGTGCAATGAATTTCTCACAG
ATATTTCAACTGTGCAGTTTGTAAATGGAAAATTCTCAAATGCTCCACTT
GTACATGCAACCTTGGAAACATTGCTCAGATTTCTGAACTGGATTCCCCTG
GGATATATTTTTGAGACCAAATTAATCAGCAGATGATTTATAAGTTCCTG
AATGTTCCAATGTTTCGAAATGTCTCTGAAAGTGCCTCACTGAGATTGCT
GGTGTGAGTGAAGCCAATATGAAGAACAATTTGTAACACTATTTACTCTG
ACAATGATGCAACTAAAGCAGATGCTTCTTTAAATACCAATATTCGACTT
GCGTACTCAAATGGAAAAGATGATGAACAGAACTTCATTCAAATCTCAGT
TTGTTTTCTGACACTTTCTTAAGGAACATGATCAACTTATAGAAAAAGA
TTAAATCTCAGGAACTCTTATGGAGGCCCTTCAATATATGTTGTTGGTA
TCTGAAGTAGAAGAACTGAAATCTTAAAATTTGTCTGAATACTGGAAT
CATTTGGCTGCTGAACTCTATAGAGAGAGTCCATCTCTACATCTGCCTCT
CCGTGCTTTCTGGAAGTCAACATTTTGATGTTCTCCAGGAGACAGCTA
TATTTGCCCATGTTATTCAAGGTCCGTTTATTAATGGTTAGTCGAATGGCT
AAACCAGAGGAAGTATTGGTTGTAGAGAATGATCAAGGAGAAGTTGTGAGA
GAATTCATGAAGGATACAGATTCATAAATTTGTATAAGAATATGAGGGAA
ACATTTGGTTTACTTACTCATCTGGATTATGTAGATACAGAAAGAATAATG
ACAGAGAAGCTTCACAATCAAGTGAATGGTACAGAGTGGTCATGGAAAAAT
TTGAATACATGTGTTGGGCAATAGGCTCCATTAGTGGAGCAATGCATGAA
GAGGACGAAAAACGATTTCTTGTACTGTTATAAAGGATCTATTAGGATTA
TGTGAACAGAAAGAGGCAAGATAATAAAGCTATTATTGCATCAAATATC
ATGTACATAGTAGGTCATACCCAGTTTTTTGTAGAGCTCACTGGAAATTT
CTGAAGACTGTAGTTAAACAAGCTGTTGAAATTCATGCATGAGACCCATGAT
GGAGTCCAGGATATGGCTTGTGATACTTTCATTAAAATAGCCAAAAATGC
CGCAGGCATTTCTGTTCAAGTTCAGGTTGAGGAAGTATGCCATTTATTGAT
GAAATTTGAACAACATTAACACTATTATTGTGATCTTCAGCTCAACAG
GTTTACATGTTTTATGAAGCTGTGGGTACATGATTTGGTGCAAAACAGAT
CAAACAGTACAAGAACAATTTGATAGAAAAGTACATGTTACTCCCTAATCAA
GTGTGGGATAGTATAATCCAGCAGGCAACCAAAAATGTGGATATACTGAAA
GATCTGAAACAGTCAAGCAGCTTGGTAGCATTTTGAAAACAAATGTGAGA
GCCTGCAAAGCTGTGGACACCCCTTTGTAATTCAGCTTGAAGAATTTAT
TTAGATATGCTTAAATGTATACAAGTGCCTCAGTGAATAATTTCTGCAGCT
ATCCAAGCTAATGGTGAATGGTTACAAAGCAACCATTTGATTAGAAGTATG
CGAAGTGTAAAAGGAACTTTAAAGTTAATATCTGGTTGGGTGAGCCGA
TCCAATGATCCACAGATGGTGCCTGAAAATTTGTTCCCTCTGTTGGAT
GCAGTTCTCATTGATTATCAGAGAAATGTCCAGCTGCTAGAGAACCAGAA
GTGCTTAGTACTATGGCCATAATTTGCAACAAGTTAGGGGACATATAACA

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GCTGAAATACCTCAAATATTTGATGCTGTTTTTGAATGCACATTGAATATG
ATAAATAAGGACTTTGAAGAATATCTGAACATAGAACGAACTTTTTCTTA
CTACTTCAGGCTGTCAATTCTCATTGTTTCCCAGCATTCTTGCTATTCCA
CCTACACAGTTTAAACTGTTTTGGATTCCATCATTGGGGCTTTCAAACAT
ACTATGAGGAATGTCGCAGATACGGGCTTACAGATACTTTTTTACACTCTTA
CAAATGTTGCACAAGAAGAGCTGCAGCTCAGAGTTTTTATCAAACCTTAT
TTTTGTGATATCTCCAGCATATCTTTCTGTGTGACAGACACTTCACAT
ACTGCTGGTTTAAACAATGCATGCATCAATCTTGATATATGTTAATTTG
GTTGAAGAAGGAAAAATAAGTACATCATAAATCTGGAAATCCAGTTAAC
AACCAAATCTTCTTCAGGAATATGTGGCTAATCTCCTTAAGTCGGCCTTC
CCTCACCTACAAGATGCTCAAGTAAAGCTCTTTGTGACAGGGCTTTTCAGC
TTAAATCAAGATATTTCTGCTTTCAGGAACATTTAAGAGATTTCTTAGTT
CAAATAAAGGAATTTGCAGGTGAAGACACTTCTGATTTGTTTTTGAAGAG
AGAGAAATAGCCCTACGGCAGGCTGATGAAGAGAAACATAAACGTCAAATG
TCTGTCCCTGGCATCTTAAATCCACATGAGATTCAGAAGAAATGTGTGAT
TAA

[0043] The sequence of a portion of an exemplary human XPO1 gene that can be edited to generate an exemplary genetically modified XPO1 gene is represented by SEQ ID NO:11:

(SEQ ID NO: 11)
TTCTCTCCTCTGTGATGGTACATTTGGGTTGTGATACCACCTTATTGGCACC
CAAGGCCTTTTAAATAAATGTCGTTCCATTAGGAGACATGATAAAAAACA
TATTGATCAACTACTATGTGAGAGATTTTGAAGTGCCTTAGGGCATGTCA
GAAGAAGCAGAGTTACTCCAGAGTTTGTGCTTATTGATAAGTATTGAAA
TCTGAGTTGTGATGAATAAAACATGAATTTTTATTTCCCTTAAGGTGTA
CAAGTGAAGAACAATTTGAAGTTGGTAATGTTAAGAATATTTTAACAGT
TTTGGTCTTCTGTGTAGGCCCTTCATTATATGTTGTTGGTATCTGAAGTAG
AAGAACTGAAATCTTAAAATTTGTCTGAACTAGGAATCATTGGCTG
CTGAACTCTATAGAGAGAGTCCATTCTCTACATCTGCCTCTCCGTTGCTTT
CTGGAAGTCAACATTTGATGTTCTCCAGGAGACAGCTATATTGCCCCA
TGTTATTCAAGTAAACAGAGCGGTTGGTTGAGTGTCTTCTGTTGCATAC
TGTGGTTTTGAGGTCTGAATCAAATACTTCTAATCTGTGTAATAAATTA
GCTATAAAAAGAGAACCAACAACCTTCTCCATGAGTGTGGAAGTATAGAAC
ATGAAAGGAGTTGAGTCTAGAACCTTGATTTCTCAAGAGTGTGGTCTTCTC
TCAGTATCAACATTGGTTGTGATTTTCGTTAGGCAATTCATTGGCCACCTG
CCAATCTACTAAACCAGAGTCTAGGAATGAGACACAGGAACTCCTGTAAAC
AGAAGTTGGTTAAAAAATCACATTAACACACTTAAATAATATAAAGC
CATTTTTGTAGAATTACAGTGAATAAATTTTTTCTTTTGGAGACAGGGT

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CTTGCTCTGTGGCTCAGGTTGGAGTGCAGTGGCGTGGTCATAGCTCACTAC
AATCTTGA

Exemplary methods for performing the editing are described in the following examples.

[0044] The threonine at position 411, the valine at position 412, and/or the serine at position 414 of the protein encoded by the genetically modified XPO1 gene of the invention are modeled after the threonine at position 411, the valine at position 412, and/or the serine at position 414 of the mouse XPO1 protein (mXPO1, Xpo1), which is represented by SEQ ID NO:12:

(SEQ ID NO: 12)
MPAIMTMLADHAARQLLDFSQKLDINLLDNVNCCLYHGEGAQQRMAQEVLT
HLKEHPDAWTRVDTILEFSQNMNTKYIQLILENVIKTRWKILPRNQCEGI
KKYVVGLIIKTSDDPTCEVEKEKVYIGKLNMLLVQILKQEWPKHWPTFISDI
VGASRTSESLCQNMNVILKLLSEEVDFSSGQITQVKAKHLKDSMCNEFSQ
IFQLCQFVMENSQNAPLVHATLETLLRFLNWIPLGYIFETKLISTLIYKFL
NVPFRNVSLKCLTEIAGVSVSQYEEQFETLFTLTMMQLKQMLPLNTNIRL
AYSNGKDDQNFIQNLSLFLCTFLKEHGQLLEKRLNLRREALMEALHYMLLV
SEVEETEIEFKICLEYWNHLAELRYESPFSASPLLSGSQHFDIPRRQL
YLTVLSKVRLLMVSRAKPEEVLVVENDQGEVVRREFMKDTSINLYKNMRE
TLVYLTHLDYVDTEIIMTKKLQNVNGTEWSWKNLNTLCWAIGSISGAMHE
EDEKRFLVTVIKDLLGLCEQKRGKDNKAIIASNIMYIVGQYPRFLRAHWKF
LKTVVNKLFEFMEHETHDGVQDMACDTFIKIAQKCRHFVQVQVGEVMPFID
EILNNINTIICDLQPPQVHTFYEAVGYMIGAQTDTQTVQEHLEIKYMLLPNQ
VWDSIIQQATKNVDILKDPETVKQLGSLKTNVRACKAVGHFVVIQLGRIY
LDMLNVYKCLSENI SAAIQANGEMVTKQPLIRSMRTVKRETLKLSIGVWSR
SNDPQMAENFVPLLDVAVLIDYQRNVPAAREPEVLSTMAIIVNKLGGHIT
AEIPQIFDAVPECTLNMINKDFEEYPEHRTNFFLLQAVNSHCFFAFLAIP
PAQPKLVLDSEIIWAFKHTMRNVADTGLQILFTLLQNVQEEAAAQSFYQTY
FCDILQHIFSVVTDTSHTAGLTMHASILAYMFNLVEEGKISTPLNPGNPNV
NQMFIQDYVANLLKSAFFHLQDAQVKLFVTLGFLSNQDIPAFKEHLRDFLV
QIKFAGEDTSDLFLEERETALRQAQEEKHKLQMSVPGILNPHIPEEMCD

[0045] An alignment of hXPO1-P411T-M412V-F414S (SEQ ID NO:7, shown as hXPO1*), hXPO1 (SEQ ID NO:9), and mXPO1 (SEQ ID NO:12) as aligned by Clustal Omega using default parameters is shown in FIGS. 2A and 2B.

[0046] With the exception of T411, V412, and/or S414, the genetically modified XPO1 gene may encode a number of differences with respect to mXPO1 or other native XPO1 proteins. These differences may comprise at least one, some, or all of: an amino acid other than aspartic acid at a position corresponding to position 100 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 118 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 151 of SEQ ID NO:7; an amino acid other than alanine at a position

corresponding to position 191 of SEQ ID NO:7; an amino acid other than serine at a position corresponding to position 215 of SEQ ID NO:7; an amino acid other than glutamic acid at a position corresponding to position 284 of SEQ ID NO:7; an amino acid other than valine at a position corresponding to position 306 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 334 of SEQ ID NO:7; an amino acid other than leucine at a position corresponding to position 337 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 346 of SEQ ID NO:7; an amino acid other than isoleucine at a position corresponding to position 402 of SEQ ID NO:7; an amino acid other than isoleucine at a position corresponding to position 474 of SEQ ID NO:7; an amino acid other than lysine at a position corresponding to position 478 of SEQ ID NO:7; an amino acid other than glutamine at a position corresponding to position 481 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 869 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 909 of SEQ ID NO:7; an amino acid other than proline at a position corresponding to position 961 of SEQ ID NO:7; an amino acid other than serine at a position corresponding to position 966 of SEQ ID NO:7; an amino acid other than serine at a position corresponding to position 969 of SEQ ID NO:7; an amino acid other than valine and/or methionine at a position corresponding to position 972 of SEQ ID NO:7; an amino acid other than isoleucine at a position corresponding to position 974 of SEQ ID NO:7; an amino acid other than aspartic acid at a position corresponding to position 976 of SEQ ID NO:7; an amino acid other than threonine at a position corresponding to position 1040 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 1043 of SEQ ID NO:7; an amino acid other than glutamine at a position corresponding to position 1046 of SEQ ID NO:7; an amino acid other than leucine at a position corresponding to position 1052 of SEQ ID NO:7; and an amino acid other than leucine at a position corresponding to position 1060 of SEQ ID NO:7.

[0047] In some versions, the differences encoded by the genetically modified XPO1 gene with respect to mXPO1 or other native XPO1 proteins may comprise at least one, some, or all of: glutamic acid at a position corresponding to position 100 of SEQ ID NO:7; threonine at a position corresponding to position 118 of SEQ ID NO:7; serine at a position corresponding to position 151 of SEQ ID NO:7; serine at a position corresponding to position 191 of SEQ ID NO:7; asparagine at a position corresponding to position 215 of SEQ ID NO:7; valine at a position corresponding to position 284 of SEQ ID NO:7; leucine at a position corresponding to position 306 of SEQ ID NO:7; aspartic acid at a position corresponding to position 334 of SEQ ID NO:7; isoleucine at a position corresponding to position 337 of SEQ ID NO:7; threonine at a position corresponding to position 346 of SEQ ID NO:7; valine at a position corresponding to position 402 of SEQ ID NO:7; arginine at a position corresponding to position 474 of SEQ ID NO:7; glutamic acid at a position corresponding to position 478 of SEQ ID NO:7; histidine at a position corresponding to position 481 of SEQ ID NO:7; threonine at a position corresponding to position 869 of SEQ ID NO:7; alanine at a position corresponding to position 909 of SEQ ID NO:7; serine at a position corresponding to position 961 of SEQ ID NO:7; asparagine at a position corresponding to position 966

of SEQ ID NO:7; asparagine at a position corresponding to position 969 of SEQ ID NO:7; isoleucine at a position corresponding to position 972 of SEQ ID NO:7; leucine at a position corresponding to position 974 of SEQ ID NO:7; glutamic acid at a position corresponding to position 976 of SEQ ID NO:7; isoleucine at a position corresponding to position 1040 of SEQ ID NO:7; arginine at a position corresponding to position 1043 of SEQ ID NO:7; aspartic acid at a position corresponding to position 1046 of SEQ ID NO:7; arginine at a position corresponding to position 1052 of SEQ ID NO:7; and phenylalanine at a position corresponding to position 1060 of SEQ ID NO:7.

[0048] In some versions, the genetically modified XPO1 gene encodes a protein comprising a sequence with a sequence identity of at least about 85%, at least about 90%, at least about 91%, at least about 92%, at least about 93%, at least about 94%, at least about 95.0%, at least about 95.1%, at least about 95.2%, at least about 95.3%, at least about 95.4%, at least about 95.5%, at least about 95.6%, at least about 95.7%, at least about 95.8%, at least about 95.9%, 96.0%, at least about 96.1%, at least about 96.2%, at least about 96.3%, at least about 96.4%, at least about 96.5%, at least about 96.6%, at least about 96.7%, at least about 96.8%, at least about 96.9%, 97.0%, at least about 97.1%, at least about 97.2%, at least about 97.3%, at least about 97.4%, at least about 97.5%, at least about 97.6%, at least about 97.7%, at least about 97.8%, at least about 97.9%, 98.0%, at least about 98.1%, at least about 98.2%, at least about 98.3%, at least about 98.4%, at least about 98.5%, at least about 98.6%, at least about 98.7%, at least about 98.8%, at least about 98.9%, 99.0%, at least about 99.1%, at least about 99.2%, at least about 99.3%, at least about 99.4%, at least about 99.5%, at least about 99.6%, at least about 99.7%, at least about 99.8%, at least about 99.9% or more with respect to SEQ ID NO:7.

[0049] Throughout the specification, a reference may be made using an abbreviation of a gene name or a polypeptide name, but it is understood that such an abbreviated gene or polypeptide name represents the genus of genes or polypeptides, respectively. Such gene names include all genes encoding the same polypeptide and homologous polypeptides having the same physiological function. Polypeptide names include all polypeptides that have the same activity (e.g., that catalyze the same fundamental chemical reaction).

[0050] Unless otherwise indicated, the accession numbers referenced herein are derived from the NCBI database (National Center for Biotechnology Information) maintained by the National Institute of Health, U.S.A.

[0051] EC numbers are established by the Nomenclature Committee of the International Union of Biochemistry and Molecular Biology (NC-IUBMB) (available at www.chem.qmul.ac.uk/iubmb/enzyme/). The EC numbers referenced herein are derived from the KEGG Ligand database, maintained by the Kyoto Encyclopedia of Genes and Genomics, sponsored in part by the University of Tokyo.

[0052] The term “alignment” refers to a method of comparing two or more polynucleotides or polypeptide sequences for the purpose of determining their relationship to each other. Alignments are typically performed by computer programs that apply various algorithms; however it is also possible to perform an alignment by hand. Alignment programs typically iterate through potential alignments of sequences and score the alignments using substitution tables, employing a variety of strategies to reach a potential

optimal alignment score. Commonly-used alignment algorithms include, but are not limited to, CLUSTALW, (see, Thompson J. D., Higgins D. G., Gibson T. J., *CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice*, *Nucleic Acids Research* 22: 4673-4680, 1994); CLUSTALV, (see, Larkin M. A., et al., *CLUSTALW2, ClustalW and ClustalX version 2*, *Bioinformatics* 23(21): 2947-2948, 2007); Jotun-Hein, Muscile et al., *MUSCLE: a multiple sequence alignment method with reduced time and space complexity*, *BMC Bioinformatics* 5: 113, 2004); Mafft, Kalign, ProbCons, and T-Coffee (see Notredame et al., *T-Coffee: A novel method for multiple sequence alignments*, *Journal of Molecular Biology* 302: 205-217, 2000). Exemplary programs that implement one or more of the above algorithms include, but are not limited to MegAlign from DNASTar (DNASTar, Inc. 3801 Regent St. Madison, Wis. 53705), MUSCLE, T-Coffee, CLUSTALX, CLUSTALV, JalView, Phylip, and Discovery Studio from Accelrys (Accelrys, Inc., 10188 Telesis Ct, Suite 100, San Diego, Calif. 92121). In a non-limiting example, MegAlign is used to implement the CLUSTALW alignment algorithm with the following parameters: Gap Penalty 10, Gap Length Penalty 0.20, Delay Divergent Seqs (30%) DNA Transition Weight 0.50, Protein Weight matrix Gonnet Series, DNA Weight Matrix IUB.

[0053] The term “consensus sequence” or “canonical sequence” refers to an archetypical amino acid sequence against which all variants of a particular protein or sequence of interest are compared. Either term also refers to a sequence that sets forth the nucleotides that are most often present in a polynucleotide sequence of interest. For each position of a protein, the consensus sequence gives the amino acid that is most abundant in that position in the sequence alignment.

[0054] The term “conservative substitutions” or “conserved substitutions” refers to, for example, a substitution of an amino acid with a conservative variant. The proteins encoded by the genetically modified CCNT1 and XPO1 genes may comprise one or more conservative substitutions for any residue at any position, except for the tyrosine at the position corresponding to position 261 of SEQ ID NO:1 in the genetically modified CCNT1 gene and the threonine at the position corresponding to position 411 of SEQ ID NO:7, the valine at the position corresponding to position 412 of SEQ ID NO:7, and the serine at the position corresponding to position 414 of SEQ ID NO:7 in the genetically modified XPO1 gene.

[0055] “Conservative variant” refers to residues that are functionally similar to a given residue. Amino acids within the following groups are conservative variants of one another: glycine, alanine, serine, and proline (very small); alanine, isoleucine, leucine, methionine, phenylalanine, valine, proline, and glycine (hydrophobic); alanine, valine, leucine, isoleucine, methionine (aliphatic-like); cysteine, serine, threonine, asparagine, tyrosine, and glutamine (polar); phenylalanine, tryptophan, tyrosine (aromatic); lysine, arginine, and histidine (basic); aspartate and glutamate (acidic); alanine and glycine; asparagine and glutamine; arginine and lysine; isoleucine, leucine, methionine, and valine; and serine and threonine.

[0056] The terms “corresponds to” or “corresponding to” refer to an amino acid residue or position in a first protein sequence being positionally equivalent to an amino acid

residue or position in a second reference protein sequence by virtue of the fact that the residue or position in the first protein sequence aligns to the residue or position in the reference sequence using bioinformatic techniques, for example, using the methods described herein for preparing a sequence alignment. The corresponding residue in the first protein sequence is then assigned the position number in the second reference protein sequence.

