10-11-2016

Methods of inhibiting Alu RNA and Therapeutic Uses Thereof

Jayakrishna Ambati
University of Kentucky, jayakrishna.ambati@uky.edu

Click here to let us know how access to this document benefits you.

Follow this and additional works at: https://uknowledge.uky.edu/ophthalmology_patents

Part of the Ophthalmology Commons

Recommended Citation
Ambati, Jayakrishna, "Methods of inhibiting Alu RNA and Therapeutic Uses Thereof" (2016). Ophthalmology and Visual Science Faculty Patents. 22.
https://uknowledge.uky.edu/ophthalmology_patents/22

This Patent is brought to you for free and open access by the Ophthalmology and Visual Science at UKnowledge. It has been accepted for inclusion in Ophthalmology and Visual Science Faculty Patents by an authorized administrator of UKnowledge. For more information, please contact UKnowledge@lsv.uky.edu.
METHODS OF INHIBITING ALU RNA AND THERAPEUTIC USES THEREOF

Applicant: University of Kentucky Research Foundation, Lexington, KY (US)

Inventor: Jayakrishna Ambati, Lexington, KY (US)

Assignee: University of Kentucky Research Foundation, Lexington, KY (US)

Notice: Subject to any disclaimer, the term of this patent is subject to a terminal disclaimer.

Appl. No.: 14/323,457
Filed: Jul. 3, 2014

Prior Publication Data

Related U.S. Application Data
Division of application No. 13/701,450, filed as application No. PCT/US2011/038753 on Jun. 1, 2011, now Pat. No. 8,809,517.

Int. Cl.
C12N 15/11 (2006.01)
C12N 15/113 (2010.01)
C12Q 1/68 (2006.01)
C12N 9/22 (2006.01)
G01N 33/53 (2006.01)

U.S. Cl.
C12N 15/113 (2013.01)
C12Q 1/6804 (2013.01)
C12Q 1/6876 (2013.01)
C12Q 1/6883 (2013.01)
G01N 33/5308 (2013.01)
C12N 2310/113 (2013.01)
C12N 2320/30 (2013.01)
C12Q 2600/166 (2013.01)
C12Q 2600/158 (2013.01)
C12Y 30/26003 (2013.01)
G01N 2500/10 (2013.01)

Field of Classification Search
None
See application file for complete search history.

References Cited
FOREIGN PATENT DOCUMENTS

OTHER PUBLICATIONS

(Continued)

Primary Examiner — Kate Poliaikova-Georgantas
Attorney, Agent, or Firm — Stites & Harbison PLLC; Mandy Wilson Decker

ABSTRACT

The presently-disclosed subject matter includes methods of identifying an Alu RNA inhibitor, and methods and compositions for inhibiting Alu RNA. Methods and compositions can be used for the treatment of geographic atrophy and other conditions of interest.
References Cited

OTHER PUBLICATIONS


* cited by examiner
FIG. 1
FIG. 1, Continued
FIG. 3
FIG. 4
FIG. 5
FIG. 7

a) Fold change in DICER1 RNA

b) DICER1
c) isotype

GA

d) Normal
e) Normal

FIG. 7
FIG. 8

- Best disease
- Retinal detachment
- Retinitis pigmentosa

Fold change in Dicer1 mRNA

<table>
<thead>
<tr>
<th></th>
<th>0</th>
<th>0.5</th>
<th>1</th>
<th>10</th>
<th>15</th>
</tr>
</thead>
<tbody>
<tr>
<td>wild-type</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ccr2/Col2-</td>
<td>*</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>wild-type</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cp/Heph-</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

NS
FIG. 9
FIG. 14
FIG. 15
FIG. 16

FIG. 17
FIG. 19

<table>
<thead>
<tr>
<th></th>
<th>Normal</th>
<th>GA</th>
</tr>
</thead>
<tbody>
<tr>
<td>L1.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HERV-WE1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>hY3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>DICER1 as</th>
<th>Ctrl as</th>
</tr>
</thead>
<tbody>
<tr>
<td>L1.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>HERV-WE1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>hY3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Fold change
FIG. 20

FIG. 21
FIG. 23

![Graph showing fold change in Alu RNA over time.]

FIG. 24

![Images comparing different treatments: (a) Human GA csRNA vs Vehicle vs DICER1-cleaved Human GA Alu csRNA vs Mock DICER1-cleaved Human GA Alu csRNA.]

FIG. 25

![Images showing different RNA targets: p7SL, tRNA, pmiR29b1, pmiR26a2.]

FIG. 24
FIG. 27

(a) Geographic atrophy

(b) Best1 Cre; Dicer1Δ

(c) pAlu

(d) Fold change in caspase-3 activation

(e) Fold change in caspase-3 activation
FIG. 28

(a) Human RPE cell viability (%)

- pNull
- pAlu
- pAlu + Alu Frag

(b) Alu Frag vs. Alu Frag + pAlu

Alu Frag

Alu Frag + pAlu
METHODS OF INHIBITING ALU RNA AND THERAPEUTIC USES THEREOF

RELATED APPLICATIONS

This application is a divisional of U.S. patent application Ser. No. 13/701,450, now allowed, which is a 371 application of International Patent Application No. PCT/US2011/038753, filed Jun. 1, 2011, which claims priority from U.S. Provisional Application Ser. No. 61/396,747, filed on Jun. 1, 2010; U.S. Provisional Application Ser. No. 61/432,110, filed Jan. 12, 2011; and U.S. Provisional Application Ser. No. 61/432,948, filed Jan. 14, 2011. The entire disclosures of these applications are incorporated herein by this reference.

TECHNICAL FIELD

The presently-disclosed subject matter relates to uses of DICER overexpression and the inhibition of Alu RNA.

INTRODUCTION

Geographic atrophy, an advanced form of age-related macular degeneration that causes blindness in millions of people worldwide and for which there is no approved treatment, results from death of retinal pigmented epithelium (RPE) cells. As described herein the present inventors show that expression of DICER, an enzyme involved in microRNA (miRNA) biogenesis, is reduced in the RPE of human eyes with geographic atrophy, and that conditional ablation of Dicer1 induces RPE degeneration in mice. Surprisingly, ablation of seven other enzymes responsible for miRNA biogenesis or function does not induce such pathology. Instead, knockdown of DICER1 leads to accumulation of Alu repeat RNA in human RPE cells and of B1 and B2 (Alu-like elements) repeat RNAs in the RPE of mice.

Alu RNA is dramatically increased in the RPE of human eyes with geographic atrophy, and introduction of this pathological RNA induces death of human RPE cells and RPE degeneration in mice. Antisense oligonucleotides targeting Alu/B1/B2 RNAs inhibit DICER1 depletion-induced RPE degeneration despite persistence of global miRNA downregulation. DICER1 degrades Alu RNA, and Alu RNA loses the ability to induce RPE degeneration in mice when digested by DICER1. These findings reveal a novel miRNA-independent cell survival function for DICER1 via degradation of retrotransposon transcripts, introduce the concept that Alu RNA can directly cause human pathology, and identify new molecular targets for treating a major cause of blindness.

Age-related macular degeneration (AMD), which is as prevalent as cancer in industrialized countries, is a leading cause of blindness worldwide. In contrast to the neovascular form of AMD, for which many approved treatments exist, the far more common atrophic form of AMD remains poorly understood and without effective clinical intervention. Extensive atrophy of the retinal pigment epithelium (RPE) leads to severe vision loss and is termed geographic atrophy, the pathogenesis of which is unclear. As described herein, the present inventors identify dysregulation of the RNase DICER1 and the resulting accumulation of transcripts of Alu elements, the most common small interspersed repetitive elements in the human genome, as a cause of geographic atrophy, and describe treatment strategies to inhibit this pathology in vivo.

METHODS OF INHIBITING ALU RNA AND THERAPEUTIC USES THEREOF

This application is a divisional of U.S. patent application Ser. No. 13/701,450, now allowed, which is a 371 application of International Patent Application No. PCT/US2011/038753, filed Jun. 1, 2011, which claims priority from U.S. Provisional Application Ser. No. 61/396,747, filed on Jun. 1, 2010; U.S. Provisional Application Ser. No. 61/432,110, filed Jan. 12, 2011; and U.S. Provisional Application Ser. No. 61/432,948, filed Jan. 14, 2011. The entire disclosures of these applications are incorporated herein by this reference.

TECHNICAL FIELD

The presently-disclosed subject matter relates to uses of DICER overexpression and the inhibition of Alu RNA.

INTRODUCTION

Geographic atrophy, an advanced form of age-related macular degeneration that causes blindness in millions of people worldwide and for which there is no approved treatment, results from death of retinal pigmented epithelium (RPE) cells. As described herein the present inventors show that expression of DICER, an enzyme involved in microRNA (miRNA) biogenesis, is reduced in the RPE of human eyes with geographic atrophy, and that conditional ablation of Dicer1 induces RPE degeneration in mice. Surprisingly, ablation of seven other enzymes responsible for miRNA biogenesis or function does not induce such pathology. Instead, knockdown of DICER1 leads to accumulation of Alu repeat RNA in human RPE cells and of B1 and B2 (Alu-like elements) repeat RNAs in the RPE of mice.

Alu RNA is dramatically increased in the RPE of human eyes with geographic atrophy, and introduction of this pathological RNA induces death of human RPE cells and RPE degeneration in mice. Antisense oligonucleotides targeting Alu/B1/B2 RNAs inhibit DICER1 depletion-induced RPE degeneration despite persistence of global miRNA downregulation. DICER1 degrades Alu RNA, and Alu RNA loses the ability to induce RPE degeneration in mice when digested by DICER1. These findings reveal a novel miRNA-independent cell survival function for DICER1 via degradation of retrotransposon transcripts, introduce the concept that Alu RNA can directly cause human pathology, and identify new molecular targets for treating a major cause of blindness.

Age-related macular degeneration (AMD), which is as prevalent as cancer in industrialized countries, is a leading cause of blindness worldwide. In contrast to the neovascular form of AMD, for which many approved treatments exist, the far more common atrophic form of AMD remains poorly understood and without effective clinical intervention. Extensive atrophy of the retinal pigment epithelium (RPE) leads to severe vision loss and is termed geographic atrophy, the pathogenesis of which is unclear. As described herein, the present inventors identify dysregulation of the RNase DICER1 and the resulting accumulation of transcripts of Alu elements, the most common small interspersed repetitive elements in the human genome, as a cause of geographic atrophy, and describe treatment strategies to inhibit this pathology in vivo.

METHODS OF INHIBITING ALU RNA AND THERAPEUTIC USES THEREOF

This application is a divisional of U.S. patent application Ser. No. 13/701,450, now allowed, which is a 371 application of International Patent Application No. PCT/US2011/038753, filed Jun. 1, 2011, which claims priority from U.S. Provisional Application Ser. No. 61/396,747, filed on Jun. 1, 2010; U.S. Provisional Application Ser. No. 61/432,110, filed Jan. 12, 2011; and U.S. Provisional Application Ser. No. 61/432,948, filed Jan. 14, 2011. The entire disclosures of these applications are incorporated herein by this reference.

TECHNICAL FIELD

The presently-disclosed subject matter relates to uses of DICER overexpression and the inhibition of Alu RNA.

INTRODUCTION

Geographic atrophy, an advanced form of age-related macular degeneration that causes blindness in millions of people worldwide and for which there is no approved treatment, results from death of retinal pigmented epithelium (RPE) cells. As described herein the present inventors show that expression of DICER, an enzyme involved in microRNA (miRNA) biogenesis, is reduced in the RPE of human eyes with geographic atrophy, and that conditional ablation of Dicer1 induces RPE degeneration in mice. Surprisingly, ablation of seven other enzymes responsible for miRNA biogenesis or function does not induce such pathology. Instead, knockdown of DICER1 leads to accumulation of Alu repeat RNA in human RPE cells and of B1 and B2 (Alu-like elements) repeat RNAs in the RPE of mice.

Alu RNA is dramatically increased in the RPE of human eyes with geographic atrophy, and introduction of this pathological RNA induces death of human RPE cells and RPE degeneration in mice. Antisense oligonucleotides targeting Alu/B1/B2 RNAs inhibit DICER1 depletion-induced RPE degeneration despite persistence of global miRNA downregulation. DICER1 degrades Alu RNA, and Alu RNA loses the ability to induce RPE degeneration in mice when digested by DICER1. These findings reveal a novel miRNA-independent cell survival function for DICER1 via degradation of retrotransposon transcripts, introduce the concept that Alu RNA can directly cause human pathology, and identify new molecular targets for treating a major cause of blindness.

Age-related macular degeneration (AMD), which is as prevalent as cancer in industrialized countries, is a leading cause of blindness worldwide. In contrast to the neovascular form of AMD, for which many approved treatments exist, the far more common atrophic form of AMD remains poorly understood and without effective clinical intervention. Extensive atrophy of the retinal pigment epithelium (RPE) leads to severe vision loss and is termed geographic atrophy, the pathogenesis of which is unclear. As described herein, the present inventors identify dysregulation of the RNase DICER1 and the resulting accumulation of transcripts of Alu elements, the most common small interspersed repetitive elements in the human genome, as a cause of geographic atrophy, and describe treatment strategies to inhibit this pathology in vivo.
embodiments, increasing levels of a DICER polypeptide compromises overexpressing the DICER polypeptide in the cells. In some embodiments, increasing levels of a DICER polypeptide comprises using a vector comprising a nucleotide encoding the DICER polypeptide. In some embodiments, the vector is a viral vector. In some embodiments, the virus is selected from an adeno-associated virus, a lentivirus, and an adenovirus. In some embodiments, the vector is a plasmid vector. In some embodiments, the nucleotide encoding the DICER polypeptide is selected from SEQ ID NO: 7 and SEQ ID NO: 8. In some embodiments, the DICER polypeptide is selected from SEQ ID NO: 9, 10, 11, 12, 13, 14, 15, 16, 18, and 20. In some embodiments, the DICER polypeptide comprises a functional fragment of the sequence of SEQ ID NO: 9, 18, or 20. In some embodiments, the DICER polypeptide comprises the following amino acid residues of the polypeptide of SEQ ID NO: 9: 605-1922, 605-1912, 1666-1922, 1666-1912, 605-1786 and 1800-1922, 605-1786 and 1800-1912, 1666-1786 and 1800-1922, 1666-1786 and 1800-1912, 1276-1922, 1276-1912, 1276-1786 and 1800-1922, 1276-1786, 800-1912, 1275-1824, or 1276-1824.

In some embodiments of the methods of the presently disclosed subject matter including inhibiting Alu RNA associated with a cell, the inhibiting Alu RNA comprises increasing levels of a DICER polypeptide comprises using DICER mRNA or a functional fragment thereof. In some embodiments, the DICER mRNA has the sequence of SEQ ID NO: 17, 19, or 21. In some embodiments, the DICER mRNA encodes a DICER polypeptide, for example, the DICER polypeptide of SEQ ID NO: 9, 18, or 20, or a functional fragment thereof.

In some embodiments of the methods of the presently disclosed subject matter including inhibiting Alu RNA associated with a cell, the inhibiting Alu RNA comprises administering an oligonucleotide targeting Alu RNA. In some embodiments, the oligonucleotide has a sequence including a sequence selected from SEQ ID NO: 22, 23, 24, 25, and 26. In some embodiments, at least two oligonucleotides are administered. The presently-disclosed subject matter further includes an isolated oligonucleotide that inhibits the expression of Alu RNA, including a sequence selected from SEQ ID NO: 22, 23, 24, 25, and 26 and including about 29 to 100 nucleotides.

In some embodiments of the methods of the presently disclosed subject matter including inhibiting Alu RNA associated with a cell, the inhibiting Alu RNA comprises administering an siRNA targeting Alu RNA. In some embodiments, the siRNA includes a first strand having a sequence selected from SEQ ID NO: 1, 2, 3, 4, 5, and 6. The presently-disclosed subject matter further includes an isolated double-stranded RNA molecule that inhibits expression of Alu RNA, wherein a first strand of the double-stranded RNA comprises a sequence selected from SEQ ID NO: 1, 2, 3, 4, 5, and 6 and including about 19 to 25 nucleotides.

**BRIEF DESCRIPTION OF THE DRAWINGS**

**FIG. 1 DICER1 deficit in geographic atrophy induces RPE degeneration.** a, DICER1 mRNA abundance, relative to 18S rRNA, monitored by real-time RT-PCR, was lower in the retinal pigmented epithelium (RPE) of a human eye with geographic atrophy (GA; n=10) compared to the RPE of normal human eyes without GA (n=11). P=0.004 by Mann Whitney U test. The abundance of DROSHA, DGCR8, and EIF2C2 (encoding AGO2) mRNA transcripts in the RPE was not significantly different (P>0.11 by Mann Whitney U test) in human eyes with geographic atrophy and control eyes. Transcript abundance quantified by real-time RT-PCR and normalized to 18S rRNA and to control eye levels. n=10-11. b, Relative quantification of DICER1 protein abundance, relative to Vinculin, assessed by Western blotting (Supplementary FIG. 1), was lower in the RPE of human eyes with geographic atrophy (GA; n=4) compared to the RPE of normal human eyes without GA (n=4). P=0.005 by Student t test. c, Immunohistochemistry for DICER1 (blue) showed reduced protein abundance in the RPE of human eyes with GA compared to normal eyes without GA. d, Fundus photographs show extensive RPE degeneration in BEST1 Cre; Dicer1/ mice but not in littermate control mice. e, Toluidine blue-stained sections show marked RPE degeneration in BEST1 Cre; Dicer1/ mice compared to normal RPE architecture (red, arrowheads point to basal surface of RPE). f, Flat mounts of the RPE and choroid stained with antibodies against zonula occludens-1 (ZO-1; red) show marked disruption of the RPE monolayer architecture in BEST1 Cre; Dicer1/ mice compared to the uniformly tesselated RPE layer in littermate control mice. g, Fundus photographs show RPE degeneration in Dicer1/ mice following subretinal injection of AAV1-BEST1-Cre but not AAV1-BEST1-GFP. h, Toluidine blue-stained sections show marked degeneration of RPE and photoreceptor outer segments in Dicer1/ mice following subretinal injection of AAV1-BEST1-Cre but not AAV1-BEST1-GFP. i, Flat mounts show marked increase in RPE cell size and distortion of RPE cell shape in Dicer1/ mice following subretinal injection of AAV1-BEST1-Cre but not AAV1-BEST1-GFP. RPE cell borders outlined by ZO-1 staining (red). Nuclei stained blue with Hoechst 33342.