[0057] The term “deletion,” when used in the context of an amino acid sequence, means a deletion in or a removal of one or more residues from the amino acid sequence of a precursor protein, resulting in a mutant protein having at least one less amino acid residue as compared to the precursor protein. The term can also be used in the context of a nucleotide sequence, which means a deletion in or removal of a nucleotide from the polynucleotide sequence of a precursor polynucleotide.

[0058] The term “expressed genes” refers to genes that are transcribed into messenger RNA (mRNA) and then translated into protein, as well as genes that are transcribed into types of RNA, such as transfer RNA (tRNA), ribosomal RNA (rRNA), and regulatory RNA, which are not translated into protein.

[0059] “Gene” refers to a polynucleotide (e.g., a DNA segment), which encodes a polypeptide, and may include regions preceding and following the coding regions as well as intervening sequences (introns) between individual coding segments (exons).

[0060] The term “homologous genes” refers to a pair of genes from different but related species, which correspond to each other and which are identical or similar to each other. The term encompasses genes that are separated by the speciation process during the development of new species (e.g., orthologous genes), as well as genes that have been separated by genetic duplication (e.g., paralogous genes).

[0061] The term “endogenous protein” refers to a protein that is native to or naturally occurring in a cell. “Endogenous polynucleotide” refers to a polynucleotide that is in the cell and was not introduced into the cell using recombinant engineering techniques, for example, a gene that was present in the cell when the cell was originally isolated from nature. Conversely, the term “heterologous” refers to a protein or a polynucleotide that does not naturally occur in a host cell.

[0062] The term “homologous recombination” refers to the exchange of DNA fragments between two DNA molecules or paired chromosomes at sites of identical or nearly identical nucleotide sequences. In certain embodiments, chromosomal integration is homologous recombination.

[0063] The term “homologous sequences” as used herein refers to a polynucleotide or polypeptide sequence having, for example, about 100%, about 99% or more, about 98% or more, about 97% or more, about 96% or more, about 95% or more, about 94% or more, about 93% or more, about 92% or more, about 91% or more, about 90% or more, about 88% or more, about 85% or more, about 80% or more, about 75% or more, about 70% or more, about 65% or more, about 60% or more, about 55% or more, about 50% or more, about 45% or more, or about 40% or more sequence identity to another polynucleotide or polypeptide sequence when optimally aligned for comparison. In particular embodiments, homologous sequences can retain the same type and/or level of a particular activity of interest. In some embodiments, homologous sequences have between 85% and 100%

sequence identity, whereas in other embodiments there is between 90% and 100% sequence identity. In particular embodiments, there is 95% and 100% sequence identity.

[0064] “Homology” refers to sequence similarity or sequence identity. Homology is determined using standard techniques known in the art (see, e.g., Smith and Waterman, *Adv. Appl. Math.*, 2:482, 1981; Needleman and Wunsch, *J. Mol. Biol.*, 48:443, 1970; Pearson and Lipman, *Proc. Natl. Acad. Sci. USA* 85:2444, 1988; programs such as GAP, BESTFIT, FASTA, and TFasta in the Wisconsin Genetics Software Package (Genetics Computer Group, Madison, Wis.); and Devereux et al., *Nucl. Acid Res.*, 12:387-395, 1984). A non-limiting example includes the use of the BLAST program (Altschul et al., Gapped BLAST and PSI-BLAST: a new generation of protein database search programs, *Nucleic Acids Res.* 25:3389-3402, 1997) to identify sequences that can be said to be “homologous.” A recent version such as version 2.2.16, 2.2.17, 2.2.18, 2.2.19, or the latest version, including sub-programs such as blastp for protein-protein comparisons, blastn for nucleotide-nucleotide comparisons, tblastn for protein-nucleotide comparisons, or blastx for nucleotide-protein comparisons, and with parameters as follows: Maximum number of sequences returned 10,000 or 100,000; E-value (expectation value) of $1e-2$ or $1e-5$, word size 3, scoring matrix BLOSUM62, gap cost existence 11, gap cost extension 1, may be suitable. An E-value of $1e-5$, for example, indicates that the chance of a homologous match occurring at random is about 1 in 10,000, thereby marking a high confidence of true homology.

[0065] The term “identical,” in the context of two polynucleotide or polypeptide sequences, means that the residues in the two sequences are the same when aligned for maximum correspondence, as measured using a sequence comparison or analysis algorithm such as those described herein. For example, if when properly aligned, the corresponding segments of two sequences have identical residues at 5 positions out of 10, it is said that the two sequences have a 50% identity. Most bioinformatic programs report percent identity over aligned sequence regions, which are typically not the entire molecules. If an alignment is long enough and contains enough identical residues, an expectation value can be calculated, which indicates that the level of identity in the alignment is unlikely to occur by random chance.

[0066] The term “insertion,” when used in the context of a polypeptide sequence, refers to an insertion in the amino acid sequence of a precursor polypeptide, resulting in a mutant polypeptide having an amino acid that is inserted between two existing contiguous amino acids, i.e., adjacent amino acids residues, which are present in the precursor polypeptide. The term “insertion,” when used in the context of a polynucleotide sequence, refers to an insertion of one or more nucleotides in the precursor polynucleotide between two existing contiguous nucleotides, i.e., adjacent nucleotides, which are present in the precursor polynucleotides.

[0067] The term “introduced” refers to, in the context of introducing a polynucleotide sequence into a cell, any method suitable for transferring the polynucleotide sequence into the cell. Such methods for introduction include but are not limited to protoplast fusion, transfection, transformation, conjugation, and transduction (see, e.g., Ferrari et al., *Genetics*, in Hardwood et al, (eds.), *Bacillus*, Plenum Publishing Corp., pp. 57-72, 1989).

[0068] The term “isolated” or “purified” means a material that is removed from its original environment, for example,

the natural environment if it is naturally occurring. A material is said to be “purified” when it is present in a particular composition in a higher or lower concentration than the concentration that exists prior to the purification step(s). For example, with respect to a composition normally found in a naturally occurring or wild type organism, such a composition is “purified” when the final composition does not include some material from the original matrix. As another example, a naturally occurring polynucleotide or polypeptide present in a living animal is not isolated, but the same polynucleotide or polypeptide, separated from some or all of the coexisting materials in the natural system, is isolated, whether such process is through genetic engineering or mechanical separation. Such polynucleotides can be parts of vectors. Alternatively, such polynucleotides or polypeptides can be parts of compositions. Such polynucleotides or polypeptides can be considered “isolated” because the vectors or compositions comprising thereof are not part of their natural environments. In another example, a polynucleotide or protein is said to be purified if it gives rise to essentially one band in an electrophoretic gel or a blot.

[0069] The term “mutation” refers to, in the context of a polynucleotide, a modification to the polynucleotide sequence resulting in a change in the sequence of a polynucleotide with reference to a precursor polynucleotide sequence. A mutant polynucleotide sequence can refer to an alteration that does not change the encoded amino acid sequence, for example, with regard to codon optimization for expression purposes, or that modifies a codon in such a way as to result in a modification of the encoded amino acid sequence. Mutations can be introduced into a polynucleotide through any number of methods known to those of ordinary skill in the art, including random mutagenesis, site-specific mutagenesis, oligonucleotide directed mutagenesis, gene shuffling, directed evolution techniques, combinatorial mutagenesis, site saturation mutagenesis among others.

[0070] “Mutation” or “mutated” means, in the context of a protein, a modification to the amino acid sequence resulting in a change in the sequence of a protein with reference to a precursor protein sequence. A mutation can refer to a substitution of one amino acid with another amino acid, an insertion or a deletion of one or more amino acid residues. A mutation can also be a truncation (e.g., a deletion or interruption) in a sequence or a subsequence from the precursor sequence. A mutation may also be an addition of a subsequence (e.g., two or more amino acids in a stretch, which are inserted between two contiguous amino acids in a precursor protein sequence) within a protein, or at either terminal end of a protein, thereby increasing the length of (or elongating) the protein. A mutation can be made by modifying the DNA sequence corresponding to a precursor protein. Mutations can be introduced into a protein sequence by known methods in the art, for example, by creating synthetic DNA sequences that encode the mutation with reference to precursor proteins, or chemically altering the protein itself. A “mutant” as used herein is a protein comprising a mutation.

[0071] A “naturally-occurring equivalent,” in the context of the present invention, refers to a naturally occurring gene or protein, or a portion thereof that comprises a naturally occurring residue.

[0072] The term “operably linked,” in the context of a polynucleotide sequence, refers to the placement of one polynucleotide sequence into a functional relationship with

another polynucleotide sequence. For example, a DNA encoding a secretory leader (e.g., a signal peptide) is operably linked to a DNA encoding a polypeptide if it is expressed as a preprotein that participates in the secretion of the polypeptide. A promoter or an enhancer is operably linked to a coding sequence if it affects the transcription of the sequence. A ribosome binding site is operably linked to a coding sequence if it is positioned so as to facilitate translation. Generally, “operably linked” means that the DNA sequences being linked are contiguous, and, in the case of a secretory leader, contiguous and in the same reading frame.

[0073] The term “optimal alignment” refers to the alignment giving the highest overall alignment score.

[0074] “Overexpressed” or “overexpression” in a host cell occurs if the enzyme is expressed in the cell at a higher level than the level at which it is expressed in a corresponding wild-type cell.

[0075] The terms “percent sequence identity,” “percent amino acid sequence identity,” “percent gene sequence identity,” and/or “percent polynucleotide sequence identity,” with respect to two polypeptides, polynucleotides and/or gene sequences (as appropriate), refer to the percentage of residues that are identical in the two sequences when the sequences are optimally aligned. Thus, 80% amino acid sequence identity means that 80% of the amino acids in two optimally aligned polypeptide sequences are identical.

[0076] A “promoter” is a polynucleotide sequence that functions to direct transcription of a downstream coding sequence. In preferred embodiments, the promoter is appropriate to the host cell in which the target coding sequence is being expressed. The promoter, together with other transcriptional and translational regulatory polynucleotide sequences (also termed “control sequences”) is necessary to express a given coding sequence in a gene. In general, the transcriptional and translational regulatory sequences include, but are not limited to, promoter sequences, ribosomal binding sites, transcriptional start and stop sequences, translational start and stop sequences, and enhancer or activator sequences.

[0077] The terms “protein” and “polypeptide” are used interchangeably herein. The 3-letter code as well as the 1-letter code for amino acid residues as defined in conformity with the IUPAC-IUB Joint Commission on Biochemical Nomenclature (JCBN) is used throughout this disclosure. It is also understood that a polypeptide may be coded for by more than one polynucleotide sequence due to the degeneracy of the genetic code.

[0078] The term “recombinant,” when used to modify the term “gene” or “protein” herein, is used synonymously with “genetically modified” and refers to a gene or protein comprising a heterologous (i.e., non-native or non-naturally occurring) sequence. The term “recombinant,” when used to modify the term “cell” herein, is used synonymously with “genetically modified” and refers to a cell that has been modified to comprise a heterologous polynucleotide sequence, or that the cell is derived from a cell so modified. Thus, for example, recombinant cells express genes that are not found in identical form within the native (non-recombinant) form of the cells or express, as a result of deliberate human intervention, native genes that are otherwise abnormally expressed, underexpressed or not expressed at all. The terms “recombination,” “recombining,” and generating a “recombined” polynucleotide refer generally to the assem-

bly of two or more polynucleotide fragments wherein the assembly gives rise to a chimeric polynucleotide made from the assembled parts.

[0079] The terms “regulatory segment,” “regulatory sequence,” or “expression control sequence” refer to a polynucleotide sequence that is operatively linked with another polynucleotide sequence that encodes the amino acid sequence of a polypeptide chain to effect the expression of that encoded amino acid sequence. The regulatory sequence can inhibit, repress, promote, or even drive the expression of the operably linked polynucleotide sequence encoding the amino acid sequence.

[0080] The term “substantially identical,” in the context of two polynucleotides or two polypeptides refers to a polynucleotide or polypeptide that comprises at least 70% sequence identity, for example, at least 75%, at least 80%, at least 85%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% sequence identity as compared to a reference sequence using the programs or algorithms (e.g., BLAST, ALIGN, CLUSTAL) using standard parameters.

[0081] “Substantially purified” means molecules that are at least about 60% free, preferably at least about 75% free, about 80% free, about 85% free, and more preferably at least about 90% free from other components with which they are naturally associated. As used herein, the term “purified” or “to purify” also refers to the removal of contaminants from a sample.

[0082] “Substitution” means replacing an amino acid in the sequence of a precursor protein with another amino acid at a particular position, resulting in a mutant of the precursor protein.

[0083] The term “transformed” or “stably transformed” cell refers to a cell that has a non-native (heterologous) polynucleotide sequence integrated into its genome or as an episomal plasmid that is maintained for at least two generations.

[0084] “Variant” is used interchangeably herein with “mutant.”

[0085] “Vector” refers to a polynucleotide construct designed to introduce polynucleotides into one or more cell types. Vectors include cloning vectors, expression vectors, shuttle vectors, plasmids, cassettes and the like. In some embodiments, the polynucleotide construct comprises a polynucleotide sequence encoding a thioesterase (e.g., a precursor or a mature thioesterase) that is operably linked to a suitable prosequence (e.g., a secretory pro-sequence) capable of effecting the expression of the polynucleotide or gene in a suitable host.

[0086] “Wild-type” means, in the context of gene or protein, a polynucleotide or protein sequence that occurs in nature. In some embodiments, the wild-type sequence refers to a sequence of interest that is a starting point for protein engineering.

Genetically Modified Cells

[0087] Another aspect of the invention is directed to genetically modified cells.

[0088] The genetically modified cells of the invention are cells comprising one or more copies of the genetically modified genes of the invention. Specifically, the genetically modified cells of the invention may comprise one or more genetically modified CCNT1 genes of the invention, one or

more genetically modified XPO1 genes of the invention, or one or more genetically modified CCNT1 genes of the invention and one or more genetically modified XPO1 genes of the invention. The genetically modified cells of the invention may comprise two or more, three or more, or four or more of one or both of the genetically modified CCNT1 and XPO1 genes of the invention. Each genetically modified CCNT1 and XPO1 gene present in the cell may be identical or different with respect to any other genetically modified CCNT1 and XPO1 gene(s) present in the cell.

[0089] The genetically modified CCNT1 and XPO1 genes may be incorporated in a chromosome in the cell or may be present extrachromosomally, such as on an extrachromosomal plasmid.

[0090] In some versions, the genetically modified cell is devoid of any native CCNT1 and/or XPO1 genes. Accordingly, the genetically modified cell may be devoid of any CCNT1 gene having an amino acid other than a tyrosine at a position corresponding to position 261 of SEQ ID NO:1; any XPO1 gene having an amino acid other than a threonine at a position corresponding to position 411 of SEQ ID NO:7, an amino acid other than a methionine at a position corresponding to position 412 of SEQ ID NO:7, and/or an amino acid other than a phenylalanine at a position corresponding to position 414 of SEQ ID NO:7; or any CCNT1 gene having an amino acid other than a tyrosine at a position corresponding to position 261 of SEQ ID NO:1 and any XPO1 gene having an amino acid other than a threonine at a position corresponding to position 411 of SEQ ID NO:7, an amino acid other than a methionine at a position corresponding to position 412 of SEQ ID NO:7, and/or an amino acid other than a phenylalanine at a position corresponding to position 414 of SEQ ID NO:7.

[0091] The genetically modified CCNT1 and/or XPO1 genes may replace one, some, or all of the native CCNT1 and/or XPO1 genes in the cell. In some versions, one, some or all of the native CCNT1 and/or XPO1 genes in the cell are directly edited to generate the genetically modified CCNT1 and/or XPO1 genes of the invention. The native genes can be edited using any gene editing tools known in the art, including CRISPR/Cas9, TALENS, etc. Exemplary methods of editing native CCNT1 and XPO1 genes to genetically modified CCNT1 and XPO1 genes of the invention are provided in the following examples.

[0092] The genetically modified cell may be a mammalian cell. In some versions, the cell is a primate cell. In some versions, the cell is a simian cell. In some versions, the cell is a human cell. In some versions, the cell is a non-human simian cell. In some versions, the cell is a feline cell. In some versions, the cell is a bovine cell.

[0093] The genetically modified cell may be a primary cell or may be an immortalized or transformed cell from a cell line.

[0094] The genetically modified cell may be an immune cell or a precursor of an immune cell. Exemplary immune cells (in various levels of generality) include white blood cells, leukocytes, lymphocytes, granulocytes, agranulocytes, myeloid cells, lymphoid cells, innate lymphoid cells, neutrophils, eosinophils (acidophilus), basophils, lymphocytes, monocytes, B cells, T cells, natural killer cells, macrophages, Kupffer cells, dendritic cells, mast cells, CD4+ T cells, CD8+ T cells, $\gamma\delta$ T cells, regulatory (suppressor) T cells. Markers for the above-referenced immune cells are well known in the art.

[0095] “Precursor” as applied to a particular cell type herein refers to a cell capable of differentiating (whether in vivo, in vitro, or ex vivo) into a particular given cell. Exemplary immune cell precursors include hematopoietic stem cells, pluripotent stem cells, multipotent progenitors, myeloid progenitors, lymphoid progenitors, myeloblasts, monocytes, small lymphocytes, B cell progenitors, and T cell progenitors. Markers for the above-referenced cells are well known in the art.

[0096] In some versions of the invention, the genetically modified cell is a T cell or a precursor thereof. Exemplary T cells include CD4+ T cells, CD8+ T cells, $\gamma\delta$ T cells, regulatory (suppressor) T cells. Exemplary precursors of T cells include hematopoietic stem cells, pluripotent stem cells, multipotent progenitors, lymphoid progenitors, and T cell progenitors. Markers for the above-referenced cells are well known in the art.

[0097] In some versions, the genetically modified cell may be a neuron or a precursor of a neuron and/or a glial cell or a precursor of a glial cell. In some versions, the genetically modified cell may be an astrocyte.

[0098] In some versions of the invention, the genetically modified cell is of a cell type susceptible to infection with a virus or a precursor of a cell type susceptible to infection with a virus. The phrase “of a cell type susceptible to infection with a virus” as applied to a particular genetically modified cell means that the cell type in unmodified form is susceptible to infection with the virus, whether or not the particular genetically modified cell itself is susceptible to infection with the virus. The term “susceptible” in the phrase “of a cell type susceptible to infection with a virus” means that the cell is capable of being infected with a virus. The term “infected” in the phrase “of a cell type susceptible to infection with a virus” means that the virus is capable of entering a cell of the cell type and, at least in the case of retroviruses, integrating part or all of its genome (in DNA form) into the cell’s genome as a provirus.

[0099] In some versions, the virus to which the cell type of the genetically modified cell is susceptible to infection is a lentivirus. In some versions, the lentivirus is a primate immunodeficiency virus. Exemplary primate immunodeficiency viruses to which the cell type of the genetically modified cell is susceptible to infection include human immunodeficiency virus (HIV), such as HIV-1 and HIV-2, and simian immunodeficiency virus (SIV). In some versions, the lentivirus is a feline immunodeficiency virus. In some versions, the lentivirus is a bovine immunodeficiency virus.

[0100] In some versions, the virus to which the cell type of the genetically modified cell is susceptible to infection is a deltaretrovirus. In some versions, the deltaretrovirus is a primate T-lymphotropic virus. Exemplary primate T-lymphotropic viruses to which the cell type of the genetically modified cell is susceptible to infection include human T-lymphotropic virus (HTLV), including HTLV-1, HTLV-2, HTLV-3, and HTLV-4, and simian T-lymphotropic virus (STLV), including STLV-1, STLV-2, STLV-3, and STLV-5.

Methods of Treatment

[0101] Another aspect of the invention is directed to methods of treating subjects infected with a lentivirus. The methods include introducing a genetically modified cell of the invention in a subject infected with a lentivirus.

[0102] The lentivirus to which the treated subject is infected may comprise any lentivirus, including any of those explicitly described herein.

[0103] The term “introduce” used with respect to treating a subject encompasses introducing genetically modified cells generated outside the body of the subject (in vitro or ex vivo) into the body, as well as generating genetically modified cells inside the body of the subject (in vivo). In some versions, the introducing comprises introducing the cell into the bloodstream of the subject. In some versions, the introducing comprises injecting or infusing the cell into the bloodstream of the subject.

[0104] The genetically modified cell introduced in the subject may comprise any genetically modified cell of the invention.

[0105] The genetically modified cell introduced in the subject is preferably of a cell type susceptible to infection with the lentivirus or a precursor of a cell type susceptible to infection with the lentivirus.

[0106] The genetically modified subject may be a mammal. In some versions, the subject is a primate. In some versions, the subject is a simian. In some versions, the subject is a human. In some versions, the subject is a non-human simian. In some versions, the subject is a feline. In some versions, the subject is a bovine. In some versions, the subject is a canine.

[0107] The genetically modified cell may be a mammalian cell. In some versions, the cell is a primate cell. In some versions, the cell is a simian cell. In some versions, the cell is a human cell. In some versions, the cell is non-human simian cell. In some versions the cell is a feline cell. In some versions the cell is a bovine cell. In some versions, the cell is a canine cell.