**FIG. 2 Alu RNA accumulation in geographic atrophy**

Representative images shown. n=1-6 (d-i); 10-12 (g-i). Scale bars, (c,e,h), 10 μm; (f,i) 20 μm. j, Transfection of adenoviral vector coding for Cre recombinase (Ad-Cre) in RPE cells isolated from Dicer1/ mice resulted in loss of cell viability, as monitored by MTS assay at 7 days, compared to transfection with Ad-Null or untreated (no Tx) cells. k, Transfection of antisense oligonucleotide (as) targeting DICER1 into human RPE cells resulted in increasing loss of cell viability over time compared to scrambled sequence antisense (Ctrl as)-treated cells. n=6-8.
difference in Alu RNA abundance in the neural retina of these two patient groups. Values normalized to relative abundance in normal eyes.

FIG. 3 DICER1 degrades Alu RNA. a, Transfection of antisense oligonucleotide (as) targeting DICER1 into human RPE cells induced a time-dependent increase in the abundance of Alu RNA transcripts. b, c, Transfection of adenoviral vector encoding for Cre recombinase (Ad-Cre) into mouse RPE cells isolated from Dicer1−/− mice, increased, in the nucleus (b) and the cytoplasm (c), the abundance of B1 and B2 RNAs, the Alu-like repetitive elements in the mouse, compared to cells transfected with adenoviral vector coding for green fluorescent protein (Ad-GFP). d, DICER1 as treatment of human RPE cells upregulated Alu RNA levels in both the nucleus (Nuc) and cytoplasm (Cyt). e, Alu RNA isolated and cloned from the RPE of human eye with geographic atrophy was degraded by recombinant DICER1 digestion (+) as visualized by agarose gel electrophoresis. Digestion with heat denatured DICER1 did not degrade Alu RNA. Image representative of 6 experiments. f, The increased abundance of Alu RNA in human RPE cells transfected with plasmid coding for Alu (pAlu) compared to pNull or no treatment (no Tx) at 24 h was reduced by co-transfection with pDICER1. *P<0.05. n=4-8 (a-d, f). RNA abundance was quantified by real-time RT-PCR, normalized to 18S rRNA levels, and normalized to levels in control as-treated (for Alu) or Ad-GFP-infected cells (for B elements).

FIG. 4 DICER1 protects RPE cells from Alu RNA cytotoxicity. a, Transfection of mouse or human retinal pigmented epithelium cells (mRPE or hRPE) with plasmid coding for Alu RNA (pAlu) compromised cell viability. b, Subretinal administration of pAlu induced RPE degeneration in wild-type mice whereas pNull did not do so. Fundus photograph (top row) shows area of degeneration in pAlu injected eye compared to the normal appearance in pNull. Flat mount preparations stained with anti-zonula occludens-1 antibody (ZO-1, red, bottom row) show marked distortion of RPE cell shape and size compared to pNull-injected eye. c, Alu RNA induced dose-dependent increase in cell death of human RPE cells. d, Cell death of human RPE cells induced by transfection of pAlu was inhibited by co-transfection with pDICER1 but not pNull. (a,d) Cell viability monitored by MTS assay at 2 days. Values normalized to null plasmid (pNull) transfected or vehicle treated cells. *P<0.05 by Student t test. n=4-6. e, Subretinal co-administration of pDICER1, but not of pNull, inhibited pAlu induced RPE degeneration in wild-type mice. f, Subretinal administration of Alu RNA isolated and cloned from the RPE of a human eye with geographic atrophy (GA) induced RPE degeneration in wild-type mice whereas subretinal injection of vehicle did not. g, Subretinal injection of this Alu RNA, when subjected to cleavage by DICER1, did not induce RPE degeneration in wild-type mice whereas Alu RNA subjected to mock cleavage by DICER1 did do so, as evident on fundus photography (top row) or flat mount preparation (bottom row). Area of degeneration outlined by blue arrowheads in fundus photographs (b, e-g). Scale bars (20 μm). n=10-15 (b, e-g).

FIG. 5 DICER1 downregulation induces RPE cell death via Alu RNA accumulation. a, Loss of human RPE cell viability, as monitored by MTS assay, induced by transfection of antisense oligonucleotide (as) targeting DICER1 was rescued by co-transfection of Alu RNA as. Levels normalized or compared to transfection with control (Ctrl) antisense oligonucleotide. b, Alu RNA as inhibition accumulation of Alu RNA induced by DICER1 as. c, Ad-Cre but not Ad-Null induced loss of cell viability of Dicer1−/− mouse RPE cells. This was rescued by transfection of antisense oligonucleotide targeting B1 and B2 RNAs but not by control (Ctrl) antisense oligonucleotide. Levels normalized to untreated cells (no Tx), d, B1/B2 RNA as inhibited accumulation of B1 and B2 RNAs induced by Ad-Cre-induced Dicer1 depletion. *P<0.05 by Student t test. n=4-6 (a-d). d, Subretinal AAV-BEST1-Cre administration induced RPE degeneration (blue arrowheads in fundus photograph on top row and marked increase in RPE cell size and distortion of RPE cell shape in ZO-1 stained (red) RPE flat mounts (bottom row) in Dicer1−/− mice 20 days after injection. Subretinal administration of cholesterol-conjugated B1/B2 as, but not Ctrl as, 10 days after AAV-BEST1-Cre injection inhibited RPE degeneration (e) and abundance of B1/B2 RNAs in the RPE of these mice, as monitored by real-time RT-PCR at 10 days after as injection, normalized to 18S rRNA levels, and normalized to levels in eyes treated with cholesterol-conjugated Ctrl as (f). n=8 (e,f). Scale bar, 20 μm. (e, g, DICER1 as treatment of human RPE cells led to global reduction of miRNA abundance at 2 days compared to Ctrl as. There was no significant difference in miRNA abundance between Alu as and Ctrl as-treated Dicer1 depleted cells. n=3.

FIG. 6 DICER1 levels in RPE are reduced in geographic atrophy. Western blots of macular RPE lysates from individual human donor eyes show that DICER1 protein abundance, normalized to the levels of the housekeeping protein Vinculin, are reduced in geographic atrophy (GA) compared to age-similar control human eyes without age-related macular degeneration.

FIG. 7 DICER1 levels in neural retina are unchanged in geographic atrophy. a, DICER1 miRNA abundance in the neural retina, as monitored by real-time RT-PCR, was not significantly different (P>0.05 by Mann Whitney U test) between normal human retina and those with geographic atrophy. Levels normalized to 18S rRNA abundance and to normal retinas. n=7, b-e, DICER1 protein immunolocalization in the neural retina was not different between human eyes with geographic atrophy (b) and normal (d) eyes. Specificity of DICER1 staining was confirmed by absence of reaction production with isotype control antibody (c,e). Representative images shown. n=8. Scale bars (20 μm, b-e).

FIG. 8 DICER1 is not generically downregulated in retinal diseases. Immunolocalization studies revealed abundant DICER1 protein expression (blue, left column) in the RPE in the eye of an 85-year-old man with Best disease (vitrelliform macular dystrophy), a 68-year-old man with retinal detachment secondary to choroidal melanoma, and a 72-year-old woman with retinitis pigmentosa. Specificity of DICER1 staining was confirmed by absence of reaction production with isotype control antibody (right column). Representative images shown. n=13. Scale bars (10 μm). DICER1 mRNA expression in the RPE was not significantly (NS) different in Ccl2−/− Ccr2−/− mice or Cpt1−/− Hep−− mice compared to their background strains. Transcript abundance quantified by real-time RT-PCR and normalized to 18S rRNA and to control eye levels. n=6. NS, not significant.

FIG. 9 Cre recombinase expression does not induce retinal pigmented epithelium (RPE) degeneration. Subretinal administration of adeno-associated viral vector coding for Cre recombinase directed by the BEST1 promoter (AAV1-BEST1-Cre) in wild-type mice did not induce retinal toxicity that was evident on fundus photography (top left) and did not disrupt the tiling pattern of the RPE monolayer (top right). Circular flash artifact is seen in the centre of the fundus photographs. RPE cell borders delineated by staining with anti-ZO-1 antibody (red) and nuclei stained by Hoechst
33342 (blue). RPE flat mounts show successful Cre recombinase expression (red) following subretinal injection of AAV1-BEST1-Cre in wild-type (bottom left) and Dicer1/−/− (bottom right) mouse eyes. Representative images shown. n=3-10. Scale bar (20 μm).

FIG. 10 Retinal pigmented epithelium (RPE) cell dysmorphism in human age-related macular degeneration eye with atrophy. In contrast to the well tessellated RPE cell monolayer observed in a normal human eye (right), marked changes in RPE cell size and shape are observed in the human eye with geographic atrophy (left). These changes resemble those observed in eyes of mice wherein Dicer1 has been depleted in the RPE. RPE cell borders delineated by staining with anti-ZO-1 antibody (green) and nuclei stained by propidium iodide (red). Representative image shown. n=8. Scale bar, 50 μm.

FIG. 11 Conditional ablation of Drosha, Dgcr8, or Ago2 in the retinal pigmented epithelium (RPE) does not induce degeneration as seen in Dicer1-ablated mice. Fundus photographs (left column) show no significant degeneration following subretinal injection of AAV-BEST1-Cre in mice “fooled” for Drosha, Dgcr8, or Ago2. Circular flash artifacts are seen near the centre of the fundus photographs. Injection site wound appears white in the fundus photograph of the Ago2/−/− eye. RPE flat mounts (middle column) stained with anti-ZO-1 antibody (red) and Hoechst 33342 (blue) show normal tiling pattern of RPE with no gross disturbance of cell size or shape. RPE flat mounts (right column) stained with anti-Cre recombinase antibody (red) and Hoechst 33342 (blue) shows successful Cre expression in these mice eyes. Representative images shown. n=8-12. Scale bar (20 μm).

FIG. 12 Deficiency of Ago1, Ago3, Ago4, or Tarbp2 does not induce RPE degeneration. Mice deficient in Ago1 Ago3 Ago4, or Tarbp2 have normal retinal appearance on fundus photography (top row) and normal RPE monolayer architecture on ZO-1 stained (red) flat mounts (bottom row). Circular flash artifact is seen in the centre of the fundus photographs. Scale bar, 20 μm.

FIG. 13 Dicer1 mutant cells impaired in miRNA biogenesis do not have compromised cell viability. There was no difference in baseline cell viability between HCT-Dicer1−/− cells, which are impaired in miRNA biogenesis, and parent HCT116 cells over 3 days of analysis of cell proliferation. n=3. NS, not significant.

FIG. 14 Human geographic atrophy eye retinal pigmented epithelium contain Alu RNA sequences. a, Top: Typical Alu element with conserved structural regions (adapted from ref. 2). The left arm consists of RNA polymerase III binding sites (Box A and Box B). The right arm occasionally contains a terminal poly A tail that may be interspersed with non-A bases. The 5′ and 3′ regions of the Alu element are linked by a mid-stretch A-rich sequence. Bottom: Representative Alu cDNA (Sequence 1). The conserved regions mentioned above are highlighted and correspond to the coloured boxes in the top figure. b, Alignment of Alu cDNA Sequences 1 and 2 isolated from human eyes with geographic atrophy to Alu Sq consensus sequence. These sequences contain the highly conserved 5′ Alu consensus elements (5′ characteristic Alu region—blue; RNA polymerase III promoter B box—red), with extensive heterogeneity located 3′ to the mid-sequence poly-A stretch that have been reported to exist in Alu sequences.4,5

FIG. 15 J2 anti-dsRNA antibody recognizes Alu RNA. a, Alu RNA duplex isolated and cloned from the retinal pigmented epithelium (RPE) of a human eye with geographic atrophy was recognized by J2 anti-dsRNA antibody in an immuno-dot blot format. J2 antibody did not recognize rRNA or tRNA (negative controls), but did recognize RNA duplexes of 325-bp or 1-kbp in length (positive controls). b, Immunofluorescent imaging of human RPE cells transfected with pAlu shows that J2 recognizes Alu expressed in these cells (left panel). Specificity of staining confirmed by absence of staining with isotype control antibody (middle panel) and by the absence of immunodetection following transfection with pNull (right panel). Representative images shown. n=3. Scale bar (20 μm).

FIG. 16 Confirmation of lack of DNA contamination in Alu RNA PCR. The relative abundance of Alu RNA in the RPE of human eyes with geographic atrophy was presented in FIG. 2/7. Shown above is the detection of the PCR product band for a sample of human geographic atrophy RPE that underwent reverse transcription (RT+). No amplification was detected in the negative controls where reverse transcriptase (RT–) was omitted or where water alone was analyzed. These data demonstrate the absence of DNA contamination in the sample.

FIG. 17 Validation of Dicer1 knockdown. Transfection of Dicer1 antisense oligonucleotides (as) into human RPE cells knocks down Dicer1 protein abundance, as monitored by Western blot analysis, over 2 days. Efficiency of protein loading is monitored by blotting for the housekeeping Vinculin protein. Representative of 3 experiments.

FIG. 18 Dicer1 is expressed in nucleus and cytoplasm. a, Western blot shows expression of Dicer1 in both the nuclear and cytoplasmic fractions of human RPE cells. Blotting of the same protein sample reveals the presence of Tubulin in the cytoplasmic fraction and not in the nuclear fraction. b, Merged images (bottom row) of Dicer1 immunofluorescence (red, top row) and nuclear DAPI fluorescence (middle row) confirms expression of Dicer1 in both the nucleus and the cytoplasm of human RPE cells. Representative images shown. Scale bar, 10 μm.

FIG. 19 Retrotransposons and repetitive RNAs are not generically activated in geographic atrophy or by Dicer1 depletion. In the RPE of human eyes with geographic atrophy (GA, n=7), there was no significant increase in the abundance of RNAs coded by LINE I1.3, a long interspersed repetitive element, human endogenous retrovirus-W envelope (HERV-WE1), a long terminal repeat retrotransposon, or hY3, a repetitive small cytoplasmic Ro RNA compared to normal human eyes (top, n=8). These RNAs also were not upregulated by Dicer1 antisense (as) knockdown, compared to control (Ctrl) as treatment, in human RPE cells (bottom). n=3. Transcript abundance monitored by real-time RT-PCR and normalized to 18S rRNA levels.

FIG. 20 Alu RNA induced by Dicer1 depletion is RNA Pol III derived. a, The upregulation of Alu RNA in RPE cells treated with antisense (as) oligonucleotides targeting Dicer1, compared to control (Ctrl), is inhibited by treatment with the Pol III inhibitor tagetitoxin (tagetin), but not by the Pol II inhibitor α-amanitin. *, P<0.05. NS, not significant, compared to treatment with Dicer1 as treatment alone. b, Northern blot (NB) shows that the abundance of Alu RNA species in the RPE of a human eye with geographic atrophy (GA) is greater than in normal human eye RPE, and is principally approximately 300 nucleotides long, consistent with the length of a non-embedded Pol III derived transcript. Reprobing these samples with a probe corresponding to the “S region” of the 7SL RNA gene that is not present in Alu elements shows that 7SL RNA abundance is not different between the RPE of normal and GA human eyes. Abundance of U6 RNA in GA and normal eyes shows loading efficiency. c, Northern blot shows that Alu
probe detects in vitro transcribed Alu RNA but not 7SL RNA in mouse liver (which lacks primate-specific Alu), and reprobing these samples confirms specificity of the 7SL probe. d, DICER1 knockdown by antisense (as) oligonucleotides in human RPE cells does not, compared to control (Ctrl) as treatment, induce upregulation of several Pol II-transcribed genes (ADAR2, NINC, NLRP, SLFN 11) that contain embedded Alu sequences in their exons. n=3.

FIG. 21 7SL RNA is not regulated in geographic atrophy or by inhibition of DICER1 or Alu. a, 7SL RNA abundance was not different in the RPE of human eyes with geographic atrophy (GA) compared to the RPE of normal human eyes without GA (n=8). b, 7SL RNA abundance was not different in human RPE cells transfected with antisense oligonucleotide (as) targeting DICER1 from those transfected with control (Ctrl) as. n=3. c, 7SL RNA abundance was not different in human RPE cells transfected with antisense oligonucleotide (as) targeting Alu from those transfected with control (Ctrl) as. n=3. 7SL RNA abundance, relative to 18S RNA, was monitored by real-time RT-PCR. NS, not significant by Student t test.

FIG. 22 Overexpression of B1 or B2 RNA induces RPE degeneration. Subretinal transfection of pB1 or pB2 RNAs, but not of pNull, induces RPE degeneration in wild-type mice. Top row shows fundus photographs demonstrating areas of degeneration outlined by blue arrowheads. Bottom row shows ZO-1 stained (red) RPE flat mounts demonstrating marked degeneration and disarray of the RPE cells in mice overexpressing B1 or B2 RNAs. Circular flash artifact is seen in the centre of the fundus photographs. n=4. Representative images shown. Scale bar, 20 μm.

FIG. 23 Alu RNA enters retinal pigmented epithelium (RPE) cells in vivo. Subretinal administration of Alu RNA in wild-type mice achieved RPE cell delivery at 8 h after injection as monitored by real-time RT-PCR in isolated cell lysates (n=3).