[0108] In some versions, the genetically modified cell is autologous to the treated subject. In some versions, the genetically modified cell is non-autologous to the treated subject.

[0109] The terms “treating,” or “ameliorating” and similar terms used herein may include prophylaxis and full or partial treatment. The terms may also include reducing symptoms, ameliorating symptoms, reducing the severity of symptoms, reducing the incidence of the disease, or any other change in the condition of the patient, which improves the therapeutic outcome. In some versions of the invention, the treating comprises increasing the proportion of genetically modified cells in the subject over a period of time. The period of time may comprise from 1 day, to a month, several months, or a year or more. In some versions of the invention, the treating comprises reducing the viral load of the lentivirus in the subject.

[0110] Some versions of the invention comprise isolating a cell from the subject, genetically modifying a native CCNT1 and/or XPO1 gene in the cell to generate a genetically modified cell of the invention, and introducing the genetically modified cell in the subject. Some versions may further comprise expanding the genetically modified cells ex vivo prior to introducing the expanded genetically modified cells in the subject. In exemplary versions, the subject is a human, the lentivirus is a primate immunodeficiency virus, such as HIV-1 or HIV-2, and the cell is a CD4+ T cell.

[0111] Methods for isolating cells from a subject, expanding the cells ex vivo after genetic modification, and introducing the expanded cells in the subject are well known in the art. See Trickett et al. 2002 (Trickett A E, Kwan Y L,

Cameron B, Dwyer J M. Ex vivo expansion of functional T lymphocytes from HIV-infected individuals. *J Immunol Methods*. 2002 Apr. 1; 262(1-2):71-83), Lieberman et al. 1997 (Lieberman J, Skolnik P R, Parkerson G R 3rd, Fabry J A, Landry B, Bethel J, Kagan J. Safety of autologous, ex vivo-expanded human immunodeficiency virus (HIV)-specific cytotoxic T-lymphocyte infusion in HIV-infected patients. *Blood*. 1997 Sep. 15; 90(6):2196-206), van Lunzen et al. 2007 (van Lunzen J, Glaunsinger T, Stahmer I, von Baehr V, Baum C, Schilz A, Kuehlcke K, Naundorf S, Martinius H, Hermann F, Giroglou T, Newrzela S, Müller I, Brauer F, Brandenburg G, Alexandrov A, von Laer D. Transfer of autologous gene-modified T cells in HIV-infected patients with advanced immunodeficiency and drug-resistant virus. *Mol Ther*. 2007 May. 15(5):1024-33), Tebas et al. 2014 (Tebas P, Stein D, Tang W W, Frank I, Wang S Q, Lee G, Spratt S K, Surosky R T, Giedlin M A, Nichol G, Holmes M C, Gregory P D, Ando D G, Kalos M, Collman R G, Binder-Scholl G, Plesa G, Hwang W T, Levine B L, June CH. Gene editing of CCR5 in autologous CD4 T cells of persons infected with HIV. *N Engl J Med*. 2014 Mar. 6; 370(10):901-10), von Laer et al. 2006, (von Laer, D, Hasselmann, S and Hasselmann, K (2006). Gene therapy for HIV infection: what does it need to make it work? *J Gene Med* 8: 658-667), and Levine et al. 2006 (Levine, B L, Humeau, L M, Boyer J, Macgregor, R R, Rebello, T, Lu, X et al. (2006). Gene transfer in humans using a conditionally replicating lentiviral vector. *Proc Natl Acad Sci USA* 103: 17372-17377).

[0112] An exemplary method for isolating cells from a subject, expanding the cells ex vivo after genetic modification, and introducing the expanded cells is as follows. Patients undergo lymphapheresis, and about 1.0×10^{10} or more mononuclear cells are collected. After overnight storage, cells are washed with a CytoMate device (Baxter, Heidelberg, Germany) and incubated with magnetic beads labeled with anti-CD8 antibodies (Miltenyi Biotech, Bergisch-Gladbach, Germany) for 30 minutes. After a second wash step, CD8+ cells are depleted using the CliniMacs (Miltenyi Biotech). A maximum of 2.5×10^8 CD3+ cells are then incubated with anti-CD³/_aanti-CD28-coated Xcyte Dynabeads (Xcyte Therapies, Seattle, Wash.) at a CD3+cell to bead ratio of 1:3 for 30 minutes on a lab rotator. Labeled cells are then enriched via the MaxSep permanent magnet (Baxter) and carefully resuspended in X-Vivo 15 medium (Cambrex) complemented with 100 U/ml rhIL-2 (Chiron, Munich, Germany), 2 mM L-glutamine (Cambrex), 5% human serum (Cambrex), and 20 mM HEPES (Invitrogen, Karlsruhe, Germany) at a cell density of 5×10^5 cells/ml and seeded into tissue culture bags (Baxter). A mixture of antivirals (1 μ M nelfinavir (Viracept), Roche, Basel, Switzerland; 0.33 μ M amprenavir (Agenerase), GlaxoSmith-Kline, Munich, Germany; 10 μ g/ml T-20 (Fuzeon), Roche) are added to the cell suspension to avoid viral replication. After 4 days of culture at 37° C. and 5% CO₂, Xcyte Dynabeads are removed from the cell suspension. Cells are then subject to gene editing to generate the genetically modified cells of the invention. After gene editing, the cells are expanded for a maximum of 7 days in a static culture until the required cell number is achieved. Finally, the remaining Xcyte Dynabeads are removed and cells are harvested with a Cyto-Mate device and cryopreserved in dimethyl sulfoxide (WAK Chemie, Steinbach, Germany), PlasmaLyte A (Baxter), Plasmasteril (6% hydroxyethyl

starch; Fresenius Kabi, Bad Homburg, Germany), and human serum albumin (20%, Baxter) for long-term storage. The genetically modified cells are infused in the patient in an amount of from about 1×10^8 to about 1×10^{12} , such as from about 1×10^9 to about 1×10^{11} . Amounts above and below these amounts are also acceptable.

[0113] The elements and method steps described herein can be used in any combination whether explicitly described or not.

[0114] All combinations of method steps as used herein can be performed in any order, unless otherwise specified or clearly implied to the contrary by the context in which the referenced combination is made.

[0115] As used herein, the singular forms “a,” “an,” and “the” include plural referents unless the content clearly dictates otherwise.

[0116] Numerical ranges as used herein are intended to include every number and subset of numbers contained within that range, whether specifically disclosed or not. Further, these numerical ranges should be construed as providing support for a claim directed to any number or subset of numbers in that range. For example, a disclosure of from 1 to 10 should be construed as supporting a range of from 2 to 8, from 3 to 7, from 5 to 6, from 1 to 9, from 3.6 to 4.6, from 3.5 to 9.9, and so forth.

[0117] All patents, patent publications, and peer-reviewed publications (i.e., “references”) cited herein are expressly incorporated by reference to the same extent as if each individual reference were specifically and individually indicated as being incorporated by reference. In case of conflict between the present disclosure and the incorporated references, the present disclosure controls.

[0118] It is understood that the invention is not confined to the particular construction and arrangement of parts herein illustrated and described, but embraces such modified forms thereof as come within the scope of the claims.

EXAMPLES

Editing Host Factors to Silence HIV Gene Expression

Methods

[0119] Cell lines and cell culture. Jurkat E6.1 T-lymphocyte (J.E6-1) cells were obtained from the American Type Culture Collection (ATCC) and were cultured in RPMI 1640 medium (Gibco) supplemented with 10% fetal bovine serum (FBS, Sigma), 1% L-glutamine (Sigma), and 1% penicillin-streptomycin antibiotics. 293T cells were cultured in Dulbecco’s modified Eagle’s medium (DMEM) supplemented with 10% fetal bovine serum, 1% L-glutamine and penicillin-streptomycin antibiotics. Cells were maintained at 37° C. and 5% CO₂ in a humidified incubator.

[0120] Gene Editing. CRISPR-Cas9 and homology directed repair (HDR) were used to edit hCCNT1 and hXPO1 to generate genetically modified CCNT1 and XPO1 genes of the invention.

[0121] The method used for editing hCCNT1 is depicted in FIG. 4. The HDR donor template for editing hCCNT1 had a sequence represented by SEQ ID NO:13

(SEQ ID NO: 13)
 GTGTTTTTTTATTATTAGTAAATTACCTAAGTAAAGAGATGCTATTTGCTTCA
 TTGCAGGCgTAcGAaGCTGCCAAGAAAACAAAAGCAGATGACCGAGGAACA
 GATGAAAAGACTTCAGA

[0122] Methods for editing hXPO1 are depicted in FIGS. 5A and 5B. These methods used two different donor templates. The donor template used in the method depicted in FIG. 5A had a sequence represented by SEQ ID NO:14:

(SEQ ID NO: 14)
 TGCTTTCTGGAAGTCAACATTTGATGTTCCCTCCAGGAGACAGCTGTAT
 TTGACTGTGTTATCAAAAGTAAACAGAGCGGITGGITGAGTGTCTTCCTG
 TTGCATACTGTGGTTTTGA

The donor template used in the method depicted in FIG. 5B had a sequence represented by SEQ ID NO:15:

(SEQ ID NO: 15)
 ATTCTCTACATCTGCgTCTCCGTTGCTTTCTGGAAGTCAACATTTGATGT
 TCCTCCAGGAGACAGCTgTATTTGaccgtgtatcaAAGGTAACAGAGCG
 GTTGcTTGAGTGTCTT

[0123] Alt-R™ recombinant S.p. Cas9 nuclease-3NLS (IDT, #1074181), Alt-R™ CRISPR-Cas9 crRNA (IDT, custom ordered), ATTO™-550 labeled Alt-R™ tracrRNA (IDT, #1075928), and Alt-R™ Cas9 Electroporation Enhancer reagent (IDT, #1075915), and nuclease-free IDTE pH 7.5 buffer (IDT, #11-01-02-02) were prepared according to the manufacturer's instructions as described in Integrated DNA Technologies User Guide ("Alt-R™ CRISPR-Cas9 System: Delivery of ribonucleoprotein complexes in Jurkat T cells using Neon® Transfection System," published at www.idtdna.com). The indicated 119-nt single-stranded oligodeoxynucleotide (ssODN) templates for homology-directed repair (HDR) were custom ordered (Sigma) and prepared as a 100 μM stock solution in TE buffer.

[0124] Jurkat cell culture preparations, crRNA:tracrRNA duplex preparations, and ribonucleoprotein (RNP) complex preparations were performed according to the reagent manufacturer's instructions in the Integrated DNA Technologies User Guide. The electroporations and final delivered material mixtures were performed according to the manufacturer's instructions in the Integrated DNA Technologies User Guide with slight modification to include ssODN HDR donor templates.

[0125] For each electroporation reaction, the crRNA:tracrRNA duplexes were assembled by combining 2.2 μL 200 μM crRNA stock, 2.2 μL 200 μM tracrRNA stock, and 5.6 nuclease-free IDTE buffer for a final volume of 10 μL. Combined reagents were heated to 95° C. for 5 minutes in a bench-top thermocycler and removed to passively cool to room temperature.

[0126] For each electroporation reaction, the ribonucleoprotein (RNP) complexes were assembled by combining 0.3 μL rCas9 and 0.2 μL resuspension buffer R for a final volume of 0.5 μL which was subsequently mixed with 0.5 μL of the prepared crRNA:tracrRNA duplex mixtures. This 1 μL total RNP mixture was incubated for approximately 15 minutes at room temperature.

[0127] For each electroporation reaction, 5×10⁵ Jurkat cells were washed with 1× PBS and resuspended in 8 μL resuspension buffer R (Invitrogen). 8 μL cell suspensions were combined with 1 μL of the prepared total RNP mixture, 2 μL of the prepared 10.8 μM Electroporation Enhancer, and 1 μL 100 μM ssODN HDR template for a total of 12 μL total. Negative controls for genome editing were included by substituting the crRNA:tracrRNA duplexes from the total RNP mixture with 10 μL nuclease-free IDTE buffer.

[0128] These reagents were delivered to JE6-1 cells using the Neon® Transfection System and Neon® Transfection 10 μL Kit (Invitrogen) according to manufacturer's instructions in the Integrated DNA Technologies User Guide. Electroporation parameters were 1600 V, 10-ms pulse width, 3 pulses and electroporated cells were cultured post-electroporation in pre-warmed antibiotic-free media (RPMI 1640 supplemented with 10% FBS) according to the manufacturer's instructions. Cells were subsequently either bulk-sorted by fluorescence-associated cell sorting (FACS) to concentrate ATTO™-550 positive cells in antibiotic-replete media (RPMI 1640 supplemented with 10% FBS and 1% penicillin-streptomycin-L-glutamine) or unsorted cell cultures were directly screened for ssODN-mediated HDR. Cell populations exhibiting positive HDR sequences based on described screening strategies were subsequently single-cell cloned, screened, and subsequently analyzed.

[0129] Cell proliferation assays. 5.0×10⁴ cells per 1 mL were plated in 12-well tissue culture dishes and maintained under normal (37° C./5% CO₂) culture conditions. At 4 and 6 days post-plating, cells were resuspended to homogenized suspensions and stained with trypan blue (Sigma) to label any dead cells with unstained cells and enumerated using a hemacytometer.

[0130] Preparation of virus stocks. 2-color HIV-1 latency reporter virus ("Dual Reporter Virus") stocks were generated by co-transfecting 293T producer cells using polyethylenimine (PEI; catalog no. 23966. Polysciences, Inc.) and the following plasmids at a 4:2:1 ratio: pE/EF1a-mChe/eGFP reporter (Calvanese et al. 2013), 2000 ng psPax2, and 1000 ng pMD.G encoding VSV-G (Ory et al. 1996). Media was exchanged 6 h post-transfection with cell culture supernatants harvested at 48 h, filtered to prevent cell contamination, aliquoted, and stored at -20° C.

[0131] For single-round primate lentivirus reporter stocks, 293T cells were co-transfected with plasmids encoding HIV-2.ROD and SIVagm.Tan-1 Env-deficient eGFP-encoding lentiviral reporters (Kane et al. 2013) at a 9:1 ratio with pMD.G encoding VSV-G.

[0132] 2-color HIV-1 gene expression reporter virus ("Early-Late Reporter Virus") stocks were generated by co-transfecting 293T producer cells using PEI, as above, and the following plasmids at a 4:4:1 ratio: pNL4-3 E-R/Gag (MA-mVenus-CA)/mChe (Knoener et al., 2017; 3×CFP gene cassette exchanged for a single mVenus reporter gene using standard molecular cloning techniques), psPax2, and pMD.G encoding VSV-G.

[0133] For all virus preparation transfections, media was exchanged at approximately 4 hours post-transfection with cell culture supernatants harvested at approximately 48 hours post-transfection, filtered to prevent cell contamination, aliquoted, and stored at ~20° C.

[0134] Viral infectivity and gene expression assays. For reporter virus experiments, 1.0×10⁶ cells were infected with equivalent amounts of virus at a multiplicity of infection of

~0.5, with cells transferred to microcentrifuge tubes at 24 hours post-infection, pelleted by centrifugation (500×G for 10 min at RT), washed with 1× PBS, and resuspended in fresh medium. 36 h post-treatment with DMSO, cells were transferred to fresh microcentrifuge tubes and pelleted by centrifugation (500×G for 10 min at RT). Supernatants were removed and cells were subsequently washed twice with 1× PBS, stained with Ghost 780 cell viability dye (Tonbo Biosciences) and washed according to the manufacturer's instructions, fixed with 4% reconstituted paraformaldehyde (PFA) and washed thrice with 1× PBS. Cells were analyzed using an analysis flow cytometer (LSRII, BD biosciences) gating for single, viable cells.

[0135] For early-late reporter virus experiments, 1.0×10^6 cells were infected with equivalent amounts of virus at a multiplicity of infection of ~0.1 or ~0.5. 48 hours post-infection, cells were transferred to microcentrifuge tubes and pelleted by centrifugation (500×G for 10 min at RT). Supernatants were removed and cells were subsequently washed twice with 1× PBS, stained with Ghost 780 cell viability dye (Tonbo Biosciences, San Diego, Calif.), and washed according to the manufacturer's instructions, fixed with 4% reconstituted paraformaldehyde (PFA) and washed thrice with 1× PBS. Cells were analyzed using an analysis flow cytometer (Attune N×T, Thermo Fisher Scientific, Waltham, Mass.) gating for single, viable cells.

[0136] All flow cytometry plots and gated cell statistics were generated using flow cytometry analysis software (FlowJo, www.flowjo.com).

[0137] Transfection-based Tat activity assay. For transient promoter activation assays, Jurkat cells (5.0×10^5 cells per well) were transfected using the Neon electroporation system (Invitrogen) following manufacturer's instructions using 1600 volts, a pulse width of 10 ms and 3 pulses. Each transfection mix consisted of 75 ng of plasmid encoding an HIV-1 U3 Tat/TAR-responsive secreted gaussia luciferase (gLuc) reporter (Nekhai et al. 2006), 250 ng of pmCherry expression plasmid (pmCherry-C1, Takara Bio), 75 ng of a *cypridina* expression plasmid (tk-Cluc, New England Biolabs, NEB) with or without plasmids encoding CCNT1 variants or Tat expression plasmids at 1200 and 25 ng/well, respectively. Vector plasmid DNA or Calf thymus DNA (NEB) was used to maintain a constant 2.5 plasmid DNA per transfection. 24 hours post-transfection, 10 μl of media was removed, diluted with 40 μl of PBS and assayed for secreted gaussia luciferase (gLuc) by injecting 30 μl coelenterazine solution (Renilla luciferase assay system, Promega, Madison, Wis.), waiting 1.6 s and then reading luminescence for 1 s. Secreted cypridina luciferase (cLuc) activity from the internal control plasmid was determined using the cypridina Luciferase kit (NEB) according to the manufacturer's instructions using the same injection conditions as for gLuc. The activity of the retroviral promoter in each well was then determined as the ratio of gLuc: cLuc.

[0138] Analysis of genomic DNA modifications to CCNT1 and XPO1. Genomic DNA was extracted from prepared bulk heterogeneous or clonal Jurkat cell lines. Briefly, Jurkat cells were washed with phosphate-buffered saline (PBS) in microcentrifuge tubes, resuspended in 10 μl 1× polymerase chain reaction (PCR) buffer (GoTaq Green Buffer, Promega) in standard PCR tubes, and subjected to a single freeze-thaw cycle at -80° C. 1 μl proteinase K (New England Biolabs, NEB) were added to each tube and incubated at 65° C. for 60 min, 95° C. for 15 min, and were

maintained at 4° C. During 4° C. hold, the remaining 40 μl for a 50 μl PCR reaction (GoTaq Flexi Kit, Promega) were added to each tube, using the following CCNT1 or XPO1 primer sets:

[0139] CCNT1 forward screening primer: 5'-TGA GAT TAG AAG TAG GCT TGA GAG G-3' (SEQ ID NO:16). CCNT1 reverse screening primer: 5'-GCT AAA TTC TCA CTA GTC CGA TGA C-3' (SEQ ID NO:17). XPO1 forward screening primer: 5'-TTC TCT CCT CTG TGA TGG TAC ATT T-3' (SEQ ID NO:18). XPO1 reverse screening primer: 5'-TCA AGA TTG TAG TGA GCT ATG ACC A-3' (SEQ ID NO:19).

[0140] CCNT1 or XPO1 genomic loci amplicons were amplified using the following PCR cycle conditions: CCNT1 PCR cycle conditions: 98° C. for 2 min, 98° C. for 15 sec, 66° C. for 45 sec, 72° C. for 2 min, repeat steps 2-4 an additional 35 times, 72° C. for 10 min, 4° C. hold. XPO1 PCR cycle conditions: 98° C. for 2 min, 98° C. for 15 sec, 60° C. for 45 sec, 72° C. for 2 min, repeat steps 2-4 an additional 35 times, 72° C. for 10 min, 4° C. hold.

[0141] Restriction enzyme digestion reactions containing candidate CCNT1 genomic DNA amplicons were carried out following the manufacturer's recommended protocol with BsiWI-HF enzyme (NEB) or no enzyme controls. Predicted BsiWI digestion products were based on the FIG. 4 design scheme: 712 bp and 288 bp. Restriction enzyme digestion reactions containing candidate XPO1 genomic DNA amplicons were carried out following the manufacturer's recommended protocol with PvuII enzyme (NEB) or no enzyme controls. Predicted PvuII digestion products were based on the FIG. 5A or FIG. 5B design scheme: 497 bp and 480 bp.

[0142] DNA amplicons and/or DNA products following restriction enzyme digestion were resolved using standard agarose gel electrophoresis.

Results

[0143] The invention encompasses the generation of primary mammalian cells or cell lines wherein orthologs of conserved genes known to regulate human immunodeficiency virus type 1 (HIV-1) gene expression are altered at their native loci within chromosomes in order to render the cells intrinsically resistant to HIV-1 replication in vitro and in vivo. This strategy also blocks replication of other important human retroviral pathogens including HIV type 2 (HIV-2) and human T lymphotropic virus types 1 and 2 (HTLV-1 and HTLV-2), as well as related retroviruses of the genera lentiviridae and deltaretoviridae that cause immunodeficiency, cancers, or other diseases in other animals. The invention is premised on our discovery that blocks to HIV-1 replication observed in mice can be made manifest in human cells using a gene knock-in strategy, with little to no discernable effect on host biology.

[0144] In people infected with HIV-1, the human CCNT1 (hCCNT1) transcription factor is recruited by the viral Tat protein to the viral promoter in order to activate robust viral mRNA transcription (Nekhai et al. 2006, Wei et al. 1998) (FIG. 3, panel A). By contrast, in mice mouse CCNT1 (mCcnt1) binds poorly to Tat, a defect previously mapped to a single species-specific amino acid (tyrosine at position 261 instead of cysteine as found in hCCNT1; a difference herein referred to as "C261Y") (Bieniasz et al. 1998, Garber et al. 1998) (FIG. 3, panel B). Similarly, the human XPO1 (hXPO1) nuclear export receptor is recruited by the viral

Rev protein to intron-retaining viral mRNAs to mediate their nuclear export and hence ensure late stage gene expression needed to accomplish infectious virion production (Fornerod et al. 1997, Neville et al. 1997, Pollard et al. 1998) (FIG. 3, panel A). By contrast, mouse XPO1 (mXpo1) interacts poorly with Rev/RNA complexes due to a species-specific cluster of three mXpo1-specific amino acids; threonine-411 instead of proline, valine-412 instead of methionine, and serine-414 instead of phenylalanine (Elinav et al. 2012, Sherer et al. 2011) (FIG. 3, panel C). Thus, in mouse cells HIV-1 is unable to express viral gene products and infectious virus particles cannot be generated.

[0145] To determine if naturally-occurring, species-specific genetic blocks to HIV-1 gene expression can be made manifest in human cells, we designed and engineered CRISPR/Cas9 clonal human Jurkat E6-1 T cell lines carrying homozygous hCCNT1-C261Y alleles and compared them to wild-type parental cells in HIV-1 gene expression assays. A depiction of how the hCCNT1-C261Y Jurkat cells were modified is shown in FIGS. 4 and 6. A depiction of how cells can similarly be modified to encode hXPO1-P411T-M412V-F414S is shown in FIGS. 5 and 6. FIG. 6 also shows how two or more gene edits (e.g., modifying both hCCNT1 and hXPO1) can be multiplexed to block multiple stages of the HIV-1 replication cycle in the same cell. Two hCCNT1-C261Y cell lines (clones 4.7F and 4.8C) were isolated and characterized. These cells proliferated identically to parental cells, thus demonstrating that the C261Y hCCNT1 codon change has no effect on basic features of cellular metabolism (FIG. 7, panel A). In contrast, clones 4.7F and 4.8C potently restricted HIV-1 Tat-dependent gene expression (FIG. 7, panels B and C).

[0146] We confirmed that the hCCNT1-C261Y block specifically inhibited HIV-1 Tat function using two independent HIV-1 gene reporter assays (FIG. 7, panels B and C). First, wild-type and hCCNT1-C261Y cells were infected using “single-round” HIV-1 reporter viruses that report on both HIV-1 Tat-dependent gene expression (based on GFP synthesis) and Tat-independent gene expression (based on mCherry synthesis) (Calvanese et al. 2013). Similar percentages of cells for all three cell lines exhibited constitutive mCherry reporter gene expression at 2.5 days post-infection, thus confirming equivalent levels of infection (FIG. 7, panel B, red bars). By contrast, the hCCNT1-C261Y cells were highly resistant to Tat activity as illustrated by a >16-fold drop to Tat-dependent GFP expression (FIG. 7, panel B, green bars). Second, to address specificity we co-transfected wild-type or hCCNT1-C261Y cell lines with DNA gene expression plasmids encoding Tat and a Tat-responsive Luciferase reporter, with or without plasmids encoding wild-type hCCNT1 (FIG. 7, panel C). Tat activity was almost completely abolished in both hCCNT1-C261Y cell lines compared to parental cells but could be rescued by co-expressing wild-type hCCNT1 (FIG. 7, panel C). Collectively, these data demonstrate that Tat-dependent viral gene expression is largely abolished in human cells simply by altering a single codon in CCNT1 to encode tyrosine and not cysteine.

[0147] Because many if not all lentiviruses resemble HIV-1 in their dependence on Tat-CCNT1 interactions in order to activate viral gene expression, we also tested if hCCNT1-C261Y modified cells could suppress HIV-2 and the simian immunodeficiency virus of the African green monkey (SIVagm) Tat-dependent viral gene expression

using previously validated GFP reporter viruses (Kane et al. 2013) (FIG. 8). HIV-2 and SIVagm Tat-dependent viral gene expression was highly suppressed in the 4.7F hCCNT1-C261Y Jurkat T cell line. These results illustrate broad-spectrum antiviral potential for hCCNT1-C261Y gene modifications.

[0148] C261Y alleles predict similar results with hXPO1, wherein clonal human Jurkat E6-1 T cell lines carrying homozygous hXPO1 genes encoding hXPO1 with either a P411T mutation (hXPO1-P411V), a M412V mutation (hXPO1-M412V), a F414S mutation (hXPO1-F414S), P411T and M412V mutations (hXPO1-P411T-M412V), P411T and F414S mutations (hXPO1-P411T-F414S), M412V and F414S mutations (hXPO1-M412V-F414S), or P411T, M412V, and F414S mutations (hXPO1-P411T-M412V-F414S) are predicted to proliferate identically to parental cells with one or more combination of the three variant amino acids restricting HIV-1, HIV-2, and SIV late stage gene expression.

[0149] We sought to define an analytical method by which HIV-1 resistance profiles can be resolved and characterized in non-clonal T cell populations. To this end, we mixed wild-type and hCCNT1-C261Y cells at defined ratios (20:80, 50:50, and 80:20) to simulate a heterogeneous cell mixture using known clonal cell lines. Next, we infected these mixed cultures with a HIV-1 reporter virus that distinguishes between early and late viral gene expression (Early-Late Reporter Virus) and examined the HIV resistance profile at a population level by flow cytometric analyses (FIGS. 9B-D). We confirmed the relative abundance of wild-type and hCCNT1-C261Y alleles in each cell mixture by PCR and restriction enzyme digestion reactions (FIG. 9A). In this analysis, we defined a gating strategy (FIG. 9B) that enables us to resolve population-level changes to the percentages of cells expressing no viral genes, only early viral genes, or early and late viral genes (“Gate 1”, “Gate 2”, and “Gate 3”, respectively). Given the ability of hCCNT1-C261Y cells to resist HIV-1 gene expression, we observed a striking and titratable reduction to the percentages of cells expressing both early and late viral genes as the relative abundance of the resistant cell population in the mixtures is increased (FIGS. 9C and 9D). In conclusion, we present a method to quantify HIV resistance in heterogeneous cell populations using defined mixtures clonal Jurkat populations.

[0150] Using the population-level flow cytometry analysis described (FIGS. 9A-9D), we examined whether cells modified to produce hXPO1-P411T-M412V-F414S could also imbue HIV-1 resistance to Jurkat T cells. According to the design scheme presented above (FIGS. 5A and 6), we carried out CRISPR/Cas9 gene editing in a similar manner to induce hXPO1-P411T-M412V-F414S gene modifications, and infected the heterogeneous cell mixtures, or wild-type control cells, with the early-late reporter virus with two virus doses. We confirmed the presence of hXPO1-P411T-M412V-F414S modifications in the CRISPR/Cas9-treated cells by PCR and restriction enzyme digestion reactions (FIG. 10A) and evaluated the infected cell populations according to the gating strategy described for FIG. 10B. Consistent with a block to HIV-1 Rev function, we observed a reduction to the percentages of cells expressing both early and late genes and an approximately similar increase to the cells expressing only early genes (FIGS. 10 C and 10D). In

sum, we demonstrate that species-informed modifications to hXPO1 can be employed to repress HIV-1 gene expression.

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EXEMPLARY EMBODIMENTS OF THE INVENTION

[0179] Embodiment 1. A genetically modified CCNT1 gene encoding a protein comprising a sequence with at least 80%, at least 85%, at least 90%, or at least 95% sequence identity to SEQ ID NO:1 and comprising a tyrosine at a position corresponding to position 261 of SEQ ID NO:1.

[0180] Embodiment 2. The genetically modified CCNT1 gene of embodiment 1, wherein the protein encoded by the genetically modified CCNT1 gene comprises one, some, or all of: an amino acid other than glutamic acid at a position corresponding to position 3 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 29 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 31 of SEQ ID NO:1; an amino acid other than leucine and/or asparagine at a position corresponding to position 37 of SEQ ID NO:1; an amino acid other than histidine at a position corresponding to position 79 of SEQ ID NO:1; an amino acid other than arginine and glutamine and/or tyrosine at a position corresponding to position 80 of SEQ ID NO:1; an amino acid other than tyrosine at a position corresponding to position 81 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 83 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 110 of SEQ ID NO:1; an amino acid other than tyrosine at a position corresponding to position 113 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 250 of SEQ ID NO:1; an amino acid other than arginine at a position corresponding to position 256 of SEQ ID NO:1; an amino acid other than glutamine at a position corresponding to position 262 of SEQ ID NO:1; an amino acid other than methionine, arginine, and/or glutamine at a position corresponding to position 265 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 269 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 274 of SEQ ID NO:1; an amino acid other than threonine and/or alanine at a position corresponding to position 276 of SEQ ID NO:1; an amino acid other than asparagine at a position corresponding to position 277 of SEQ ID NO:1; an amino acid other than threonine at a position corresponding to position 290 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 304 of SEQ ID NO:1; an amino acid other than alanine and/or threonine at a position corresponding to position 305 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 306 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 307 of SEQ ID NO:1; an amino acid other than arginine and/or valine at a position corresponding to position 313 of SEQ ID NO:1; an amino acid

other than serine, alanine, and/or valine at a position corresponding to position 315 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 322 of SEQ ID NO:1; an amino acid other than asparagine at a position corresponding to position 325 of SEQ ID NO:1; an amino acid other than aspartic acid at a position corresponding to position 327 of SEQ ID NO:1; an amino acid other than glutamine at a position corresponding to position 330 of SEQ ID NO:1; an amino acid other than glutamic acid at a position corresponding to position 332 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 340 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 345 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 346 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 354 of SEQ ID NO:1; an amino acid other than isoleucine and/or methionine at a position corresponding to position 358 of SEQ ID NO:1; an amino acid other than glutamine at a position corresponding to position 365 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 370 of SEQ ID NO:1; an amino acid other than glycine at a position corresponding to position 373 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 378 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 443 of SEQ ID NO:1; an amino acid other than aspartic acid at a position corresponding to position 453 of SEQ ID NO:1; an amino acid other than serine and/or alanine at a position corresponding to position 458 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 464 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 468 of SEQ ID NO:1; an amino acid other than valine at a position corresponding to position 473 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 488 of SEQ ID NO:1; an amino acid other than glycine at a position corresponding to position 490 of SEQ ID NO:1; an amino acid other than isoleucine at a position corresponding to position 496 of SEQ ID NO:1; an amino acid other than glutamine at a position corresponding to position 510 of SEQ ID NO:1; an amino acid other than arginine at a position corresponding to position 511 of SEQ ID NO:1; an amino acid other than arginine at a position corresponding to position 527 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 531 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 535 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 537 of SEQ ID NO:1; an amino acid other than valine at a position corresponding to position 538 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 539 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 543 of SEQ ID NO:1; an amino acid other than threonine at a position corresponding to position 553 of SEQ ID NO:1; an amino acid other than threonine at a position corresponding to position 564 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 565 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 577 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 582 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 603 of SEQ

ID NO:1; an amino acid other than proline at a position corresponding to position 606 of SEQ ID NO:1; an amino acid other than threonine and/or alanine at a position corresponding to position 611 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 613 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 624 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 637 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 644 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 651 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 654 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 678 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 679 of SEQ ID NO:1; an amino acid other than aspartic acid at a position corresponding to position 682 of SEQ ID NO:1; an amino acid other than histidine at a position corresponding to position 685 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 686 of SEQ ID NO:1; an amino acid other than glycine at a position corresponding to position 688 of SEQ ID NO:1; an amino acid other than glutamic acid at a position corresponding to position 689 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 691 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 695 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 697 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 698 of SEQ ID NO:1; an amino acid other than threonine at a position corresponding to position 704 of SEQ ID NO:1; and an amino acid other than leucine at a position corresponding to position 710 of SEQ ID NO:1.

[0181] Embodiment 3. The genetically modified CCNT1 gene of any prior embodiment, wherein the protein encoded by the genetically modified CCNT1 gene comprises one, some or all of: proline at a position corresponding to position 31 of SEQ ID NO:1; tyrosine at a position corresponding to position 37 of SEQ ID NO:1; proline at a position corresponding to position 79 of SEQ ID NO:1; glycine at a position corresponding to position 80 of SEQ ID NO:1; asparagine at a position corresponding to position 81 of SEQ ID NO:1; valine at a position corresponding to position 83 of SEQ ID NO:1; threonine at a position corresponding to position 110 of SEQ ID NO:1; asparagine at a position corresponding to position 250 of SEQ ID NO:1; tryptophan at a position corresponding to position 256 of SEQ ID NO:1; glutamic acid at a position corresponding to position 262 of SEQ ID NO:1; lysine at a position corresponding to position 265 of SEQ ID NO:1; alanine at a position corresponding to position 269 of SEQ ID NO:1; threonine at a position corresponding to position 274 of SEQ ID NO:1; lysine at a position corresponding to position 277 of SEQ ID NO:1; serine at a position corresponding to position 290 of SEQ ID NO:1; serine at a position corresponding to position 305 of SEQ ID NO:1; threonine at a position corresponding to position 306 of SEQ ID NO:1; threonine at a position corresponding to position 307 of SEQ ID NO:1; leucine at a position corresponding to position 313 of SEQ ID NO:1; valine at a position corresponding to position 315 of SEQ ID NO:1; serine at a position corresponding to position 316 of SEQ ID NO:1; asparagine at a position corresponding to

position 322 of SEQ ID NO:1; serine at a position corresponding to position 325 of SEQ ID NO:1; glutamic acid at a position corresponding to position 327 of SEQ ID NO:1; proline at a position corresponding to position 330 of SEQ ID NO:1; lysine at a position corresponding to position 332 of SEQ ID NO:1; serine at a position corresponding to position 340 of SEQ ID NO:1; proline at a position corresponding to position 345 of SEQ ID NO:1; threonine at a position corresponding to position 346 of SEQ ID NO:1; asparagine at a position corresponding to position 354 of SEQ ID NO:1; threonine at a position corresponding to position 358 of SEQ ID NO:1; proline at a position corresponding to position 365 of SEQ ID NO:1; asparagine at a position corresponding to position 370 of SEQ ID NO:1; isoleucine at a position corresponding to position 373 of SEQ ID NO:1; asparagine at a position corresponding to position 378 of SEQ ID NO:1; histidine at a position corresponding to position 429 of SEQ ID NO:1; glycine at a position corresponding to position 443 of SEQ ID NO:1; glutamic acid at a position corresponding to position 453 of SEQ ID NO:1; threonine at a position corresponding to position 458 of SEQ ID NO:1; isoleucine at a position corresponding to position 464 of SEQ ID NO:1; glycine at a position corresponding to position 468 of SEQ ID NO:1; alanine at a position corresponding to position 473 of SEQ ID NO:1; alanine at a position corresponding to position 488 of SEQ ID NO:1; alanine at a position corresponding to position 490 of SEQ ID NO:1; valine at a position corresponding to position 496 of SEQ ID NO:1; histidine at a position corresponding to position 510 of SEQ ID NO:1; lysine at a position corresponding to position 511 of SEQ ID NO:1; lysine at a position corresponding to position 527 of SEQ ID NO:1; serine at a position corresponding to position 531 of SEQ ID NO:1; valine at a position corresponding to position 535 of SEQ ID NO:1; threonine at a position corresponding to position 537 of SEQ ID NO:1; glycine at a position corresponding to position 538 of SEQ ID NO:1; asparagine at a position corresponding to position 539 of SEQ ID NO:1; glycine at a position corresponding to position 543 of SEQ ID NO:1; asparagine at a position corresponding to position 553 of SEQ ID NO:1; serine at a position corresponding to position 564 of SEQ ID NO:1; phenylalanine at a position corresponding to position 565 of SEQ ID NO:1; serine at a position corresponding to position 577 of SEQ ID NO:1; glycine at a position corresponding to position 582 of SEQ ID NO:1; serine at a position corresponding to position 599 of SEQ ID NO:1; serine at a position corresponding to position 603 of SEQ ID NO:1; serine at a position corresponding to position 606 of SEQ ID NO:1; glycine at a position corresponding to position 611 of SEQ ID NO:1; methionine at a position corresponding to position 613 of SEQ ID NO:1; serine at a position corresponding to position 624 of SEQ ID NO:1; serine at a position corresponding to position 637 of SEQ ID NO:1; threonine at a position corresponding to position 644 of SEQ ID NO:1; threonine at a position corresponding to position 651 of SEQ ID NO:1; threonine at a position corresponding to position 654 of SEQ ID NO:1; proline at a position corresponding to position 678 of SEQ ID NO:1; threonine at a position corresponding to position 679 of SEQ ID NO:1; glutamic acid at a position corresponding to position 682 of SEQ ID NO:1; arginine at a position corresponding to position 685 of SEQ ID NO:1; proline at a position corresponding to position 686 of SEQ ID NO:1; serine at a

position corresponding to position 688 of SEQ ID NO:1; aspartic acid at a position corresponding to position 689 of SEQ ID NO:1; leucine at a position corresponding to position 691 of SEQ ID NO:1; serine at a position corresponding to position 695 of SEQ ID NO:1; glycine at a position corresponding to position 697 of SEQ ID NO:1; isoleucine at a position corresponding to position 698 of SEQ ID NO:1; asparagine at a position corresponding to position 704 of SEQ ID NO:1; and proline at a position corresponding to position 710 of SEQ ID NO:1.

[0182] Embodiment 4. A genetically modified XPO1 gene encoding a protein comprising a sequence with at least 80%, at least 85%, at least 90%, or at least 95% sequence identity to SEQ ID NO:7 and having at least one, at least two, or all three of: threonine at a position corresponding to position 411 of SEQ ID NO:7; valine at a position corresponding to position 412 of SEQ ID NO:7; and serine at a position corresponding to position 414 of SEQ ID NO:7.

[0183] Embodiment 5. The genetically modified XPO1 gene of embodiment 4, wherein the protein encoded by the genetically modified XPO1 gene comprises one, some, or all of: an amino acid other than aspartic acid at a position corresponding to position 100 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 118 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 151 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 191 of SEQ ID NO:7; an amino acid other than serine at a position corresponding to position 215 of SEQ ID NO:7; an amino acid other than glutamic acid at a position corresponding to position 284 of SEQ ID NO:7; an amino acid other than valine at a position corresponding to position 306 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 334 of SEQ ID NO:7; an amino acid other than leucine at a position corresponding to position 337 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 346 of SEQ ID NO:7; an amino acid other than isoleucine at a position corresponding to position 402 of SEQ ID NO:7; an amino acid other than isoleucine at a position corresponding to position 474 of SEQ ID NO:7; an amino acid other than lysine at a position corresponding to position 478 of SEQ ID NO:7; an amino acid other than glutamine at a position corresponding to position 481 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 869 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 909 of SEQ ID NO:7; an amino acid other than proline at a position corresponding to position 961 of SEQ ID NO:7; an amino acid other than serine at a position corresponding to position 966 of SEQ ID NO:7; an amino acid other than serine at a position corresponding to position 969 of SEQ ID NO:7; an amino acid other than valine and/or methionine at a position corresponding to position 972 of SEQ ID NO:7; an amino acid other than isoleucine at a position corresponding to position 974 of SEQ ID NO:7; an amino acid other than aspartic acid at a position corresponding to position 976 of SEQ ID NO:7; an amino acid other than threonine at a position corresponding to position 1040 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 1043 of SEQ ID NO:7; an amino acid other than glutamine at a position corresponding to position 1046 of SEQ ID NO:7; an amino acid other than leucine at a position corresponding to position 1052 of SEQ

ID NO:7; and an amino acid other than leucine at a position corresponding to position 1060 of SEQ ID NO:7.