FIG. 24 Human GA Alu dsRNA does not induce RPE degeneration when cleared by DICER1. a, Subretinal administration of a fully complementary synthetic Alu RNA (dsRNA) corresponding to the sequence of an Alu RNA isolated from a human eye with geographic atrophy (GA) induces RPE degeneration in wild-type mice. Vehicle administration does not damage the retina. Top panels show fundus photographs with the area of RPE degeneration outlined by blue arrowheads. Circular flash artifact is seen in the centre of the fundus photographs. Bottom panels show ZO-1 stained (red) RPE flat mounts that are well arrayed in vehicle (bottom) but disorganized in Alu dsRNA (top). b, This Alu dsRNA did not induce RPE degeneration when it was first subjected to cleavage by recombinant DICER1. However, when subjected to mock cleavage by DICER1, this Alu dsRNA did induce RPE degeneration. n=4. Representative images shown. Scale bar, 20 μm.

FIG. 25 RPE degeneration does not occur in response to a variety of structured RNAs. Subretinal transfection of transfer RNA (tRNA) or of plasmids coding for 7SL RNA, pri-miRNA-29b1 or pri-miRNA26a2 in wild-type mice did not induce retinal toxicity that was evident on fundus photography. Circular flash artifact is seen in the centre of the fundus photographs. N=4. Representative images shown.

FIG. 26 Alu RNA does not cause RPE degeneration via TLR3. a, Western blot shows that transfection of pAlu or pNull does not induce TLR3 phosphorylation, relative to the levels of the housekeeping protein Vinculin, in human RPE cells. b, Subretinal transfection of pAlu induced RPE degeneration in Tlr3−/− mice where pNull transfection did not do so. Representative images shown. n=4. Scale bar, 20 μm.

FIG. 27 DICER1 reduction or Alu RNA augmentation induces caspase-3 activation. a, Immunolocalization of activated caspase-3 (red) in the RPE of human eyes with geographic atrophy (left panel). Specificity of immunolabeling revealed by absence of staining with isotype control antibody (middle panel) and in control eyes stained with antibody against cleaved caspase-3 (right panel). Autofluorescence of RPE and choroid seen in green channel. Nuclei stained by DAPI (blue). b, Flat mounts of BEST1 Cre; Dicer1−/− mice show evidence of caspase-3 activation (red staining, top left panel). Specificity of immunolabeling revealed by absence of staining with isotype control antibody (top right panel). No caspase-3 activation was detectable in the RPE of littermate control BEST1 Cre or Dicer1+/+ mice (bottom panels). c, Human RPE cells transfected with Alu showed evidence of caspase-3 activation (red staining, top left panel). DAPI (blue staining) and merged images are also shown. Scale bars (20 μm, a; 10 μm, c). Representative images shown. n=4-6. d, Exposure of human RPE cells to Alu RNA induced dose-dependent increase in caspase-3 activation, as monitored by fluorometric plate assay. n=3, *P<0.05 compared to vehicle by Student t test. e, Transfection of human RPE cells with pAlu induced increase in caspase-3 activation. n=3, *P=0.47 by Student t test.

FIG. 28 Alu RNA cleavage fragments do not modulate RPE degeneration. a, Transfection of pAlu induced cell death in human RPE cells. Cotransfection of DICER1-cleaved Alu RNA fragments did not change the degree of cell death. n=3. b, Subretinal transfection of DICER1-cleaved Alu RNA fragments (Frag) in wild-type mice did not cause RPE degeneration as seen by fundus photography (top left) or ZO-1-stained (red) RPE flat mounts (bottom left). Cotransfections of these fragments did not prevent the RPE degeneration induced by pAlu in wild-type mice (right panels). n=4. Representative images shown. Scale bar, 20 μm.

FIG. 29 Impaired DICER1 processing of microRNAs does not increase Alu RNA abundance or modulate Alu RNA cytotoxicity. a, There was no significant difference (P>0.05) in Alu RNA transcript abundance between HCT116 parent cells and HCT mutant cells carrying a mutation in exon 5 (ex5) of DICER1 which renders it incapable of processing microRNAs. b, Transfection of anti-sense oligonucleotide (as) targeting DICER1 into HCT116 cells increased the abundance of Alu RNA transcripts compared to control anti-sense oligonucleotide (Ctrl as) at 48 h. Transcript abundance monitored by real-time RT-PCR and normalized to 18S rRNA levels. c, Alu RNA induced similar levels of cell death in HCT116 parent and HCT-DICER1ex5 cells. *P<0.05 by Student t test. n=4-6. d, Oxidative stress downregulates DICER1 in human RPE cells. Human retinal pigmented epithelium (RPE) cells exposed to varying concentrations of hydrogen peroxide (H2O2) display a dose- and time-dependent reduction in DICER1 mRNA abundance, as monitored by real-time RT-PCR and normalized to 18S rRNA levels. n=3.

**BRIEF DESCRIPTION OF THE SEQUENCE LISTING**

SEQ ID NO: 1 is an embodiment of a first strand of an siRNA provided in accordance with the presently-disclosed subject matter.
SEQ ID NO: 2 is an embodiment of a first strand of an siRNA provided in accordance with the presently-disclosed subject matter.

SEQ ID NO: 3 is an embodiment of a first strand of an siRNA provided in accordance with the presently-disclosed subject matter.

SEQ ID NO: 4 is an embodiment of a first strand of an siRNA provided in accordance with the presently-disclosed subject matter.

SEQ ID NO: 5 is an embodiment of a first strand of an siRNA provided in accordance with the presently-disclosed subject matter.

SEQ ID NO: 6 is an embodiment of a first strand of an siRNA provided in accordance with the presently-disclosed subject matter.

SEQ ID NO: 7 is a nucleotide sequence encoding a human DICER polypeptide, including all untranslated regions (GenBank Accession Number NM_177438).

SEQ ID NO: 8 is a cDNA sequence encoding a human DICER polypeptide.

SEQ ID NO: 9 is a polypeptide sequence for a human DICER polypeptide.

SEQ ID NO: 10 is a polypeptide sequence for a human DICER polypeptide, including residues 1276-1922 of SEQ ID NO: 9.

SEQ ID NO: 11 is a polypeptide sequence for a human DICER polypeptide, including residues 605-1922 of SEQ ID NO: 9.

SEQ ID NO: 12 is a polypeptide sequence for a human DICER polypeptide, including residues 1666-1922 of SEQ ID NO: 9.

SEQ ID NO: 13 is a polypeptide sequence for a human DICER polypeptide, including residues 1666-1912 of SEQ ID NO: 9.

SEQ ID NO: 14 is a polypeptide sequence for a human DICER polypeptide, including residues 1666-1786 and 1800-1912 of SEQ ID NO: 9.

SEQ ID NO: 15 is a polypeptide sequence for a human DICER polypeptide, including residues 1275-1824 of SEQ ID NO: 9.

SEQ ID NO: 16 is a polypeptide sequence for a human DICER polypeptide, including residues 1276-1824 of SEQ ID NO: 9.

SEQ ID NO: 17 is an mRNA sequence encoding a human DICER polypeptide.

SEQ ID NO: 18 is a mRNA sequence encoding a Schizosaccharomyces pombe DICER polypeptide.

SEQ ID NO: 19 is an mRNA sequence encoding a Schizosaccharomyces pombe DICER polypeptide.

SEQ ID NO: 20 is a polypeptide sequence for a Giardia lamblia DICER polypeptide.

SEQ ID NO: 21 is an mRNA sequence encoding a Giardia lamblia DICER polypeptide.

SEQ ID NO: 22 is an embodiment of an antisense oligonucleotide sequence provided in accordance with the presently-disclosed subject matter.

SEQ ID NO: 23 is an embodiment of an antisense oligonucleotide sequence provided in accordance with the presently-disclosed subject matter.

SEQ ID NO: 24 is an embodiment of an antisense oligonucleotide sequence provided in accordance with the presently-disclosed subject matter.

SEQ ID NO: 25 is an embodiment of an antisense oligonucleotide sequence provided in accordance with the presently-disclosed subject matter.

SEQ ID NO: 26 is an embodiment of an antisense oligonucleotide sequence provided in accordance with the presently-disclosed subject matter.

**DESCRIPTION OF EXEMPLARY EMBODIMENTS**

The presently-disclosed subject matter includes methods for identifying Alu RNA inhibitors and methods and compositions for inhibiting Alu RNA and therapeutic uses thereof.

As disclosed herein, Alu RNA (including Alu repeat RNA in human cells and B1 and B2, Alu-like element repeat RNAs) increases are associated with cells that are associated with certain conditions of interest. For example, Alu RNA increase is associated with the retinal pigment epithelium (RPE) cells of eyes with geographic atrophy. This increase of Alu RNA induces the death of RPE cells. Methods and compositions disclosed herein can protect a cell from Alu RNA-triggered cell death, thereby treating conditions associated with such cell death.

The presently-disclosed subject matter further includes methods useful for identifying an Alu RNA inhibitor and uses of such inhibitors, including therapeutic and protective uses. In some embodiments, the method makes use of a cultured cell wherein Alu RNA is upregulated. Candidate compounds can be screened using the cultured cell to determine efficacy as antagonists of Alu RNA. Candidate compounds include, for example, small molecules, biologics, and combinations thereof, such as compositions including multiple compounds. The term small molecules is inclusive of polypeptides and nucleotides.

In some embodiments, the screening method includes providing a cell in culture wherein Alu RNA is upregulated, and contacting a candidate compound with the cell. The method can further include identifying a change in Alu RNA. For example, a measurable change in Alu RNA levels can be indicative of efficacy associated with the candidate compound. In some embodiments, wherein the change in the Alu RNA is a measurable decrease in Alu RNA, the change is an indication that the candidate compound is an Alu RNA inhibitor. Such Alu RNA inhibitors can have utility for therapeutic applications as disclosed herein.

In some embodiments, the Alu RNA can be upregulated by decreasing native levels of DICER polypeptides in the cell using methods known to those skilled in the art. In some embodiments, the Alu RNA associated with cultured cell can be upregulated by using heat shock stress using methods known to those skilled in the art. In some embodiments, the cultured cell is an RPE cell.

Methods and compositions of the presently-disclosed subject matter for treating a condition of interest include inhibiting Alu RNA associated with a cell, such as a cell of a subject in need of treatment. Examples of conditions of interest include, but are not limited to: geographic atrophy, dry age-related macular degeneration, thalassemia, familial hypercholesterolemia, Dent’s disease, acute intermittent porphyria, anterior pituitary aplasia, Apert syndrome, Hemophilia A, Hemophilia B, glycogen kinase deficiency, autoimmune lymphoproliferative syndrome, X-linked agammaglobulinemia, X-linked severe combined immunodeficiency, adrenoleukodystrophy, Menkes disease, hyperimmunoglobulin M syndrome, retinal blinding, Type I anti-thrombin deficiency, Muckle-Wells syndrome, hypocalcemic hypercalcemia and hyperparathyroidism, cholesterol esterase deficiency, hereditary desmoid disease, chronic...

As used herein, the terms treatment or treating relate to any treatment of a condition of interest, including but not limited to prophylactic treatment and therapeutic treatment. As such, the terms treatment or treating include, but are not limited to: preventing a condition of interest or the development of a condition of interest; inhibiting the progression of a condition of interest; arresting or preventing the development of a condition of interest; reducing the severity of a condition of interest; ameliorating or relieving symptoms associated with a condition of interest; and causing a regression of the condition of interest or one or more of the symptoms associated with the condition of interest.

As used herein, the term “subject” refers to a target of treatment. The subject of the herein disclosed methods can be a vertebrate, such as a mammal, a fish, a bird, a reptile, or an amphibian. Thus, the subject of the herein disclosed methods can be a human or non-human. Thus, veterinary therapeutic uses are provided in accordance with the presently disclosed subject matter.

In some embodiments, the condition of interest is geographic atrophy and the cell is an RPE cell. In this regard, a subject having age-related macular degeneration can be achieved in a number of manners. For example, overexpressing the DICER polypeptide in the cell, for example, by overexpressing the DICER polypeptide in the cell. For another example, a DICER mRNA could be used. For another example, in some embodiments, inhibiting Alu RNA associated with a cell comprises administering a DICER polypeptide or a small RNA molecule targeting the Alu RNA. As used herein, inhibiting Alu RNA associated with a cell refers to a reduction in the levels of Alu RNA inside and/or outside the cell in the extracellular space.

The term DICER Polypeptide refers to polypeptides known to those of ordinary skill in the art as DICER, including, but not limited to polypeptides comprising the sequences of SEQ ID NO: 9, 18, and 20, and functional fragments or functional variants thereof.

It is noted that one of ordinary skill in the art will be able to readily obtain publicly-available information related to DICER, including relevant nucleotide and polypeptide sequences included in publicly-accessible databases, such as GENBANK®. Some of the sequences disclosed herein are cross-referenced to GENBANK® accession numbers, e.g., GenBank Accession Number NM_177438. The sequences cross-referenced in the GENBANK® database are expressly incorporated by reference as are equivalent and related sequences present in GENBANK® or other public databases. Also expressly incorporated herein by reference are all annotations present in the GENBANK® database associated with the sequences disclosed herein. Unless otherwise indicated or apparent, the references to the GENBANK® database are references to the most recent version of the database as of the filing date of this application.

The terms “polypeptide”, “protein”, and “peptide”, which are used interchangeably herein, refer to a polymer of the 20 protein amino acids, or amino acid analogs, regardless of its size. The terms “polypeptide fragment” or “fragment”, when used in reference to a reference polypeptide, refers to a polypeptide in which amino acid residues are deleted as compared to the reference polypeptide itself, but where the remaining amino acid sequence is usually identical to the corresponding positions in the reference polypeptide. Such deletions can occur at the amino-terminus (e.g., removing residues 1-604, 1-1274, 1-1275, or 1-1665 of SEQ ID NO: 9) or carboxy-terminus of the reference polypeptide (e.g., removing residues 1825-1922, or 1913-1922 of SEQ ID NO: 9), from internal portions of the reference polypeptide (e.g., removing residues 1787-1799 of SEQ ID NO: 9), or a combination thereof.

A fragment can also be a “functional fragment,” in which case the fragment retains some or all of the activity of the reference polypeptide as described herein. For example, in some embodiments, a functional fragment of the polypeptide of SEQ ID NO: 9 can retain some or all of the ability of the polypeptide of SEQ ID NO: 9 to degrade Alu RNA. Examples of functional fragments of the polypeptide of SEQ ID NO: 9 include the polypeptides of SEQ ID NOS: 10-16. Additional examples include, but are not limited to, the polypeptide of SEQ ID NO: 9, including the following residues: 605-1922, 605-1912, 1666-1922, 1666-1912, 605-1786 and 1800-1922, 605-1786 and 1800-1912, 1666-1786 and 1800-1912, 1276-1922, 1276-1912, 1276-1786 and 1800-1922, 1276-1786 and 1800-1912, 1275-1824, or 1276-1824.

The terms “modified amino acid”, “modified polypeptide”, and “variant” refer to an amino acid sequence that is different from the reference polypeptide by one or more amino acids, e.g., one or more amino acid substitutions. A variant of a reference polypeptide also refers to a variant of a fragment of the reference polypeptide, for example, a fragment wherein one or more amino acid substitutions have been made relative to the reference polypeptide. A variant can also be a “functional variant,” in which the variant retains some or all of the activity of the reference protein as described herein. The term functional variant includes a functional variant of a functional fragment of a reference polypeptide.

In some embodiments, the DICER Polypeptide can be overexpressed in the cell using a vector comprising a nucleotide encoding the DICER polypeptide, for example, the nucleotide of SEQ ID NOS: 7 or 8, or appropriate fragment thereof, or a nucleotide encoding a DICER Polypeptide, for example, a nucleotide encoding SEQ ID NOS: 9, 10, 11, 12, 13, 14, 15, 16, 18, or 20. As will be recognized by those skilled in the art, the vector can be a plasmid vector or a viral vector (e.g., adeno-associated virus, lentivirus, adenovirus).

As noted above, in some embodiments, inhibiting Alu RNA comprises use of a DICER mRNA. In some embodiments, a functional fragment of a DICER mRNA could be used. In some embodiments, a DICER mRNA having the sequence of SEQ ID NOS: 17, 19, or 21, or a functional fragment thereof could be used. In some embodiments an mRNA encoding a DICER Polypeptide could be used, for example, an mRNA encoding SEQ ID NOS: 9, 10, 11, 12, 13, 14, 15, 16, 18, or 20.

As noted above, in some embodiments, inhibiting Alu RNA comprises administering an oligonucleotide or a small RNA molecule targeting the Alu RNA. Such nucleotides can target and degrade Alu RNA.
Methods, devices, and materials similar or equivalent to those set forth in SEQ ID NOS: 1-6. The details of one or more embodiments of the presently-disclosed subject matter are set forth in this document. Modifications to embodiments described in this document, and other embodiments, will be evident to those of ordinary skill in the art after a study of the information provided in this document. The information provided in this document, and particularly the specific details of the described exemplary embodiments, is provided primarily for clearness of understanding and no unnecessary limitations are to be understood therefrom. In case of conflict, the specification of this document, including definitions, will control.

While the terms used herein are believed to be well understood by one of ordinary skill in the art, definitions are set forth to facilitate explanation of the presently-disclosed subject matter. Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which the presently-disclosed subject matter belongs. Although any methods, devices, and materials similar or equivalent to those described herein can be used in the practice or testing of the presently-disclosed subject matter, representative methods, devices, and materials are now described. Following long-standing patent law convention, the terms “a”, “an”, and “the” refer to “one or more” when used in this application, including the claims. Thus, for example, reference to “a cell” includes a plurality of such cells, and so forth.

Unless otherwise indicated, all numbers expressing quantities of ingredients, properties such as reaction conditions, and so forth used in the specification and claims are to be understood as being modified in all instances by the term “about”. Accordingly, unless indicated to the contrary, the numerical parameters set forth in this specification and claims are approximations that can vary depending upon the desired properties sought to be obtained by the presently-disclosed subject matter. As used herein, the term “about,” when referring to a value or to an amount of mass, weight, time, volume, concentration or percentage is meant to encompass variations of in some embodiments ±20%, in some embodiments ±10%, in some embodiments ±5%, in some embodiments ±1%, in some embodiments ±0.5%, and in some embodiments ±0.1% from the specified amount, as such variations are appropriate to perform the disclosed method.