[0184] Embodiment 6. The genetically modified XPO1 gene of any one of embodiments 4-5, wherein the protein encoded by the genetically modified XPO1 gene comprises one, some, or all of: glutamic acid at a position corresponding to position 100 of SEQ ID NO:7; threonine at a position corresponding to position 118 of SEQ ID NO:7; serine at a position corresponding to position 151 of SEQ ID NO:7; serine at a position corresponding to position 191 of SEQ ID NO:7; asparagine at a position corresponding to position 215 of SEQ ID NO:7; valine at a position corresponding to position 284 of SEQ ID NO:7; leucine at a position corresponding to position 306 of SEQ ID NO:7; aspartic acid at a position corresponding to position 334 of SEQ ID NO:7; isoleucine at a position corresponding to position 337 of SEQ ID NO:7; threonine at a position corresponding to position 346 of SEQ ID NO:7; valine at a position corresponding to position 402 of SEQ ID NO:7; arginine at a position corresponding to position 474 of SEQ ID NO:7; glutamic acid at a position corresponding to position 478 of SEQ ID NO:7; histidine at a position corresponding to position 481 of SEQ ID NO:7; threonine at a position corresponding to position 869 of SEQ ID NO:7; alanine at a position corresponding to position 909 of SEQ ID NO:7; serine at a position corresponding to position 961 of SEQ ID NO:7; asparagine at a position corresponding to position 966 of SEQ ID NO:7; asparagine at a position corresponding to position 969 of SEQ ID NO:7; isoleucine at a position corresponding to position 972 of SEQ ID NO:7; leucine at a position corresponding to position 974 of SEQ ID NO:7; glutamic acid at a position corresponding to position 976 of SEQ ID NO:7; isoleucine at a position corresponding to position 1040 of SEQ ID NO:7; arginine at a position corresponding to position 1043 of SEQ ID NO:7; aspartic acid at a position corresponding to position 1046 of SEQ ID NO:7; arginine at a position corresponding to position 1052 of SEQ ID NO:7; and phenylalanine at a position corresponding to position 1060 of SEQ ID NO:7.

[0185] Embodiment 7. A genetically modified cell comprising at least one of: one or more copies of the genetically modified gene of any one of embodiments 1-3; and one or more copies of the genetically modified gene of embodiments 4-6.

[0186] Embodiment 8. The cell of embodiment 7, wherein the cell is an immune cell or a precursor of an immune cell.

[0187] Embodiment 9. The cell of any one of embodiments 7-8, wherein the cell is selected from the group consisting of a hematopoietic stem cell, a myeloid progenitor cell, a lymphoid progenitor cell, a myeloblast, a monocyte, a macrophage, a dendritic cell, a small lymphocyte, a T cell, and an astrocyte.

[0188] Embodiment 10. The cell of any one of embodiments 7-9, wherein the cell is a T cell or a precursor thereof.

[0189] Embodiment 11. The cell of any one of embodiments 7-10, wherein the cell is a CD4⁺T cell or a precursor thereof.

[0190] Embodiment 12. The cell of any one of embodiments 7-11, wherein the cell is a mammalian cell.

[0191] Embodiment 13. The cell of any one of embodiments 7-12, wherein the cell comprises at least one of: two copies of the genetically modified CCNT1 gene; and two copies of the genetically modified XPO1 gene.

[0192] Embodiment 14. The cell of any one of embodiments 7-13, wherein the cell is devoid of at least one of: a CCNT1 gene having an amino acid other than a tyrosine at a position corresponding to position 261 of SEQ ID NO:1; and an XPO1 gene having at least one, at least two, or all three of an amino acid other than a threonine at a position corresponding to position 411 of SEQ ID NO:7, an amino acid other than a methionine at a position corresponding to position 412 of SEQ ID NO:7, and an amino acid other than a phenylalanine at a position corresponding to position 414 of SEQ ID NO:7.

[0193] Embodiment 15. A method of treating a subject infected with a virus, the method comprising introducing the genetically modified cell of any one of embodiments 7-14 in the subject, wherein the genetically modified cell is of a cell type susceptible to infection with the virus or a precursor of a cell type susceptible to infection with the virus.

[0194] Embodiment 16. The method of embodiment 15, wherein the subject is a mammal.

[0195] Embodiment 17. The method of any one of embodiments 15-16, wherein the subject is a human.

[0196] Embodiment 18. The method of any one of embodiments 15-17, wherein the virus is selected from the group consisting of a lentivirus and a deltaretrovirus.

[0197] Embodiment 19. The method of any one of embodiments 15-18, wherein the virus is selected from the group consisting of a primate immunodeficiency virus and a primate T-lymphotropic virus.

[0198] Embodiment 20. The method of any one of embodiments 15-19, wherein the virus is selected from the group consisting of a human immunodeficiency virus and a human T-lymphotropic virus.

[0199] Embodiment 21. The method of any one of embodiments 15-20, wherein the virus is a human immunodeficiency virus.

[0200] Embodiment 22. The method of any one of embodiments 15-21, wherein the cell is autologous to the subject.

[0201] Embodiment 23. The method of any one of embodiments 15-22, wherein the introducing comprises introducing the cell into the bloodstream of the subject.

[0202] Embodiment 24. The method of any one of embodiments 15-23, wherein the introducing comprises injecting or infusing the cell into the bloodstream of the subject.

SEQUENCE LISTING

<160> NUMBER OF SEQ ID NOS: 19

<210> SEQ ID NO 1

<211> LENGTH: 726

<212> TYPE: PRT

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: C261Y mutant of human CCNT1

<400> SEQUENCE: 1

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Lys Glu Leu Ser Tyr Arg Gln Gln Ala Ala Asn Leu Leu Gln Asp Met
35 40 45

Gly Gln Arg Leu Asn Val Ser Gln Leu Thr Ile Asn Thr Ala Ile Val
50 55 60

Tyr Met His Arg Phe Tyr Met Ile Gln Ser Phe Thr Gln Phe Pro Gly
65 70 75 80

Asn Ser Val Ala Pro Ala Ala Leu Phe Leu Ala Ala Lys Val Glu Glu
85 90 95

Gln Pro Lys Lys Leu Glu His Val Ile Lys Val Ala His Thr Cys Leu
100 105 110

His Pro Gln Glu Ser Leu Pro Asp Thr Arg Ser Glu Ala Tyr Leu Gln
115 120 125

Gln Val Gln Asp Leu Val Ile Leu Glu Ser Ile Ile Leu Gln Thr Leu
130 135 140

Gly Phe Glu Leu Thr Ile Asp His Pro His Thr His Val Val Lys Cys
145 150 155 160

Thr Gln Leu Val Arg Ala Ser Lys Asp Leu Ala Gln Thr Ser Tyr Phe
165 170 175

Met Ala Thr Asn Ser Leu His Leu Thr Thr Phe Ser Leu Gln Tyr Thr

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180					185					190					
Pro	Pro	Val	Val	Ala	Cys	Val	Cys	Ile	His	Leu	Ala	Cys	Lys	Trp	Ser
		195					200					205			
Asn	Trp	Glu	Ile	Pro	Val	Ser	Thr	Asp	Gly	Lys	His	Trp	Trp	Glu	Tyr
		210					215					220			
Val	Asp	Ala	Thr	Val	Thr	Leu	Glu	Leu	Leu	Asp	Glu	Leu	Thr	His	Glu
							230					235			240
Phe	Leu	Gln	Ile	Leu	Glu	Lys	Thr	Pro	Asn	Arg	Leu	Lys	Arg	Ile	Trp
				245								250			255
Asn	Trp	Arg	Ala	Tyr	Glu	Ala	Ala	Lys	Lys	Thr	Lys	Ala	Asp	Asp	Arg
				260								265			270
Gly	Thr	Asp	Glu	Lys	Thr	Ser	Glu	Gln	Thr	Ile	Leu	Asn	Met	Ile	Ser
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Gln	Ser	Ser	Ser	Asp	Thr	Thr	Ile	Ala	Gly	Leu	Met	Ser	Met	Ser	Thr
				290								295			300
Ser	Thr	Thr	Ser	Ala	Val	Pro	Ser	Leu	Pro	Val	Ser	Glu	Glu	Ser	Ser
				305								310			315
Ser	Asn	Leu	Thr	Ser	Val	Glu	Met	Leu	Pro	Gly	Lys	Arg	Trp	Leu	Ser
				325								330			335
Ser	Gln	Pro	Ser	Phe	Lys	Leu	Glu	Pro	Thr	Gln	Gly	His	Arg	Thr	Ser
				340								345			350
Glu	Asn	Leu	Ala	Leu	Thr	Gly	Val	Asp	His	Ser	Leu	Pro	Gln	Asp	Gly
				355								360			365
Ser	Asn	Ala	Phe	Ile	Ser	Gln	Lys	Gln	Asn	Ser	Lys	Ser	Val	Pro	Ser
				370								375			380
Ala	Lys	Val	Ser	Leu	Lys	Glu	Tyr	Arg	Ala	Lys	His	Ala	Glu	Glu	Leu
				385								390			395
Ala	Ala	Gln	Lys	Arg	Gln	Leu	Glu	Asn	Met	Glu	Ala	Asn	Val	Lys	Ser
				405								410			415
Gln	Tyr	Ala	Tyr	Ala	Ala	Gln	Asn	Leu	Leu	Ser	His	His	Asp	Ser	His
				420								425			430
Ser	Ser	Val	Ile	Leu	Lys	Met	Pro	Ile	Glu	Gly	Ser	Glu	Asn	Pro	Glu
				435								440			445
Arg	Pro	Phe	Leu	Glu	Lys	Ala	Asp	Lys	Thr	Ala	Leu	Lys	Met	Arg	Ile
				450								455			460
Pro	Val	Ala	Gly	Gly	Asp	Lys	Ala	Ala	Ser	Ser	Lys	Pro	Glu	Glu	Ile
				465								470			475
Lys	Met	Arg	Ile	Lys	Val	His	Ala	Ala	Ala	Asp	Lys	His	Asn	Ser	Val
				485								490			495
Glu	Asp	Ser	Val	Thr	Lys	Ser	Arg	Glu	His	Lys	Glu	Lys	His	Lys	Thr
				500								505			510
His	Pro	Ser	Asn	His	His	His	His	His	Asn	His	His	Ser	His	Lys	His
				515								520			525
Ser	His	Ser	Gln	Leu	Pro	Val	Gly	Thr	Gly	Asn	Lys	Arg	Pro	Gly	Asp
				530								535			540
Pro	Lys	His	Ser	Ser	Gln	Thr	Ser	Asn	Leu	Ala	His	Lys	Thr	Tyr	Ser
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Leu	Ser	Ser	Ser	Phe	Ser	Ser	Ser	Ser	Ser	Thr	Arg	Lys	Arg	Gly	Pro
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Ser	Glu	Glu	Thr	Gly	Gly	Ala	Val	Phe	Asp	His	Pro	Ala	Lys	Ile	Ala
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Lys Ser Thr Lys Ser Ser Ser Leu Asn Phe Ser Phe Pro Ser Leu Pro
595 600 605

Thr Met Gly Gln Met Pro Gly His Ser Ser Asp Thr Ser Gly Leu Ser
610 615 620

Phe Ser Gln Pro Ser Cys Lys Thr Arg Val Pro His Ser Lys Leu Asp
625 630 635 640

Lys Gly Pro Thr Gly Ala Asn Gly His Asn Thr Thr Gln Thr Ile Asp
645 650 655

Tyr Gln Asp Thr Val Asn Met Leu His Ser Leu Leu Ser Ala Gln Gly
660 665 670

Val Gln Pro Thr Gln Pro Thr Ala Phe Glu Phe Val Arg Pro Tyr Ser
675 680 685

Asp Tyr Leu Asn Pro Arg Ser Gly Gly Ile Ser Ser Arg Ser Gly Asn
690 695 700

Thr Asp Lys Pro Arg Pro Pro Pro Leu Pro Ser Glu Pro Pro Pro Pro
705 710 715 720

Leu Pro Pro Leu Pro Lys
725

<210> SEQ ID NO 2

<211> LENGTH: 2181

<212> TYPE: DNA

<213> ORGANISM: Artificial Sequence

<220> FEATURE:

<223> OTHER INFORMATION: C261Y mutant of human CCNT1

<400> SEQUENCE: 2

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gcgccaatc tgcttcagga catggggcag cgtcttaacg tctcacaatt gactatcaac 180
actgctatag tatacatgca tcgattctac atgattcagt ccttcacaca gttccctgga 240
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ttggaacatg tcatcaaggt agcacatact tgtctccatc ctcaggaatc cttcctgat 360
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actcaacttg ttcgagcaag caaggactta gcacagactt cttacttcat ggcaaccaac 540
agcctgcatt tgaccacatt tagcctgcag tacacacctc ctgtggtggc ctgtgtctgc 600
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<210> SEQ ID NO 3

<211> LENGTH: 726

<212> TYPE: PRT

<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 3

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20          25          30
Lys Glu Leu Ser Tyr Arg Gln Gln Ala Ala Asn Leu Leu Gln Asp Met
35          40          45
Gly Gln Arg Leu Asn Val Ser Gln Leu Thr Ile Asn Thr Ala Ile Val
50          55          60
Tyr Met His Arg Phe Tyr Met Ile Gln Ser Phe Thr Gln Phe Pro Gly
65          70          75          80
Asn Ser Val Ala Pro Ala Ala Leu Phe Leu Ala Ala Lys Val Glu Glu
85          90          95
Gln Pro Lys Lys Leu Glu His Val Ile Lys Val Ala His Thr Cys Leu
100         105         110
His Pro Gln Glu Ser Leu Pro Asp Thr Arg Ser Glu Ala Tyr Leu Gln
115         120         125
Gln Val Gln Asp Leu Val Ile Leu Glu Ser Ile Ile Leu Gln Thr Leu
130         135         140
Gly Phe Glu Leu Thr Ile Asp His Pro His Thr His Val Val Lys Cys
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Thr Gln Leu Val Arg Ala Ser Lys Asp Leu Ala Gln Thr Ser Tyr Phe
165         170         175

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Met Ala Thr Asn Ser Leu His Leu Thr Thr Phe Ser Leu Gln Tyr Thr
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Pro Pro Val Val Ala Cys Val Cys Ile His Leu Ala Cys Lys Trp Ser
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Asn Trp Glu Ile Pro Val Ser Thr Asp Gly Lys His Trp Trp Glu Tyr
210 215 220

Val Asp Ala Thr Val Thr Leu Glu Leu Leu Asp Glu Leu Thr His Glu
225 230 235 240

Phe Leu Gln Ile Leu Glu Lys Thr Pro Asn Arg Leu Lys Arg Ile Trp
245 250 255

Asn Trp Arg Ala Cys Glu Ala Ala Lys Lys Thr Lys Ala Asp Asp Arg
260 265 270

Gly Thr Asp Glu Lys Thr Ser Glu Gln Thr Ile Leu Asn Met Ile Ser
275 280 285

Gln Ser Ser Ser Asp Thr Thr Ile Ala Gly Leu Met Ser Met Ser Thr
290 295 300

Ser Thr Thr Ser Ala Val Pro Ser Leu Pro Val Ser Glu Glu Ser Ser
305 310 315 320

Ser Asn Leu Thr Ser Val Glu Met Leu Pro Gly Lys Arg Trp Leu Ser
325 330 335

Ser Gln Pro Ser Phe Lys Leu Glu Pro Thr Gln Gly His Arg Thr Ser
340 345 350

Glu Asn Leu Ala Leu Thr Gly Val Asp His Ser Leu Pro Gln Asp Gly
355 360 365

Ser Asn Ala Phe Ile Ser Gln Lys Gln Asn Ser Lys Ser Val Pro Ser
370 375 380

Ala Lys Val Ser Leu Lys Glu Tyr Arg Ala Lys His Ala Glu Glu Leu
385 390 395 400

Ala Ala Gln Lys Arg Gln Leu Glu Asn Met Glu Ala Asn Val Lys Ser
405 410 415

Gln Tyr Ala Tyr Ala Ala Gln Asn Leu Leu Ser His His Asp Ser His
420 425 430

Ser Ser Val Ile Leu Lys Met Pro Ile Glu Gly Ser Glu Asn Pro Glu
435 440 445

Arg Pro Phe Leu Glu Lys Ala Asp Lys Thr Ala Leu Lys Met Arg Ile
450 455 460

Pro Val Ala Gly Gly Asp Lys Ala Ala Ser Ser Lys Pro Glu Glu Ile
465 470 475 480

Lys Met Arg Ile Lys Val His Ala Ala Ala Asp Lys His Asn Ser Val
485 490 495

Glu Asp Ser Val Thr Lys Ser Arg Glu His Lys Glu Lys His Lys Thr
500 505 510

His Pro Ser Asn His His His His His Asn His His Ser His Lys His
515 520 525

Ser His Ser Gln Leu Pro Val Gly Thr Gly Asn Lys Arg Pro Gly Asp
530 535 540

Pro Lys His Ser Ser Gln Thr Ser Asn Leu Ala His Lys Thr Tyr Ser
545 550 555 560

Leu Ser Ser Ser Phe Ser Ser Ser Ser Thr Arg Lys Arg Gly Pro
565 570 575

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Ser Glu Glu Thr Gly Gly Ala Val Phe Asp His Pro Ala Lys Ile Ala
 580 585 590

Lys Ser Thr Lys Ser Ser Ser Leu Asn Phe Ser Phe Pro Ser Leu Pro
 595 600 605

Thr Met Gly Gln Met Pro Gly His Ser Ser Asp Thr Ser Gly Leu Ser
 610 615 620

Phe Ser Gln Pro Ser Cys Lys Thr Arg Val Pro His Ser Lys Leu Asp
 625 630 635 640

Lys Gly Pro Thr Gly Ala Asn Gly His Asn Thr Thr Gln Thr Ile Asp
 645 650 655

Tyr Gln Asp Thr Val Asn Met Leu His Ser Leu Leu Ser Ala Gln Gly
 660 665 670

Val Gln Pro Thr Gln Pro Thr Ala Phe Glu Phe Val Arg Pro Tyr Ser
 675 680 685

Asp Tyr Leu Asn Pro Arg Ser Gly Gly Ile Ser Ser Arg Ser Gly Asn
 690 695 700

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Leu Pro Pro Leu Pro Lys
 725

<210> SEQ ID NO 4
 <211> LENGTH: 2181
 <212> TYPE: DNA
 <213> ORGANISM: Homo sapiens

<400> SEQUENCE: 4

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actgctatag tatacatgca tcgattctac atgattcagt ccttcacaca gttccctgga      240
aattctgtgg ctccagcagc cttgtttcta gcagctaaag tggaggagca gccccaaaaa      300
ttggaacatg tcatcaaggt agcacatact tgtctccatc ctcaggaatc ctttcctgat      360
actagaagtg aggcttattt gcaacaagtt caagatctgg tcattttaga aagcataatt      420
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agcctgcatt tgaccacatt tagcctgcag tacacacctc ctgtggtggc ctgtgtctgc      600
attcacctgg cttgcaagtg gtccaattgg gagatcccag tctcaactga cgggaagcac      660
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tgcgaggctg ccaagaaaac aaaagcagat gaccgaggaa cagatgaaaa gacttcagag      840
cagacaatcc tcaatatgat tcccagagc tcttcagaca caaccattgc aggtttaatg      900
agcatgtcaa cttctaccac aagtgcagtg ctttcctctc cagtctccga agagtcatcc      960
agcaacttaa ccagtgtgga gatgttgcg ggcaagcgtt ggctgtcctc ccaaccttct     1020
ttcaaactag aacctactca gggtcacgag actagtgaga attagcact tacaggagtt     1080
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atagaggggtt cagaaaaacc cgagcggcct tttctgaaa aggctgacaa aacagctctc 1380
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aaaatgcgca taaaagtcca tgctgcagct gataagcaca attctgtaga ggacagtgtt 1500
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aaagggccca ctggggccaa tggtcacaac acgaccaga caatagacta tcaagacact 1980
gtgaatatgc ttcactccct gctcagtgcc caggggtgtc agcccactca gctactgca 2040
tttgaatttg ttcgtcctta tagtgactat ctgaatcctc ggtctggtgg aatctcctcg 2100
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cttccacccc ttctaagta a 2181

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<210> SEQ ID NO 5

<211> LENGTH: 1000

<212> TYPE: DNA

<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 5

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catggtgaaa cctcgtctct actaaaaata caaaaattag ccaggcatgg tgatgcacac 180
ctgtagtcc agctacttgg gaggtgaga caggagaatc gcttgaactc gggacgttag 240
gttgcagtga gccagatg tgccactgca ctccagcctg gatgacaaag tgagactctg 300
tctcaacaa acaaacaaac aaaaaaac agtaacaaca aaaaagaagt aggcttgaga 360
gcacatcttt tacttttagca taaaacctca ccaaaatttc tagaactcag ttatggacta 420
actataatca taagcgaagg catggatgtt catgtatgaa ttttagataa gcatagattc 480
tttgttgta ttattgcttt gtaacgtttg gatagattgc tgtgactctt aattgaaggt 540
tttaaatct tctcttgatg gtaatattta ttggattaca tggtaggata gcctcctgcc 600
tgtggcctat ccagaacttc cagtgttgct gcaagtacaa tctactcacc tcagtgtttt 660
tttatttagt aaattaccta agtaaagaga tgctatttgc ttcattgcag gcatgcgagg 720
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tctcaatat gatttcccag agctcttcag acacaacat tgcaggttta atgagcatgt 840
caacttctac cacaagtgca gtgccttccc tgccagtctc cgaagagtca tccagcaact 900
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<210> SEQ ID NO 6