As used herein, ranges can be expressed from “about one particular value, and/or to “about” another particular value. It is also understood that there are a number of values disclosed herein, and that each value is also herein disclosed as “about” that particular value in addition to the value itself. For example, if the value “10” is disclosed, then “about 10” is also disclosed. It is also understood that each unit between two particular units are also disclosed. For example, if 10 and 15 are disclosed, then 11, 12, 13, and 14 are also disclosed.

The presently-disclosed subject matter is further illustrated by the following specific but non-limiting examples.

**EXAMPLES**

DICER1 Reduction in Geographic Atrophy

In human donor eyes with geographic atrophy (n=10), the present inventors found using real-time RT-PCR that DICER1 mRNA abundance was reduced in the macular RPE by 65±3% (mean±SEM; P=0.0036; Mann-Whitney U test) compared to age-similar human eyes without geographic atrophy (n=11) (FIG. 1a). Because the best understood function of DICER1 is miRNA generation, the present inventors measured the expression of other enzymes involved in miRNA biogenesis. The abundance of the genes encoding DROSHA or the double stranded RNA (dsRNA) binding protein DGC8, which form a complex that processes pri-miRNAs into pre-miRNAs, was not reduced in the RPE of human eyes with geographic atrophy. There was also no reduction in the expression of the gene encoding Argonaute 2 (AGO2, encoded by EIF2C2), the core component of the miRNA effector complex, in the RPE of human eyes with geographic atrophy. Corroborating the mRNA data, the present inventors observed a marked reduction of DICER1 protein expression in the RPE layer of human eyes with geographic atrophy compared to controls in Western blot (FIG. 1b and FIG. 6) and immunohistochemistry analyses (FIG. 1c). Interestingly, DICER1 mRNA and protein abundance in the adjacent neural retina was similar between the two groups (FIG. 7).

Because DICER1 downregulation is observed in some cell types in culture conditions in response to various chemical stresses, the present inventors wondered whether DICER1 reduction in geographic atrophy might be a common downstream pathway in dying retina. DICER1 protein levels were not reduced in the RPE of human eyes with several other retinal disorders such as vitelliform macular dystrophy, retinitis pigmentosa, or retinal detachment (FIG. 8). Also, Dicer1 mRNA abundance in the RPE in two animal models of retinal degeneration—Ccl2+/-Cer2+/- (refs. 9,10) and Cp+/-Heph+/- mice—was unchanged compared to their background strains (FIG. 8). Gene expression studies in numerous other mouse models of retinal degeneration also have not reported modulation of Dicer1 (Supplemental Notes). These data argue that DICER1 depletion in the RPE of eyes with geographic atrophy is not a generic response of damaged or dying retinal cells in vivo.

DICER1 Depletion Induces RPE Degeneration

To determine the functional consequence of reduced DICER1 levels, the present inventors conditionally ablated Dicer1 in mouse RPE cells by interbreeding “foxed” Dicer1 mice12 (Dicer+/foxed) with BEST1 Cre mice13, which express Cre recombinase under the control of the RPE cell-specific BEST1 promoter. BEST1 Cre; Dicer1foxed mice uniformly exhibited dramatic RPE cell degeneration (FIG. 1d-f) that was evident by the time of weaning. None of the littermate controls exhibited similar pathology. The present inventors also deleted Dicer1 in adult mouse RPE by subretinal injection of an adenov-associated viral vector coding for Cre recombinase under the control of the BEST1 promoter4 (AAV1-BEST1-Cre) in Dicer1foxed mice (FIG. 9). These eyes uniformly displayed RPE cell degeneration at 28 days after injection similar to that observed in mice depleted of Dicer1 expression during development (FIG. 1g-i; FIG. 9).
contrast, neither the contralateral eyes of Dicer1/−/− mice that underwent subretinal injection of AAV1-BEST1-GFP nor the eyes of wild-type mice injected with subretinal AAV1-BEST1-Cre developed RPE cell degeneration (FIG. 1g-i and FIG. 9). The RPE cell morphogenesis in mice depleted of Dicer1 expression resembled that observed in the eyes of humans with RPE atrophy due to AMD (FIG. 10). When cultured RPE cells isolated from Dicer1/−/− mice were infected with an adenoviral vector coding for Cre recombinase (Ad-Cre), the present inventors observed a reduction of cell viability compared to infection with AdNull (FIG. 1j).

Similarly, antisense oligonucleotide mediated knockdown of DICER1 in human RPE cells resulted in increasing cell death over time (FIG. 1k). Collectively, these data support the hypothesis that DICER1 dysregulation is involved in the pathogenesis of geographic atrophy.

DICER1 Depletion Phenotype not Due to miRNA Dysregulation
The present inventors tested whether depletion of other enzymes involved in miRNA biogenesis also would induce RPE degeneration. Subretinal injection of AAV1-BEST1-Cre in Drosha/−/− (ref. 13), Dgcr8/−/− (refs. 15, 16), or 10 Ago2/−/− mice did not result in the dramatic RPE cell damage that was evident in similarly treated Dicer1/−/− mice (FIG. 11). These data suggest that miRNA imbalances are not responsible for RPE degeneration induced by Dicer1 depletion. However, the present inventors and others have reported that a small subset (approximately 7%) of mammalian miRNAs is generated by Dicer1 independent of Drosha and the hypothesis that DICER1 dysregulation is involved in the miRNA biogenesis in these HCT-DICER1 mice did not result in the dramatic RPE cell damage that was evident in similarly treated Dicer1/−/− mice (FIG. 11). Collectively, these data support the hypothesis that DICER1 dysregulation is involved in the pathogenesis of geographic atrophy.

The present inventors confirmed that the J2 monoclonal antibody recognized Alu RNA both in immunoblotting and in immunofluorescence assays (FIG. 15). The present inventors also detected a greater than 40-fold increase in the abundance of Alu RNAs in the RPE of human eyes with geographic atrophy compared to control eyes (n=7), but no significant difference in Alu RNA abundance was detected in the adjacent neural retina between the two groups (FIG. 2, FIG. 16). The present inventors did not identify exact matches to these Alu sequences in the reference human genome. This could be attributed to genetic variations or regions not represented in the reference genome or to chimeric Alu formation. Further studies are needed to elucidate the genomic origin of and regulatory factors involved in transcription of these Alu RNAs.

DICER1 Depletion Induces Alu RNA Accumulation
The present inventors tested whether Alu RNA accumulation in the RPE of geographic atrophy was the result of deficient DICER1 processing activity. DICER1 knockout in human RPE cells using antisense oligonucleotides resulted in increasing Alu RNA accumulation over time (FIG. 3a, FIG. 17). Similarly, Ad-Cre infection of RPE cells isolated from Dicer1/−/− mice resulted in accumulation of B1 and B2 repeat RNAs (FIG. 3b, c), which are Alu-like short interspersed repetitive elements in the mouse. Interestingly, DICER1 was expressed in both the nucleus and cytoplasm of RPE cells and its depletion led to accumulation of Alu/B1/B2 RNA in both cellular compartments (FIG. 3d, FIG. 18). In addition, recombinant DICER1 degraded Alu RNA, and the biological specificity of this cleavage was confirmed by the inability of heat-denatured DICER1 to degrade Alu RNA (FIG. 3e). AluRNA accumulation in human RPE cells reduced Alu RNA abundance following enforced expression of Alu RNA (FIG. 3f), consistent with degradation of these repetitive transcripts by DICER1 in vivo. Collectively these data confirm that DICER1 dysregulation can trigger Alu/B1/B2 RNA accumulation.

Because cell stresses such as heat shock or viral infection can induce generalized retrotransposon activation, the present inventors wondered whether Alu RNA accumulation in geographic atrophy might be a generic response in dying retina. However, in the RPE of human eyes with geographic atrophy and in Dicer1-depleted human RPE cells, there was no increase in the abundance of RNAs coded by L1.3 (a long interspersed repetitive element), human endogenous retrovirus-W envelope (a long terminal repeat retrotransposon), or hY3 (a repetitive small cytoplasmic Ro RNA)
These data demonstrate that Alu RNA accumulation is a biologically specific response to DICER1 depletion. To determine whether Alu RNA accumulation was derived from RNA polymerase II (Pol II) or Pol III transcription, the present inventors performed experiments using α-amanitin (a Pol II inhibitor) and tagetitoxin (a Pol III inhibitor). Alu RNA upregulation induced by DICER1 knockdown was inhibited by tagetitoxin but not α-amanitin (FIG. 20). The present inventors also found using Northern blotting that Alu RNA from the RPE of human eyes with geographic atrophy was approximately 300 nucleotides in length, consistent with the length of non-embedded Pol III Alu transcripts. Because homology between Alu RNA and 7SL RNA, the evolutionary precursor of Alu, can complicate interpretation of northern blot analysis, the present inventors reprobed these samples using a probe that specifically detects the non-Alu “S domain” of 7SL RNA. In contrast to the increased amounts of RNA species detected by the Alu-targeting probe in geographic atrophy RPE, there was no difference in 7SL RNA abundance. The present inventors also confirmed that the Alu probe did not detect endogenous 7SL RNA under the stringent conditions the present inventors employed. Corroborating these data, real-time RT-PCR analysis showed that 7SL RNA was not dysregulated in the RPE of human eyes with geographic atrophy or in DICER1-depleted human RPE cells (FIG. 21).