<211> LENGTH: 724

<212> TYPE: PRT

<213> ORGANISM: Mus musculus

<400> SEQUENCE: 6

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 Lys Glu Leu Ser Tyr Arg Gln Gln Ala Ala Asn Leu Leu Gln Asp Met
 35 40 45
 Gly Gln Arg Leu Asn Val Ser Gln Leu Thr Ile Asn Thr Ala Ile Val
 50 55 60
 Tyr Met His Arg Phe Tyr Met Ile Gln Ser Phe Thr Gln Phe His Arg
 65 70 75 80
 Tyr Ser Met Ala Pro Ala Ala Leu Phe Leu Ala Ala Lys Val Glu Glu
 85 90 95
 Gln Pro Lys Lys Leu Glu His Val Ile Lys Val Ala His Thr Cys Leu
 100 105 110
 His Pro Gln Glu Ser Leu Pro Asp Thr Arg Ser Glu Ala Tyr Leu Gln
 115 120 125
 Gln Val Gln Asp Leu Val Ile Leu Glu Ser Ile Ile Leu Gln Thr Leu
 130 135 140
 Gly Phe Glu Leu Thr Ile Asp His Pro His Thr His Val Val Lys Cys
 145 150 155 160
 Thr Gln Leu Val Arg Ala Ser Lys Asp Leu Ala Gln Thr Ser Tyr Phe
 165 170 175
 Met Ala Thr Asn Ser Leu His Leu Thr Thr Phe Ser Leu Gln Tyr Thr
 180 185 190
 Pro Pro Val Val Ala Cys Val Cys Ile His Leu Ala Cys Lys Trp Ser
 195 200 205
 Asn Trp Glu Ile Pro Val Ser Thr Asp Gly Lys His Trp Trp Glu Tyr
 210 215 220
 Val Asp Ala Thr Val Thr Leu Glu Leu Leu Asp Glu Leu Thr His Glu
 225 230 235 240
 Phe Leu Gln Ile Leu Glu Lys Thr Pro Ser Arg Leu Lys Arg Ile Arg
 245 250 255
 Asn Trp Arg Ala Tyr Gln Ala Ala Met Lys Thr Lys Pro Asp Asp Arg
 260 265 270
 Gly Ala Asp Glu Asn Thr Ser Glu Gln Thr Ile Leu Asn Met Ile Ser
 275 280 285
 Gln Thr Ser Ser Asp Thr Thr Ile Ala Gly Leu Met Ser Met Ser Thr
 290 295 300
 Ala Ser Thr Ser Ala Val Pro Ser Leu Pro Ser Ser Glu Glu Ser Ser
 305 310 315 320
 Ser Ser Leu Thr Ser Val Asp Met Leu Gln Gly Glu Arg Trp Leu Ser
 325 330 335
 Ser Gln Pro Pro Phe Lys Leu Glu Ala Ala Gln Gly His Arg Thr Ser
 340 345 350

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Glu Ser Leu Ala Leu Ile Gly Val Asp His Ser Leu Gln Gln Asp Gly
 355 360 365

 Ser Ser Ala Phe Gly Ser Gln Lys Gln Ala Ser Lys Ser Val Pro Ser
 370 375 380

 Ala Lys Val Ser Leu Lys Glu Tyr Arg Ala Lys His Ala Glu Glu Leu
 385 390 395 400

 Ala Ala Gln Lys Arg Gln Leu Glu Asn Met Glu Ala Asn Val Lys Ser
 405 410 415

 Gln Tyr Ala Tyr Ala Ala Gln Asn Leu Leu Ser His Asp Ser His Ser
 420 425 430

 Ser Val Ile Leu Lys Met Pro Ile Glu Ser Ser Glu Asn Pro Glu Arg
 435 440 445

 Pro Phe Leu Asp Lys Ala Asp Lys Ser Ala Leu Lys Met Arg Leu Pro
 450 455 460

 Val Ala Ser Gly Asp Lys Ala Val Ser Ser Lys Pro Glu Glu Ile Lys
 465 470 475 480

 Met Arg Ile Lys Val His Ser Ala Gly Asp Lys His Asn Ser Ile Glu
 485 490 495

 Asp Ser Val Thr Lys Ser Arg Glu His Lys Glu Lys Gln Arg Thr His
 500 505 510

 Pro Ser Asn His His His His His Asn His His Ser His Arg His Ser
 515 520 525

 His Leu Gln Leu Pro Ala Gly Pro Val Ser Lys Arg Pro Ser Asp Pro
 530 535 540

 Lys His Ser Ser Gln Thr Ser Thr Leu Ala His Lys Thr Tyr Ser Leu
 545 550 555 560

 Ser Ser Thr Leu Ser Ser Ser Ser Ser Thr Arg Lys Arg Gly Pro Pro
 565 570 575

 Glu Glu Thr Gly Ala Ala Val Phe Asp His Pro Ala Lys Ile Ala Lys
 580 585 590

 Ser Thr Lys Ser Ser Leu Asn Phe Pro Phe Pro Pro Leu Pro Thr Met
 595 600 605

 Thr Gln Leu Pro Gly His Ser Ser Asp Thr Ser Gly Leu Pro Phe Ser
 610 615 620

 Gln Pro Ser Cys Lys Thr Arg Val Pro His Met Lys Leu Asp Lys Gly
 625 630 635 640

 Pro Pro Gly Ala Asn Gly His Asn Ala Thr Gln Ser Ile Asp Tyr Gln
 645 650 655

 Asp Thr Val Asn Met Leu His Ser Leu Leu Ser Ala Gln Gly Val Gln
 660 665 670

 Pro Thr Gln Ala Pro Ala Phe Glu Phe Val His Ser Tyr Gly Glu Tyr
 675 680 685

 Met Asn Pro Arg Ala Gly Ala Ile Ser Ser Arg Ser Gly Thr Thr Asp
 690 695 700

 Lys Pro Arg Pro Pro Pro Leu Pro Ser Glu Pro Pro Pro Pro Leu Pro
 705 710 715 720

 Pro Leu Pro Lys

<210> SEQ ID NO 7

<211> LENGTH: 1071

<212> TYPE: PRT

<213> ORGANISM: Artificial Sequence

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<220> FEATURE:

<223> OTHER INFORMATION: P411T, M412V, F414S human XPO1 mutant

<400> SEQUENCE: 7

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Met Pro Ala Ile Met Thr Met Leu Ala Asp His Ala Ala Arg Gln Leu
1           5           10           15

Leu Asp Phe Ser Gln Lys Leu Asp Ile Asn Leu Leu Asp Asn Val Val
20           25           30

Asn Cys Leu Tyr His Gly Glu Gly Ala Gln Gln Arg Met Ala Gln Glu
35           40           45

Val Leu Thr His Leu Lys Glu His Pro Asp Ala Trp Thr Arg Val Asp
50           55           60

Thr Ile Leu Glu Phe Ser Gln Asn Met Asn Thr Lys Tyr Tyr Gly Leu
65           70           75           80

Gln Ile Leu Glu Asn Val Ile Lys Thr Arg Trp Lys Ile Leu Pro Arg
85           90           95

Asn Gln Cys Glu Gly Ile Lys Lys Tyr Val Val Gly Leu Ile Ile Lys
100          105          110

Thr Ser Ser Asp Pro Thr Cys Val Glu Lys Glu Lys Val Tyr Ile Gly
115          120          125

Lys Leu Asn Met Ile Leu Val Gln Ile Leu Lys Gln Glu Trp Pro Lys
130          135          140

His Trp Pro Thr Phe Ile Ser Asp Ile Val Gly Ala Ser Arg Thr Ser
145          150          155          160

Glu Ser Leu Cys Gln Asn Asn Met Val Ile Leu Lys Leu Leu Ser Glu
165          170          175

Glu Val Phe Asp Phe Ser Ser Gly Gln Ile Thr Gln Val Lys Ser Lys
180          185          190

His Leu Lys Asp Ser Met Cys Asn Glu Phe Ser Gln Ile Phe Gln Leu
195          200          205

Cys Gln Phe Val Met Glu Asn Ser Gln Asn Ala Pro Leu Val His Ala
210          215          220

Thr Leu Glu Thr Leu Leu Arg Phe Leu Asn Trp Ile Pro Leu Gly Tyr
225          230          235          240

Ile Phe Glu Thr Lys Leu Ile Ser Thr Leu Ile Tyr Lys Phe Leu Asn
245          250          255

Val Pro Met Phe Arg Asn Val Ser Leu Lys Cys Leu Thr Glu Ile Ala
260          265          270

Gly Val Ser Val Ser Gln Tyr Glu Glu Gln Phe Val Thr Leu Phe Thr
275          280          285

Leu Thr Met Met Gln Leu Lys Gln Met Leu Pro Leu Asn Thr Asn Ile
290          295          300

Arg Leu Ala Tyr Ser Asn Gly Lys Asp Asp Glu Gln Asn Phe Ile Gln
305          310          315          320

Asn Leu Ser Leu Phe Leu Cys Thr Phe Leu Lys Glu His Asp Gln Leu
325          330          335

Ile Glu Lys Arg Leu Asn Leu Arg Glu Thr Leu Met Glu Ala Leu His
340          345          350

Tyr Met Leu Leu Val Ser Glu Val Glu Glu Thr Glu Ile Phe Lys Ile
355          360          365

Cys Leu Glu Tyr Trp Asn His Leu Ala Ala Glu Leu Tyr Arg Glu Ser
370          375          380

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Pro Phe Ser Thr Ser Ala Ser Pro Leu Leu Ser Gly Ser Gln His Phe
 385 390 395 400
 Asp Val Pro Pro Arg Arg Gln Leu Tyr Leu Thr Val Leu Ser Lys Val
 405 410 415
 Arg Leu Leu Met Val Ser Arg Met Ala Lys Pro Glu Glu Val Leu Val
 420 425 430
 Val Glu Asn Asp Gln Gly Glu Val Val Arg Glu Phe Met Lys Asp Thr
 435 440 445
 Asp Ser Ile Asn Leu Tyr Lys Asn Met Arg Glu Thr Leu Val Tyr Leu
 450 455 460
 Thr His Leu Asp Tyr Val Asp Thr Glu Arg Ile Met Thr Glu Lys Leu
 465 470 475 480
 His Asn Gln Val Asn Gly Thr Glu Trp Ser Trp Lys Asn Leu Asn Thr
 485 490 495
 Leu Cys Trp Ala Ile Gly Ser Ile Ser Gly Ala Met His Glu Glu Asp
 500 505 510
 Glu Lys Arg Phe Leu Val Thr Val Ile Lys Asp Leu Leu Gly Leu Cys
 515 520 525
 Glu Gln Lys Arg Gly Lys Asp Asn Lys Ala Ile Ile Ala Ser Asn Ile
 530 535 540
 Met Tyr Ile Val Gly Gln Tyr Pro Arg Phe Leu Arg Ala His Trp Lys
 545 550 555 560
 Phe Leu Lys Thr Val Val Asn Lys Leu Phe Glu Phe Met His Glu Thr
 565 570 575
 His Asp Gly Val Gln Asp Met Ala Cys Asp Thr Phe Ile Lys Ile Ala
 580 585 590
 Gln Lys Cys Arg Arg His Phe Val Gln Val Gln Val Gly Glu Val Met
 595 600 605
 Pro Phe Ile Asp Glu Ile Leu Asn Asn Ile Asn Thr Ile Ile Cys Asp
 610 615 620
 Leu Gln Pro Gln Gln Val His Thr Phe Tyr Glu Ala Val Gly Tyr Met
 625 630 635 640
 Ile Gly Ala Gln Thr Asp Gln Thr Val Gln Glu His Leu Ile Glu Lys
 645 650 655
 Tyr Met Leu Leu Pro Asn Gln Val Trp Asp Ser Ile Ile Gln Gln Ala
 660 665 670
 Thr Lys Asn Val Asp Ile Leu Lys Asp Pro Glu Thr Val Lys Gln Leu
 675 680 685
 Gly Ser Ile Leu Lys Thr Asn Val Arg Ala Cys Lys Ala Val Gly His
 690 695 700
 Pro Phe Val Ile Gln Leu Gly Arg Ile Tyr Leu Asp Met Leu Asn Val
 705 710 715 720
 Tyr Lys Cys Leu Ser Glu Asn Ile Ser Ala Ala Ile Gln Ala Asn Gly
 725 730 735
 Glu Met Val Thr Lys Gln Pro Leu Ile Arg Ser Met Arg Thr Val Lys
 740 745 750
 Arg Glu Thr Leu Lys Leu Ile Ser Gly Trp Val Ser Arg Ser Asn Asp
 755 760 765
 Pro Gln Met Val Ala Glu Asn Phe Val Pro Pro Leu Leu Asp Ala Val
 770 775 780

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Leu Ile Asp Tyr Gln Arg Asn Val Pro Ala Ala Arg Glu Pro Glu Val
 785 790 795 800

Leu Ser Thr Met Ala Ile Ile Val Asn Lys Leu Gly Gly His Ile Thr
 805 810 815

Ala Glu Ile Pro Gln Ile Phe Asp Ala Val Phe Glu Cys Thr Leu Asn
 820 825 830

Met Ile Asn Lys Asp Phe Glu Glu Tyr Pro Glu His Arg Thr Asn Phe
 835 840 845

Phe Leu Leu Leu Gln Ala Val Asn Ser His Cys Phe Pro Ala Phe Leu
 850 855 860

Ala Ile Pro Pro Thr Gln Phe Lys Leu Val Leu Asp Ser Ile Ile Trp
 865 870 875 880

Ala Phe Lys His Thr Met Arg Asn Val Ala Asp Thr Gly Leu Gln Ile
 885 890 895

Leu Phe Thr Leu Leu Gln Asn Val Ala Gln Glu Glu Ala Ala Ala Gln
 900 905 910

Ser Phe Tyr Gln Thr Tyr Phe Cys Asp Ile Leu Gln His Ile Phe Ser
 915 920 925

Val Val Thr Asp Thr Ser His Thr Ala Gly Leu Thr Met His Ala Ser
 930 935 940

Ile Leu Ala Tyr Met Phe Asn Leu Val Glu Glu Gly Lys Ile Ser Thr
 945 950 955 960

Ser Leu Asn Pro Gly Asn Pro Val Asn Asn Gln Ile Phe Leu Gln Glu
 965 970 975

Tyr Val Ala Asn Leu Leu Lys Ser Ala Phe Pro His Leu Gln Asp Ala
 980 985 990

Gln Val Lys Leu Phe Val Thr Gly Leu Phe Ser Leu Asn Gln Asp Ile
 995 1000 1005

Pro Ala Phe Lys Glu His Leu Arg Asp Phe Leu Val Gln Ile Lys
 1010 1015 1020

Glu Phe Ala Gly Glu Asp Thr Ser Asp Leu Phe Leu Glu Glu Arg
 1025 1030 1035

Glu Ile Ala Leu Arg Gln Ala Asp Glu Glu Lys His Lys Arg Gln
 1040 1045 1050

Met Ser Val Pro Gly Ile Phe Asn Pro His Glu Ile Pro Glu Glu
 1055 1060 1065

Met Cys Asp
 1070

<210> SEQ ID NO 8
 <211> LENGTH: 3216
 <212> TYPE: DNA
 <213> ORGANISM: Artificial Sequence
 <220> FEATURE:
 <223> OTHER INFORMATION: P411T, M412V, F414S human XPO1 mutant

<400> SEQUENCE: 8

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caaaaactgg atatcaactt attagataat gtggtgaatt gcttatacca tggagaagga 120

gccagcaaaa gaatggctca agaagtactg acacatttaa aggagcatcc tgatgcttgg 180

acaagagtcg acacaatttt ggaattttct cagaatatga atacgaaata ctatggacta 240

caaattttgg aaaatgtgat aaaaacaagg tggaagattc ttccaaggaa ccagtgcgaa 300

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ggaataaaaa aatacgttgt tggcctcatt atcaagacgt catctgacce aacttggtga	360
gagaaagaaa aggtgtatat cggaaaatta aatatgatcc ttgttcagat actgaaacaa	420
gaatggccca aacattggcc aacttttacc agtgatattg ttggagcaag taggaccagc	480
gaaagtctct gtcaaaataa tatggtgatt cttaaactct tgagtgaaga agtatttgat	540
ttctctagtg gacagataac ccaagtcaaa tctaagcatt taaaagacag catgtgcaat	600
gaattctcac agatatttca actgtgtcag tttgtaatgg aaaattctca aaatgctcca	660
ctgttacatg caaccttggg aacattgctc agatttctga actggattcc cctgggatat	720
atthttgaga ccaaattaat cagcacattg atttataagt tcctgaatgt tccaatgtht	780
cgaaatgtct ctctgaagtg cctcactgag attgctggtg tgagtgtaa ccaatatgaa	840
gaacaatttg taacactatt tactctgaca atgatgcaac taaagcagat gcttccttta	900
aataccaata ttcgacttgc gtactcaaat ggaaaagatg atgaacagaa cttcattcaa	960
aatctcagtt tgtttctctg cacctttctt aaggaacatg atcaacttat agaaaaaaga	1020
ttaaatctca gggaaactct tatggaggcc cttcattata tgttggttgt atctgaagta	1080
gaagaaactg aaatctttaa aatttgtctt gaatactgga atcatttggc tgctgaactc	1140
tatagagaga gtccattctc tacatctcgg tctccgttgc tttctggaag tcaacatttt	1200
gatgttctcc ccaggagaca gctgtatttg accgtgttat caaaggctcc tttattaatg	1260
gttagtcgaa tggctaaacc agaggaagta ttggtttagt agaatgatca aggagaagtt	1320
gtgagagaat tcactgaagga tacagattcc ataaatttgt ataagaatat gagggaaaca	1380
ttggtttacc ttactcatct ggattatgta gatacagaaa gaataatgac agagaagctt	1440
cacaatcaag tgaatggtac agagtgttca tggaaaaatt tgaatacatt gtgttgggca	1500
ataggctcca ttagtggagc aatgcatgaa gaggacgaaa aacgatttct tgttactggt	1560
ataaaggatc tattaggatt atgtgaacag aaaagaggca aagataataa agctattatt	1620
gcatcaaata tcactgacat agtaggtcaa taccacgctt ttttgagagc tcaactggaaa	1680
tttctgaaga ctgtagttaa caagctgttc gaattcatgc atgagacca tgatggagtc	1740
caggatatgg cttgtgatac tttcattaaa atagcccaaa aatgccgcag gcatttctgt	1800
caggttcagg ttggagaagt gatgccattt attgatgaaa ttttgaacaa cattaacact	1860
attatttgtg atcttcagcc tcaacagggt catacgtttt atgaagctgt ggggtacatg	1920
attggtgcac aaacagatca aacagtacaa gaacacttga tagaaaagta catgttactc	1980
cctaatacaag tgtgggatag tataatccag caggcaacca aaaatgtgga tatactgaaa	2040
gatctgaaa cagtcaagca gcttggtagc attttgaaaa caaatgtgag agcctgcaaa	2100
gctgttggac accccttctg aattcagctt ggaagaattt atttagatat gcttaatgta	2160
tacaagtgcc tcagtgaaaa tatttctgca gctatccaag ctaatggtga aatggttaca	2220
aagcaacctc tgattagaag tatgccaact gtaaaaaggg aaactttaaa gttaatatct	2280
ggttgggtga gccgatccaa tgatccacag atggtcgcctg aaaatttctg tccccctctg	2340
ttggtgagc ttctcattga ttatcagaga aatgtcccag ctgctagaga accagaagtg	2400
cttagtacta tggccataat tgtcaacaag ttagggggac atataacagc tgaataacct	2460
caaatatttg atgctgtttt tgaatgcaca ttgaatatga taaataagga ctttgaagaa	2520
tactctgaac atagaacgaa ctttttctta ctacttcagg ctgtcaattc tcattgttcc	2580

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ccagcattcc ttgctattcc acctacacag tttaaacttg ttttgattc catcatttg 2640
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ttacaaaatg ttgcacaaga agaagctgca gctcagagt tttatcaaac ttattttgt 2760
gatattctcc agcatatctt tctgtgtg acagacact cacatactgc tggtttaaca 2820
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tcattaaatc ctggaaatcc agttaacaac caaatctttc ttcaggaata tgtggcta 2940
ctccttaagt cggecttccc tcacctacaa gatgctcaag taaagctctt tgtgacagg 3000
cttttcagct taaatcaaga tattctgct ttcaaggaac atttaagaga ttcctagtt 3060
caaataaagg aatttgcagg tgaagacact tctgattgt ttttgaaga gagagaaata 3120
gccctacggc aggctgatga agagaaacat aaacgtcaaa tgtctgtccc tggcatctt 3180
aatccacatg agattccaga agaagtgtg gattaa 3216

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<210> SEQ ID NO 9

<211> LENGTH: 1071

<212> TYPE: PRT

<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 9

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Met Pro Ala Ile Met Thr Met Leu Ala Asp His Ala Ala Arg Gln Leu
1           5           10          15
Leu Asp Phe Ser Gln Lys Leu Asp Ile Asn Leu Leu Asp Asn Val Val
                20           25           30
Asn Cys Leu Tyr His Gly Glu Gly Ala Gln Gln Arg Met Ala Gln Glu
                35           40           45
Val Leu Thr His Leu Lys Glu His Pro Asp Ala Trp Thr Arg Val Asp
                50           55           60
Thr Ile Leu Glu Phe Ser Gln Asn Met Asn Thr Lys Tyr Tyr Gly Leu
                65           70           75           80
Gln Ile Leu Glu Asn Val Ile Lys Thr Arg Trp Lys Ile Leu Pro Arg
                85           90           95
Asn Gln Cys Glu Gly Ile Lys Lys Tyr Val Val Gly Leu Ile Ile Lys
                100          105          110
Thr Ser Ser Asp Pro Thr Cys Val Glu Lys Glu Lys Val Tyr Ile Gly
                115          120          125
Lys Leu Asn Met Ile Leu Val Gln Ile Leu Lys Gln Glu Trp Pro Lys
                130          135          140
His Trp Pro Thr Phe Ile Ser Asp Ile Val Gly Ala Ser Arg Thr Ser
                145          150          155          160
Glu Ser Leu Cys Gln Asn Asn Met Val Ile Leu Lys Leu Leu Ser Glu
                165          170          175
Glu Val Phe Asp Phe Ser Ser Gly Gln Ile Thr Gln Val Lys Ser Lys
                180          185          190
His Leu Lys Asp Ser Met Cys Asn Glu Phe Ser Gln Ile Phe Gln Leu
                195          200          205
Cys Gln Phe Val Met Glu Asn Ser Gln Asn Ala Pro Leu Val His Ala
                210          215          220
Thr Leu Glu Thr Leu Leu Arg Phe Leu Asn Trp Ile Pro Leu Gly Tyr
                225          230          235          240

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Ile	Phe	Glu	Thr	Lys	Leu	Ile	Ser	Thr	Leu	Ile	Tyr	Lys	Phe	Leu	Asn
				245					250					255	
Val	Pro	Met	Phe	Arg	Asn	Val	Ser	Leu	Lys	Cys	Leu	Thr	Glu	Ile	Ala
			260					265					270		
Gly	Val	Ser	Val	Ser	Gln	Tyr	Glu	Glu	Gln	Phe	Val	Thr	Leu	Phe	Thr
		275					280					285			
Leu	Thr	Met	Met	Gln	Leu	Lys	Gln	Met	Leu	Pro	Leu	Asn	Thr	Asn	Ile
	290					295						300			
Arg	Leu	Ala	Tyr	Ser	Asn	Gly	Lys	Asp	Asp	Glu	Gln	Asn	Phe	Ile	Gln
305					310					315					320
Asn	Leu	Ser	Leu	Phe	Leu	Cys	Thr	Phe	Leu	Lys	Glu	His	Asp	Gln	Leu
				325					330					335	
Ile	Glu	Lys	Arg	Leu	Asn	Leu	Arg	Glu	Thr	Leu	Met	Glu	Ala	Leu	His
			340					345					350		
Tyr	Met	Leu	Leu	Val	Ser	Glu	Val	Glu	Glu	Thr	Glu	Ile	Phe	Lys	Ile
		355					360					365			
Cys	Leu	Glu	Tyr	Trp	Asn	His	Leu	Ala	Ala	Glu	Leu	Tyr	Arg	Glu	Ser
	370					375						380			
Pro	Phe	Ser	Thr	Ser	Ala	Ser	Pro	Leu	Leu	Ser	Gly	Ser	Gln	His	Phe
385					390					395					400
Asp	Val	Pro	Pro	Arg	Arg	Gln	Leu	Tyr	Leu	Pro	Met	Leu	Phe	Lys	Val
				405					410					415	
Arg	Leu	Leu	Met	Val	Ser	Arg	Met	Ala	Lys	Pro	Glu	Glu	Val	Leu	Val
			420					425					430		
Val	Glu	Asn	Asp	Gln	Gly	Glu	Val	Val	Arg	Glu	Phe	Met	Lys	Asp	Thr
		435					440					445			
Asp	Ser	Ile	Asn	Leu	Tyr	Lys	Asn	Met	Arg	Glu	Thr	Leu	Val	Tyr	Leu
	450					455					460				
Thr	His	Leu	Asp	Tyr	Val	Asp	Thr	Glu	Arg	Ile	Met	Thr	Glu	Lys	Leu
465					470					475					480
His	Asn	Gln	Val	Asn	Gly	Thr	Glu	Trp	Ser	Trp	Lys	Asn	Leu	Asn	Thr
				485					490					495	
Leu	Cys	Trp	Ala	Ile	Gly	Ser	Ile	Ser	Gly	Ala	Met	His	Glu	Glu	Asp
			500					505					510		
Glu	Lys	Arg	Phe	Leu	Val	Thr	Val	Ile	Lys	Asp	Leu	Leu	Gly	Leu	Cys
		515					520					525			
Glu	Gln	Lys	Arg	Gly	Lys	Asp	Asn	Lys	Ala	Ile	Ile	Ala	Ser	Asn	Ile
	530					535					540				
Met	Tyr	Ile	Val	Gly	Gln	Tyr	Pro	Arg	Phe	Leu	Arg	Ala	His	Trp	Lys
545					550					555					560
Phe	Leu	Lys	Thr	Val	Val	Asn	Lys	Leu	Phe	Glu	Phe	Met	His	Glu	Thr
				565					570					575	
His	Asp	Gly	Val	Gln	Asp	Met	Ala	Cys	Asp	Thr	Phe	Ile	Lys	Ile	Ala
			580					585					590		
Gln	Lys	Cys	Arg	Arg	His	Phe	Val	Gln	Val	Gln	Val	Gly	Glu	Val	Met
		595					600					605			
Pro	Phe	Ile	Asp	Glu	Ile	Leu	Asn	Asn	Ile	Asn	Thr	Ile	Ile	Cys	Asp
	610					615					620				
Leu	Gln	Pro	Gln	Gln	Val	His	Thr	Phe	Tyr	Glu	Ala	Val	Gly	Tyr	Met
625					630					635					640
Ile	Gly	Ala	Gln	Thr	Asp	Gln	Thr	Val	Gln	Glu	His	Leu	Ile	Glu	Lys

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645					650					655					
Tyr	Met	Leu	Leu	Pro	Asn	Gln	Val	Trp	Asp	Ser	Ile	Ile	Gln	Gln	Ala
		660						665					670		
Thr	Lys	Asn	Val	Asp	Ile	Leu	Lys	Asp	Pro	Glu	Thr	Val	Lys	Gln	Leu
		675					680					685			
Gly	Ser	Ile	Leu	Lys	Thr	Asn	Val	Arg	Ala	Cys	Lys	Ala	Val	Gly	His
	690					695					700				
Pro	Phe	Val	Ile	Gln	Leu	Gly	Arg	Ile	Tyr	Leu	Asp	Met	Leu	Asn	Val
	705				710					715					720
Tyr	Lys	Cys	Leu	Ser	Glu	Asn	Ile	Ser	Ala	Ala	Ile	Gln	Ala	Asn	Gly
			725						730						735
Glu	Met	Val	Thr	Lys	Gln	Pro	Leu	Ile	Arg	Ser	Met	Arg	Thr	Val	Lys
			740						745					750	
Arg	Glu	Thr	Leu	Lys	Leu	Ile	Ser	Gly	Trp	Val	Ser	Arg	Ser	Asn	Asp
			755					760					765		
Pro	Gln	Met	Val	Ala	Glu	Asn	Phe	Val	Pro	Pro	Leu	Leu	Asp	Ala	Val
			770			775					780				
Leu	Ile	Asp	Tyr	Gln	Arg	Asn	Val	Pro	Ala	Ala	Arg	Glu	Pro	Glu	Val
			785			790					795				800
Leu	Ser	Thr	Met	Ala	Ile	Ile	Val	Asn	Lys	Leu	Gly	Gly	His	Ile	Thr
				805						810					815
Ala	Glu	Ile	Pro	Gln	Ile	Phe	Asp	Ala	Val	Phe	Glu	Cys	Thr	Leu	Asn
			820					825						830	
Met	Ile	Asn	Lys	Asp	Phe	Glu	Glu	Tyr	Pro	Glu	His	Arg	Thr	Asn	Phe
			835					840					845		
Phe	Leu	Leu	Leu	Gln	Ala	Val	Asn	Ser	His	Cys	Phe	Pro	Ala	Phe	Leu
			850				855					860			
Ala	Ile	Pro	Pro	Thr	Gln	Phe	Lys	Leu	Val	Leu	Asp	Ser	Ile	Ile	Trp
			865			870					875				880
Ala	Phe	Lys	His	Thr	Met	Arg	Asn	Val	Ala	Asp	Thr	Gly	Leu	Gln	Ile
				885						890					895
Leu	Phe	Thr	Leu	Leu	Gln	Asn	Val	Ala	Gln	Glu	Glu	Ala	Ala	Ala	Gln
			900					905						910	
Ser	Phe	Tyr	Gln	Thr	Tyr	Phe	Cys	Asp	Ile	Leu	Gln	His	Ile	Phe	Ser
			915					920					925		
Val	Val	Thr	Asp	Thr	Ser	His	Thr	Ala	Gly	Leu	Thr	Met	His	Ala	Ser
			930				935					940			
Ile	Leu	Ala	Tyr	Met	Phe	Asn	Leu	Val	Glu	Glu	Gly	Lys	Ile	Ser	Thr
			945			950					955				960
Ser	Leu	Asn	Pro	Gly	Asn	Pro	Val	Asn	Asn	Gln	Ile	Phe	Leu	Gln	Glu
				965						970					975
Tyr	Val	Ala	Asn	Leu	Leu	Lys	Ser	Ala	Phe	Pro	His	Leu	Gln	Asp	Ala
			980						985						990
Gln	Val	Lys	Leu	Phe	Val	Thr	Gly	Leu	Phe	Ser	Leu	Asn	Gln	Asp	Ile
			995					1000						1005	
Pro	Ala	Phe	Lys	Glu	His	Leu	Arg	Asp	Phe	Leu	Val	Gln	Ile	Lys	
			1010					1015					1020		
Glu	Phe	Ala	Gly	Glu	Asp	Thr	Ser	Asp	Leu	Phe	Leu	Glu	Glu	Arg	
			1025					1030					1035		
Glu	Ile	Ala	Leu	Arg	Gln	Ala	Asp	Glu	Glu	Lys	His	Lys	Arg	Gln	
			1040					1045						1050	

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Met Ser Val Pro Gly Ile Phe Asn Pro His Glu Ile Pro Glu Glu
1055 1060 1065

Met Cys Asp
1070

<210> SEQ ID NO 10
<211> LENGTH: 3216
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens

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gcccagcaaa gaatggctca agaagtactg acacatttaa aggagcatcc tgatgcttgg 180
acaagagtgc acacaatttt ggaattttct cagaatatga atacgaaata ctatggacta 240
caaattttgg aaaatgtgat aaaaacaagg tggagattc ttccaaggaa ccagtgcgaa 300
ggaataaaaa aatacgttgt tggcctcatt atcaagacgt catctgaccc aacttgtgta 360
gagaaagaaa aggtgtatat cggaaaatta aatgatgacc ttgttcagat actgaaacaa 420
gaatggccca aacattggcc aacttttacc agtgatattg ttggagcaag taggaccagc 480
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gaattctcac agatatttca actgtgtcag tttgtaattgg aaaattctca aaatgctcca 660
ctgttacatg caaccttggg aacattgctc agattttctga actggattcc cctgggatat 720
atthttgaga ccaaattaat cagcacattg atttataagt tcctgaatgt tccaatgttt 780
cgaaatgtct ctctgaagtg cctcaactgag attgctggtg tgagtgtaa ccaatatgaa 840
gaacaatttg taacactatt tactctgaca atgatgcaac taaagcagat gcttccttta 900
aataccaata ttcgacttgc gtactcaaat ggaaaagatg atgaacagaa cttcattcaa 960
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gaagaaactg aaatctttaa aatttgcctt gaatactgga atcatttggc tgctgaactc 1140
tatagagaga gtccattctc tacatctgcc tctccgttgc tttctggaag tcaacatttt 1200
gatgttctcc ccaggagaca gctatatattg cccatgttat tcaaggtccg tttattaatg 1260
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caggatattg cttgtgatac tttcattaaa atagcccaaa aatgccgcag gcatttcggt 1800
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attattttgtg atcttcagcc tcaacagggt catacgtttt atgaagctgt ggggtacatg 1920
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gatcctgaaa cagtcaagca gcttggtagc attttgaaa caaatgtgag agcctgcaaa 2100
gctgttgac acccctttgt aattcagctt ggaagaattt atttagatat gcttaatgta 2160
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caaataaagg aatttgcagg tgaagacact tctgattgt ttttgaaga gagagaaata 3120
gccctacggc aggctgatga agagaacat aaacgtcaaa tgtctgtccc tggcatctt 3180
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<210> SEQ ID NO 11

<211> LENGTH: 977

<212> TYPE: DNA

<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 11

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tgagagattt ttgaagtgtc ttagggcacg tcagaagaag cagagttact ccagagtttg 180
ctgtctatct gataagtatt gaaatctgag ttgtgatgaa taaaacatga atttttatct 240
tcccctaagg tgtaacaagt gaaaagcaat ttgaagttgg taatgtttaa gaattatctt 300
aacagttttg gtcttctgtg taggccttc attatatggt gttggatctt gaagtagaag 360
aaactgaaat ctttaaaatt tgtcttgaat actggaatca tttggctgct gaactctata 420
gagagagtcc attctctaca tctgcctctc cgttgcttcc tgggaagcaa cttttgatg 480
ttcctcccag gagacagcta tatttgocca tgttattcaa ggtaacagag cggttggttg 540
agtgttcttc ctgttgcata ctgtggttt gaggtctgaa tccaaact tctaactctgt 600
gtaaataaat tagctataaa aagagaaccc aacaacttct ccatgagtgt ggaaaactag 660

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aacatgaaag gagttgagtc tagaaccttg atttcaaga gtgtggtoct tctctcagta 720
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agagtctagg aatgagacac aggaaactcc tgtaacagaa gttgggtaaa aaaatcacat 840
taaaacacac ttaataaatt ataaagccat tttttagata ttacagttaa aaaaaatfff 900
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gctcactaca atcttga 977

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<210> SEQ ID NO 12
<211> LENGTH: 1071
<212> TYPE: PRT
<213> ORGANISM: Mus musculus

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<400> SEQUENCE: 12

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Leu Asp Phe Ser Gln Lys Leu Asp Ile Asn Leu Leu Asp Asn Val Val
20          25          30
Asn Cys Leu Tyr His Gly Glu Gly Ala Gln Gln Arg Met Ala Gln Glu
35          40          45
Val Leu Thr His Leu Lys Glu His Pro Asp Ala Trp Thr Arg Val Asp
50          55          60
Thr Ile Leu Glu Phe Ser Gln Asn Met Asn Thr Lys Tyr Tyr Gly Leu
65          70          75          80
Gln Ile Leu Glu Asn Val Ile Lys Thr Arg Trp Lys Ile Leu Pro Arg
85          90          95
Asn Gln Cys Glu Gly Ile Lys Lys Tyr Val Val Gly Leu Ile Ile Lys
100         105         110
Thr Ser Ser Asp Pro Thr Cys Val Glu Lys Glu Lys Val Tyr Ile Gly
115         120         125
Lys Leu Asn Met Ile Leu Val Gln Ile Leu Lys Gln Glu Trp Pro Lys
130         135         140
His Trp Pro Thr Phe Ile Ser Asp Ile Val Gly Ala Ser Arg Thr Ser
145         150         155         160
Glu Ser Leu Cys Gln Asn Asn Met Val Ile Leu Lys Leu Leu Ser Glu
165         170         175
Glu Val Phe Asp Phe Ser Ser Gly Gln Ile Thr Gln Val Lys Ala Lys
180         185         190
His Leu Lys Asp Ser Met Cys Asn Glu Phe Ser Gln Ile Phe Gln Leu
195         200         205
Cys Gln Phe Val Met Glu Asn Ser Gln Asn Ala Pro Leu Val His Ala
210         215         220
Thr Leu Glu Thr Leu Leu Arg Phe Leu Asn Trp Ile Pro Leu Gly Tyr
225         230         235         240
Ile Phe Glu Thr Lys Leu Ile Ser Thr Leu Ile Tyr Lys Phe Leu Asn
245         250         255
Val Pro Met Phe Arg Asn Val Ser Leu Lys Cys Leu Thr Glu Ile Ala
260         265         270
Gly Val Ser Val Ser Gln Tyr Glu Glu Gln Phe Glu Thr Leu Phe Thr
275         280         285
Leu Thr Met Met Gln Leu Lys Gln Met Leu Pro Leu Asn Thr Asn Ile

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Arg	Leu	Ala	Tyr	Ser	Asn	Gly	Lys	Asp	Asp	Glu	Gln	Asn	Phe	Ile	Gln
305					310					315					320
Asn	Leu	Ser	Leu	Phe	Leu	Cys	Thr	Phe	Leu	Lys	Glu	His	Gly	Gln	Leu
				325					330					335	
Leu	Glu	Lys	Arg	Leu	Asn	Leu	Arg	Glu	Ala	Leu	Met	Glu	Ala	Leu	His
			340					345					350		
Tyr	Met	Leu	Leu	Val	Ser	Glu	Val	Glu	Glu	Thr	Glu	Ile	Phe	Lys	Ile
		355					360					365			
Cys	Leu	Glu	Tyr	Trp	Asn	His	Leu	Ala	Ala	Glu	Leu	Tyr	Arg	Glu	Ser
370					375							380			
Pro	Phe	Ser	Thr	Ser	Ala	Ser	Pro	Leu	Leu	Ser	Gly	Ser	Gln	His	Phe
385					390					395					400
Asp	Ile	Pro	Pro	Arg	Arg	Gln	Leu	Tyr	Leu	Thr	Val	Leu	Ser	Lys	Val
				405					410					415	
Arg	Leu	Leu	Met	Val	Ser	Arg	Met	Ala	Lys	Pro	Glu	Glu	Val	Leu	Val
			420					425					430		
Val	Glu	Asn	Asp	Gln	Gly	Glu	Val	Val	Arg	Glu	Phe	Met	Lys	Asp	Thr
		435					440					445			
Asp	Ser	Ile	Asn	Leu	Tyr	Lys	Asn	Met	Arg	Glu	Thr	Leu	Val	Tyr	Leu
450						455					460				
Thr	His	Leu	Asp	Tyr	Val	Asp	Thr	Glu	Ile	Ile	Met	Thr	Lys	Lys	Leu
465					470					475					480
Gln	Asn	Gln	Val	Asn	Gly	Thr	Glu	Trp	Ser	Trp	Lys	Asn	Leu	Asn	Thr
				485					490					495	
Leu	Cys	Trp	Ala	Ile	Gly	Ser	Ile	Ser	Gly	Ala	Met	His	Glu	Glu	Asp
			500					505					510		
Glu	Lys	Arg	Phe	Leu	Val	Thr	Val	Ile	Lys	Asp	Leu	Leu	Gly	Leu	Cys
		515					520					525			
Glu	Gln	Lys	Arg	Gly	Lys	Asp	Asn	Lys	Ala	Ile	Ile	Ala	Ser	Asn	Ile
530						535					540				
Met	Tyr	Ile	Val	Gly	Gln	Tyr	Pro	Arg	Phe	Leu	Arg	Ala	His	Trp	Lys
545					550					555					560
Phe	Leu	Lys	Thr	Val	Val	Asn	Lys	Leu	Phe	Glu	Phe	Met	His	Glu	Thr
				565					570					575	
His	Asp	Gly	Val	Gln	Asp	Met	Ala	Cys	Asp	Thr	Phe	Ile	Lys	Ile	Ala
			580					585					590		
Gln	Lys	Cys	Arg	Arg	His	Phe	Val	Gln	Val	Gln	Val	Gly	Glu	Val	Met
		595					600					605			
Pro	Phe	Ile	Asp	Glu	Ile	Leu	Asn	Asn	Ile	Asn	Thr	Ile	Ile	Cys	Asp
610						615					620				
Leu	Gln	Pro	Gln	Gln	Val	His	Thr	Phe	Tyr	Glu	Ala	Val	Gly	Tyr	Met
625					630					635					640
Ile	Gly	Ala	Gln	Thr	Asp	Gln	Thr	Val	Gln	Glu	His	Leu	Ile	Glu	Lys
				645					650					655	
Tyr	Met	Leu	Leu	Pro	Asn	Gln	Val	Trp	Asp	Ser	Ile	Ile	Gln	Gln	Ala
			660					665					670		
Thr	Lys	Asn	Val	Asp	Ile	Leu	Lys	Asp	Pro	Glu	Thr	Val	Lys	Gln	Leu
		675					680					685			
Gly	Ser	Ile	Leu	Lys	Thr	Asn	Val	Arg	Ala	Cys	Lys	Ala	Val	Gly	His
690						695					700				

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Pro Phe Val Ile Gln Leu Gly Arg Ile Tyr Leu Asp Met Leu Asn Val
 705 710 715 720
 Tyr Lys Cys Leu Ser Glu Asn Ile Ser Ala Ala Ile Gln Ala Asn Gly
 725 730 735
 Glu Met Val Thr Lys Gln Pro Leu Ile Arg Ser Met Arg Thr Val Lys
 740 745 750
 Arg Glu Thr Leu Lys Leu Ile Ser Gly Trp Val Ser Arg Ser Asn Asp
 755 760 765
 Pro Gln Met Val Ala Glu Asn Phe Val Pro Pro Leu Leu Asp Ala Val
 770 775 780
 Leu Ile Asp Tyr Gln Arg Asn Val Pro Ala Ala Arg Glu Pro Glu Val
 785 790 795 800
 Leu Ser Thr Met Ala Ile Ile Val Asn Lys Leu Gly Gly His Ile Thr
 805 810 815
 Ala Glu Ile Pro Gln Ile Phe Asp Ala Val Phe Glu Cys Thr Leu Asn
 820 825 830
 Met Ile Asn Lys Asp Phe Glu Glu Tyr Pro Glu His Arg Thr Asn Phe
 835 840 845
 Phe Leu Leu Leu Gln Ala Val Asn Ser His Cys Phe Pro Ala Phe Leu
 850 855 860
 Ala Ile Pro Pro Ala Gln Phe Lys Leu Val Leu Asp Ser Ile Ile Trp
 865 870 875 880
 Ala Phe Lys His Thr Met Arg Asn Val Ala Asp Thr Gly Leu Gln Ile
 885 890 895
 Leu Phe Thr Leu Leu Gln Asn Val Ala Gln Glu Glu Ala Ala Ala Gln
 900 905 910
 Ser Phe Tyr Gln Thr Tyr Phe Cys Asp Ile Leu Gln His Ile Phe Ser
 915 920 925
 Val Val Thr Asp Thr Ser His Thr Ala Gly Leu Thr Met His Ala Ser
 930 935 940
 Ile Leu Ala Tyr Met Phe Asn Leu Val Glu Glu Gly Lys Ile Ser Thr
 945 950 955 960
 Pro Leu Asn Pro Gly Asn Pro Val Asn Asn Gln Met Phe Ile Gln Asp
 965 970 975
 Tyr Val Ala Asn Leu Leu Lys Ser Ala Phe Pro His Leu Gln Asp Ala
 980 985 990
 Gln Val Lys Leu Phe Val Thr Gly Leu Phe Ser Leu Asn Gln Asp Ile
 995 1000 1005
 Pro Ala Phe Lys Glu His Leu Arg Asp Phe Leu Val Gln Ile Lys
 1010 1015 1020
 Glu Phe Ala Gly Glu Asp Thr Ser Asp Leu Phe Leu Glu Glu Arg
 1025 1030 1035
 Glu Thr Ala Leu Arg Gln Ala Gln Glu Glu Lys His Lys Leu Gln
 1040 1045 1050
 Met Ser Val Pro Gly Ile Leu Asn Pro His Glu Ile Pro Glu Glu
 1055 1060 1065
 Met Cys Asp
 1070

<210> SEQ ID NO 13

<211> LENGTH: 119

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<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: hCCNT1 HDR donor template

<400> SEQUENCE: 13

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<210> SEQ ID NO 14
<211> LENGTH: 119
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: hXPO1 donor template

<400> SEQUENCE: 14

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tatcaaaggt aacagagcgg ttggttgagt gttcttctcg ttgcatactg tggttttga 119

<210> SEQ ID NO 15
<211> LENGTH: 119
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: hXPO1 donor template

<400> SEQUENCE: 15

attctctaca tctcgcgtctc cgttgcttctc tggaagtcaa cattttgatg ttctctccag 60
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<210> SEQ ID NO 16
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: CCNT1 forward screening primer

<400> SEQUENCE: 16

tgagattaga agtaggcttg agagg 25

<210> SEQ ID NO 17
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: CCNT1 reverse screening primer

<400> SEQUENCE: 17

gctaaattct cactagtccg atgac 25

<210> SEQ ID NO 18
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: XPO1 forward screening primer

<400> SEQUENCE: 18

ttctctctctc tggatggta cattt 25

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<210> SEQ ID NO 19
<211> LENGTH: 25
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: XPO1 reverse screening primer

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<400> SEQUENCE: 19

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25

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What is claimed is:

1. A genetically modified CCNT1 gene encoding a protein comprising a sequence with at least 95% sequence identity to SEQ ID NO:1 and comprising a tyrosine at a position corresponding to position 261 of SEQ ID NO:1.

2. The genetically modified CCNT1 gene of claim 1, wherein the protein encoded by the genetically modified CCNT1 gene comprises one, some, or all of: an amino acid other than glutamic acid at a position corresponding to position 3 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 29 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 31 of SEQ ID NO:1; an amino acid other than leucine and/or asparagine at a position corresponding to position 37 of SEQ ID NO:1; an amino acid other than histidine at a position corresponding to position 79 of SEQ ID NO:1; an amino acid other than arginine and glutamine and/or tyrosine at a position corresponding to position 80 of SEQ ID NO:1; an amino acid other than tyrosine at a position corresponding to position 81 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 83 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 110 of SEQ ID NO:1; an amino acid other than tyrosine at a position corresponding to position 113 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 250 of SEQ ID NO:1; an amino acid other than arginine at a position corresponding to position 256 of SEQ ID NO:1; an amino acid other than glutamine at a position corresponding to position 262 of SEQ ID NO:1; an amino acid other than methionine, arginine, and/or glutamine at a position corresponding to position 265 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 269 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 274 of SEQ ID NO:1; an amino acid other than threonine and/or alanine at a position corresponding to position 276 of SEQ ID NO:1; an amino acid other than asparagine at a position corresponding to position 277 of SEQ ID NO:1; an amino acid other than threonine at a position corresponding to position 290 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 304 of SEQ ID NO:1; an amino acid other than alanine and/or threonine at a position corresponding to position 305 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 306 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 307 of SEQ ID NO:1; an amino acid other than arginine and/or valine at a position corresponding to position 313 of SEQ ID NO:1; an amino acid other than serine, alanine, and/or valine at a position corresponding to position 315 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position

322 of SEQ ID NO:1; an amino acid other than asparagine at a position corresponding to position 325 of SEQ ID NO:1; an amino acid other than aspartic acid at a position corresponding to position 327 of SEQ ID NO:1; an amino acid other than glutamine at a position corresponding to position 330 of SEQ ID NO:1; an amino acid other than glutamic acid at a position corresponding to position 332 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 340 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 345 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 346 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 354 of SEQ ID NO:1; an amino acid other than isoleucine and/or methionine at a position corresponding to position 358 of SEQ ID NO:1; an amino acid other than glutamine at a position corresponding to position 365 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 370 of SEQ ID NO:1; an amino acid other than glycine at a position corresponding to position 373 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 378 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 443 of SEQ ID NO:1; an amino acid other than aspartic acid at a position corresponding to position 453 of SEQ ID NO:1; an amino acid other than serine and/or alanine at a position corresponding to position 458 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 464 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 468 of SEQ ID NO:1; an amino acid other than valine at a position corresponding to position 473 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 488 of SEQ ID NO:1; an amino acid other than glycine at a position corresponding to position 490 of SEQ ID NO:1; an amino acid other than isoleucine at a position corresponding to position 496 of SEQ ID NO:1; an amino acid other than glutamine at a position corresponding to position 510 of SEQ ID NO:1; an amino acid other than arginine at a position corresponding to position 511 of SEQ ID NO:1; an amino acid other than arginine at a position corresponding to position 527 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 531 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 535 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 537 of SEQ ID NO:1; an amino acid other than valine at a position corresponding to position 538 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 539 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 543 of SEQ ID NO:1; an amino acid other than

threonine at a position corresponding to position 553 of SEQ ID NO:1; an amino acid other than threonine at a position corresponding to position 564 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 565 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 577 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 582 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 603 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 606 of SEQ ID NO:1; an amino acid other than threonine and/or alanine at a position corresponding to position 611 of SEQ ID NO:1; an amino acid other than leucine at a position corresponding to position 613 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 624 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 637 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 644 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 651 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 654 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 678 of SEQ ID NO:1; an amino acid other than proline at a position corresponding to position 679 of SEQ ID NO:1; an amino acid other than aspartic acid at a position corresponding to position 682 of SEQ ID NO:1; an amino acid other than histidine at a position corresponding to position 685 of SEQ ID NO:1; an amino acid other than serine at a position corresponding to position 686 of SEQ ID NO:1; an amino acid other than glycine at a position corresponding to position 688 of SEQ ID NO:1; an amino acid other than glutamic acid at a position corresponding to position 689 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 691 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 695 of SEQ ID NO:1; an amino acid other than alanine at a position corresponding to position 697 of SEQ ID NO:1; an amino acid other than methionine at a position corresponding to position 698 of SEQ ID NO:1; an amino acid other than threonine at a position corresponding to position 704 of SEQ ID NO:1; and an amino acid other than leucine at a position corresponding to position 710 of SEQ ID NO:1.

3. The genetically modified CCNT1 gene of claim 1, wherein the protein encoded by the genetically modified CCNT1 gene comprises one, some or all of: proline at a position corresponding to position 31 of SEQ ID NO:1; tyrosine at a position corresponding to position 37 of SEQ ID NO:1; proline at a position corresponding to position 79 of SEQ ID NO:1; glycine at a position corresponding to position 80 of SEQ ID NO:1; asparagine at a position corresponding to position 81 of SEQ ID NO:1; valine at a position corresponding to position 83 of SEQ ID NO:1; threonine at a position corresponding to position 110 of SEQ ID NO:1; asparagine at a position corresponding to position 250 of SEQ ID NO:1; tryptophan at a position corresponding to position 256 of SEQ ID NO:1; glutamic acid at a position corresponding to position 262 of SEQ ID NO:1; lysine at a position corresponding to position 265 of SEQ ID NO:1; alanine at a position corresponding to position 269 of SEQ ID NO:1; threonine at a position corresponding to position 274 of SEQ ID NO:1; lysine at a position corre-

sponding to position 277 of SEQ ID NO:1; serine at a position corresponding to position 290 of SEQ ID NO:1; serine at a position corresponding to position 305 of SEQ ID NO:1; threonine at a position corresponding to position 306 of SEQ ID NO:1; threonine at a position corresponding to position 307 of SEQ ID NO:1; leucine at a position corresponding to position 313 of SEQ ID NO:1; valine at a position corresponding to position 315 of SEQ ID NO:1; serine at a position corresponding to position 316 of SEQ ID NO:1; asparagine at a position corresponding to position 322 of SEQ ID NO:1; serine at a position corresponding to position 325 of SEQ ID NO:1; glutamic acid at a position corresponding to position 327 of SEQ ID NO:1; proline at a position corresponding to position 330 of SEQ ID NO:1; lysine at a position corresponding to position 332 of SEQ ID NO:1; serine at a position corresponding to position 340 of SEQ ID NO:1; proline at a position corresponding to position 345 of SEQ ID NO:1; threonine at a position corresponding to position 346 of SEQ ID NO:1; asparagine at a position corresponding to position 354 of SEQ ID NO:1; threonine at a position corresponding to position 358 of SEQ ID NO:1; proline at a position corresponding to position 365 of SEQ ID NO:1; asparagine at a position corresponding to position 370 of SEQ ID NO:1; isoleucine at a position corresponding to position 373 of SEQ ID NO:1; asparagine at a position corresponding to position 378 of SEQ ID NO:1; histidine at a position corresponding to position 429 of SEQ ID NO:1; glycine at a position corresponding to position 443 of SEQ ID NO:1; glutamic acid at a position corresponding to position 453 of SEQ ID NO:1; threonine at a position corresponding to position 458 of SEQ ID NO:1; isoleucine at a position corresponding to position 464 of SEQ ID NO:1; glycine at a position corresponding to position 468 of SEQ ID NO:1; alanine at a position corresponding to position 473 of SEQ ID NO:1; alanine at a position corresponding to position 488 of SEQ ID NO:1; alanine at a position corresponding to position 490 of SEQ ID NO:1; valine at a position corresponding to position 496 of SEQ ID NO:1; histidine at a position corresponding to position 510 of SEQ ID NO:1; lysine at a position corresponding to position 511 of SEQ ID NO:1; lysine at a position corresponding to position 527 of SEQ ID NO:1; serine at a position corresponding to position 531 of SEQ ID NO:1; valine at a position corresponding to position 535 of SEQ ID NO:1; threonine at a position corresponding to position 537 of SEQ ID NO:1; glycine at a position corresponding to position 538 of SEQ ID NO:1; asparagine at a position corresponding to position 539 of SEQ ID NO:1; glycine at a position corresponding to position 543 of SEQ ID NO:1; asparagine at a position corresponding to position 553 of SEQ ID NO:1; serine at a position corresponding to position 564 of SEQ ID NO:1; phenylalanine at a position corresponding to position 565 of SEQ ID NO:1; serine at a position corresponding to position 577 of SEQ ID NO:1; glycine at a position corresponding to position 582 of SEQ ID NO:1; serine at a position corresponding to position 599 of SEQ ID NO:1; serine at a position corresponding to position 603 of SEQ ID NO:1; serine at a position corresponding to position 606 of SEQ ID NO:1; glycine at a position corresponding to position 611 of SEQ ID NO:1; methionine at a position corresponding to position 613 of SEQ ID NO:1; serine at a position corresponding to position 624 of SEQ ID NO:1; serine at a position corresponding to position 637 of SEQ ID NO:1; threonine at a position corresponding to position 644

of SEQ ID NO:1; threonine at a position corresponding to position 651 of SEQ ID NO:1; threonine at a position corresponding to position 654 of SEQ ID NO:1; proline at a position corresponding to position 678 of SEQ ID NO:1; threonine at a position corresponding to position 679 of SEQ ID NO:1; glutamic acid at a position corresponding to position 682 of SEQ ID NO:1; arginine at a position corresponding to position 685 of SEQ ID NO:1; proline at a position corresponding to position 686 of SEQ ID NO:1; serine at a position corresponding to position 688 of SEQ ID NO:1; aspartic acid at a position corresponding to position 689 of SEQ ID NO:1; leucine at a position corresponding to position 691 of SEQ ID NO:1; serine at a position corresponding to position 695 of SEQ ID NO:1; glycine at a position corresponding to position 697 of SEQ ID NO:1; isoleucine at a position corresponding to position 698 of SEQ ID NO:1; asparagine at a position corresponding to position 704 of SEQ ID NO:1; and proline at a position corresponding to position 710 of SEQ ID NO:1.

4. A genetically modified XPO1 gene encoding a protein comprising a sequence with at least 95% sequence identity to SEQ ID NO:7 and having at least one, at least two, or all three of:

- threonine at a position corresponding to position 411 of SEQ ID NO:7;
- valine at a position corresponding to position 412 of SEQ ID NO:7; and
- serine at a position corresponding to position 414 of SEQ ID NO:7.

5. The genetically modified XPO1 gene of claim 4, wherein the protein encoded by the genetically modified XPO1 gene comprises one, some, or all of: an amino acid other than aspartic acid at a position corresponding to position 100 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 118 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 151 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 191 of SEQ ID NO:7; an amino acid other than serine at a position corresponding to position 215 of SEQ ID NO:7; an amino acid other than glutamic acid at a position corresponding to position 284 of SEQ ID NO:7; an amino acid other than valine at a position corresponding to position 306 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 334 of SEQ ID NO:7; an amino acid other than leucine at a position corresponding to position 337 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 346 of SEQ ID NO:7; an amino acid other than isoleucine at a position corresponding to position 402 of SEQ ID NO:7; an amino acid other than isoleucine at a position corresponding to position 474 of SEQ ID NO:7; an amino acid other than lysine at a position corresponding to position 478 of SEQ ID NO:7; an amino acid other than glutamine at a position corresponding to position 481 of SEQ ID NO:7; an amino acid other than alanine at a position corresponding to position 869 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 909 of SEQ ID NO:7; an amino acid other than proline at a position corresponding to position 961 of SEQ ID NO:7; an amino acid other than serine at a position corresponding to position 966 of SEQ ID NO:7; an amino acid other than serine at a position corresponding to position 969 of SEQ ID NO:7; an amino acid other than valine and/or methionine at a position

corresponding to position 972 of SEQ ID NO:7; an amino acid other than isoleucine at a position corresponding to position 974 of SEQ ID NO:7; an amino acid other than aspartic acid at a position corresponding to position 976 of SEQ ID NO:7; an amino acid other than threonine at a position corresponding to position 1040 of SEQ ID NO:7; an amino acid other than glycine at a position corresponding to position 1043 of SEQ ID NO:7; an amino acid other than glutamine at a position corresponding to position 1046 of SEQ ID NO:7; an amino acid other than leucine at a position corresponding to position 1052 of SEQ ID NO:7; and an amino acid other than leucine at a position corresponding to position 1060 of SEQ ID NO:7.

6. The genetically modified XPO1 gene of claim 4, wherein the protein encoded by the genetically modified XPO1 gene comprises one, some, or all of: glutamic acid at a position corresponding to position 100 of SEQ ID NO:7; threonine at a position corresponding to position 118 of SEQ ID NO:7; serine at a position corresponding to position 151 of SEQ ID NO:7; serine at a position corresponding to position 191 of SEQ ID NO:7; asparagine at a position corresponding to position 215 of SEQ ID NO:7; valine at a position corresponding to position 284 of SEQ ID NO:7; leucine at a position corresponding to position 306 of SEQ ID NO:7; aspartic acid at a position corresponding to position 334 of SEQ ID NO:7; isoleucine at a position corresponding to position 337 of SEQ ID NO:7; threonine at a position corresponding to position 346 of SEQ ID NO:7; valine at a position corresponding to position 402 of SEQ ID NO:7; arginine at a position corresponding to position 474 of SEQ ID NO:7; glutamic acid at a position corresponding to position 478 of SEQ ID NO:7; histidine at a position corresponding to position 481 of SEQ ID NO:7; threonine at a position corresponding to position 869 of SEQ ID NO:7; alanine at a position corresponding to position 909 of SEQ ID NO:7; serine at a position corresponding to position 961 of SEQ ID NO:7; asparagine at a position corresponding to position 966 of SEQ ID NO:7; asparagine at a position corresponding to position 969 of SEQ ID NO:7; isoleucine at a position corresponding to position 972 of SEQ ID NO:7; leucine at a position corresponding to position 974 of SEQ ID NO:7; glutamic acid at a position corresponding to position 976 of SEQ ID NO:7; isoleucine at a position corresponding to position 1040 of SEQ ID NO:7; arginine at a position corresponding to position 1043 of SEQ ID NO:7; aspartic acid at a position corresponding to position 1046 of SEQ ID NO:7; arginine at a position corresponding to position 1052 of SEQ ID NO:7; and phenylalanine at a position corresponding to position 1060 of SEQ ID NO:7.

7. A genetically modified cell comprising at least one of:

one or more copies of a genetically modified CCNT1 gene encoding a protein comprising a sequence with at least 95% sequence identity to SEQ ID NO:1 and comprising a tyrosine at a position corresponding to position 261 of SEQ ID NO:1; and

one or more copies of a genetically modified XPO1 gene encoding a protein comprising a sequence with at least 95% sequence identity to SEQ ID NO:7 and having at least one, at least two, or all three of:

- threonine at a position corresponding to position 411 of SEQ ID NO:7;
- valine at a position corresponding to position 412 of SEQ ID NO:7; and

- serine at a position corresponding to position 414 of SEQ ID NO:7.
8. The cell of claim 7, wherein the cell is an immune cell or a precursor of an immune cell.
9. The cell of claim 7, wherein the cell is selected from the group consisting of a hematopoietic stem cell, a myeloid progenitor cell, a lymphoid progenitor cell, a myeloblast, a monocyte, a macrophage, a dendritic cell, a small lymphocyte, a T cell, and an astrocyte.
10. The cell of claim 7, wherein the cell is a T cell or a precursor thereof.
11. The cell of claim 7, wherein the cell is a CD4+T cell or a precursor thereof.
12. The cell of claim 7, wherein the cell is a mammalian cell.
13. The cell of claim 7, wherein the cell comprises at least one of:
two copies of the genetically modified CCNT1 gene; and
two copies of the genetically modified XPO1 gene.
14. The cell of claim 7, wherein the cell is devoid of at least one of:
a CCNT1 gene having an amino acid other than a tyrosine at a position corresponding to position 261 of SEQ ID NO:1; and
an XPO1 gene having at least one, at least two, or all three of an amino acid other than a threonine at a position corresponding to position 411 of SEQ ID NO:7, an amino acid other than a methionine at a position corresponding to position 412 of SEQ ID NO:7, and an amino acid other than a phenylalanine at a position corresponding to position 414 of SEQ ID NO:7.
15. A method of treating a subject infected with a virus, the method comprising introducing the genetically modified cell of claim 7 in the subject, wherein the genetically modified cell is of a cell type susceptible to infection with the virus or a precursor of a cell type susceptible to infection with the virus.
16. The method of claim 15, wherein the subject is a mammal.
17. The method of claim 15, wherein the subject is a human.
18. The method of claim 15, wherein the virus is selected from the group consisting of a lentivirus and a deltaretrovirus.
19. The method of claim 15, wherein the virus is selected from the group consisting of a primate immunodeficiency virus and a primate T-lymphotropic virus.
20. The method of claim 15, wherein the virus is selected from the group consisting of a human immunodeficiency virus and a human T-lymphotropic virus.
21. The method of claim 15, wherein the virus is a human immunodeficiency virus.
22. The method of claim 15, wherein the cell is autologous to the subject.
23. The method of claim 15, wherein the introducing comprises introducing the cell into the bloodstream of the subject.
24. The method of claim 15, wherein the introducing comprises injecting or infusing the cell into the bloodstream of the subject.

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