DICER1 knockdown also did not induce upregulation of several Pol II-transcribed genes (ADAR2, NICN, NLRP, SLFN 11) that contain embedded Alu sequences in their exons. Collectively, these data suggest that Alu RNA detected in the RPE of human eyes with geographic atrophy are primary Alu transcripts and not passenger or bystander sequences embedded in other RNAs. Conclusive assignment of these Alu sequences as Pol III transcripts must await precise determination of their transcription start site.

Alu RNA Induces RPE Degeneration

Next the present inventors tested whether accumulation of Alu RNA might promote the development of geographic atrophy. Transfecting human or wild-type mouse RPE cells with a plasmid coding for Alu (pAlu) reduced cell viability (FIG. 4a). Subretinal transfection of plasmids coding for two different Alu RNAs or for B1 or B2 RNAs induced RPE degeneration in wild-type mice (FIG. 4b, FIG. 22, and data not shown). Treatment of human RPE cells with a recombinant 281 nucleotide (nt)-long Alu RNA that is identical to a Pol III derived Alu RNA isolated from a human embryonal carcinoma cell line, i.e., a single RNA strand that folds into a defined secondary structure, resulted in a dose-dependent increase in cell death (FIG. 4c). These findings suggest that endogenous DICER1 can degrade small amounts of Alu RNA but are overwhelmed by high levels. Consistent with this concept, overexpression of DICER1 blocked Alu-induced cell death in human RPE cells (FIG. 4d) and RPE degeneration in wild-type mice (FIG. 4e).

The present inventors verified that subretinal injection of Alu RNA resulted in its delivery to RPE cells in wild-type mice (FIG. 23), consistent with the ability of long RNAs with duplex motifs to enter cells. The present inventors then cloned a 302-nt long Alu RNA isolated from the RPE of a human eye with geographic atrophy and transcribed it in vitro to generate partially and completely annealed structures that mimic Alu RNAs transcribed by Pol III and Pol II, respectively. Subretinal injection of either of these Alu RNAs resulted in RPE degeneration in wild-type mice (FIG. 4f, FIG. 24), supporting the assignment of disease causality in accord with the molecular Koch’s postulates. In contrast, subretinal injection of these Alu RNAs digested with Dicer1 did not induce RPE degeneration in wild-type mice (FIG. 4g, FIG. 24). When these Alu RNAs were subjected to mock Dicer1 digestion, they retained their ability to induce RPE degeneration, suggesting a role for Dicer1 in protecting against Alu RNA-induced degeneration.

The present inventors tested whether other structured RNAs of similar length as Alu would damage the retina. Subretinal transfection of transfer RNA or plasmids coding for 7SL RNA or two different primary miRNAs did not induce RPE degeneration in wild-type mice (FIG. 25). The present inventors reported that chemically synthesized dsRNAs that mimic viral dsRNA can induce RPE degeneration by activating toll like receptor-3 (TLR3)29, a pattern receptor that generically recognizes dsRNA. However, transfection of a plasmid coding for Alu RNA did not induce TLR3 phosphorylation in human RPE cells and did induce RPE degeneration in Tlr3−/− mice (FIG. 26). These results indicate that the ability of Alu RNA to induce RPE degeneration cannot be attributed solely to its repetitive or double stranded nature, as it exerted effects distinct from other structured dsRNAs of similar length.

The mechanism of RPE cell death in geographic atrophy has not been previously defined. DNA fragmentation has been identified in RPE cells in human eyes with geographic atrophy30, and Dicer1 knockdown has been associated with induction of apoptosis in diverse tissues. The present inventors now provide evidence of caspase-3 cleavage in regions of RPE degeneration in human eyes with geographic atrophy (FIG. 27). Caspase-3 cleavage was also observed in the RPE cells of BEST1Cre; Dicer1−/− mice and in Alu RNA-stimulated or -overexpressing human RPE cells. These data suggest a role for Alu RNA-induced RPE cell apoptosis triggered by Dicer1 dysregulation in geographic atrophy.

Although the present inventors show that Alu RNA induces RPE degeneration, the presented observations could be consistent with the idea that an imbalance in small RNA species produced from long Alu RNAs could contribute to the RPE degeneration phenotype. To study this question, the present inventors exposed human RPE cells or wild-type mice to DICER1 cleavage fragments of Alu RNA. Subretinal transfection of these fragments alone in wild-type mice had no detectable effect on RPE cell morphology, and co-administering these fragments did not prevent RPE cell degeneration induced by subretinal transfection of a plasmid coding for Alu RNA (FIG. 28). Similarly, these fragments did not prevent human RPE cell death induced by overexpression of Alu RNA. These data suggest that upregulation of long Alu RNA rather than imbalance in Alu RNA-derived small RNA fragments is responsible for RPE degeneration induced by Dicer1 reduction.

As these experiments were performed with in vitro cleavage fragments the present inventors cannot be certain whether in vivo cleavage fragments would function similarly. However, Alu RNAs with varying sequences induced RPE degeneration in vivo. Because the cleavage fragments of these different Alu RNAs would not be identical it is unlikely that they all execute identical biological functions, particularly if they functioned as miRNAs. Another line of evidence that Alu RNA, and not its cleavage fragments, is responsible for RPE degeneration comes from functional rescue experiments (see below) wherein antisense-mediated inhibition of Alu RNA blocks human RPE cell death induced by Dicer1 knockdown and inhibition of B1/B2 RNA blocks RPE degeneration in Dicer1-depleted mice and mouse RPE cells. Because these antisense treatments would not be expected to alter the reduced levels of Dicer1-
cleaved Alu/B1/B2 RNA fragments, the imbalance in these fragments is unlikely to have induced RPE degeneration. Nevertheless, subtle functions of these small RNAs in modulating Alu RNA induced pathology cannot be excluded.

To dissect the contribution of Alu RNA accumulation versus that of miRNA dysregulation to RPE degeneration in the context of reduced DICER1 expression, the present inventors re-examined HCT-DICER1ex5 cells in which miRNA biogenesis is impaired but long dsRNA cleavage is preserved due to the intact Rnase III domains. The present inventors found no significant difference in Alu RNA levels between HCT-DICER1ex5 and parent HCT116 cells (FIG. 29). In contrast, when DICER1 was knocked down by antisense oligonucleotides in HCT116 cells, increased Alu RNA accumulation was observed. Also, Alu RNA induces similar levels of cytotoxicity in HCT-DICER1ex5 and parent HCT116 cells, suggesting that coexisting miRNA expression deficits do not augment Alu RNA induced RPE degeneration. In conjunction with the discordance in the RPE degeneration phenotype between ablation of Dicer1 and that of various other small RNA biogenesis pathway genes in mice, the findings suggest that Alu RNA accumulation is critical to cytotoxicity induced by DICER1 reduction.

RPE Degeneration Blocked by Alu RNA Inhibition

The present inventors then tested whether the cytotoxic effects of DICER1 reduction could be attributed to Alu RNA accumulation. DICER1 knockdown in human RPE cells by antisense oligonucleotides reduced cell viability (FIG. 5a). This cytotoxic effect of DICER1 reduction was inhibited by antisense oligonucleotides targeting Alu RNA sequences but not by a scrambled antisense control (FIG. 5a, b and FIG. 21). Ad-Cre infection of RPE cells isolated from Dicer1-/- mice resulted in reduced cell viability, and this was blocked by antisense oligonucleotides targeting both B1 and B2 repeat RNAs but not by a scrambled antisense control (FIG. 5a, d). Subretinal administration of antisense oligonucleotides that reduced accumulation of B1 and B2 RNAs also inhibited RPE degeneration in Dicer1-/- mice treated with AAV1-BEST1-Cre (FIG. 5e, f), providing evidence of in vivo functional rescue.

The present inventors tested whether Alu inhibition also rescued miRNA expression deficits as a potential explanation for the functional rescue of RPE degeneration induced by DICER1 depletion. As expected, DICER1 knockdown in human RPE cells reduced the abundance of numerous miRNAs including let-7a, which is ubiquitously expressed, miR-184, miR-204/211, and miR-221/222, which are enriched in the RPE, and miR-320a, and miR-484 and miR-877, which are DROSHA/DGCR8-independent and DICER1-dependent (FIG. 5g). However, inhibition of Alu RNA did not lead to recovery of miRNA expression in these DICER1-depleted cells. Thus the rescue of RPE cell viability by Alu RNA inhibition despite the persistence of global miRNA expression deficits argues that RPE degeneration induced by DICER1 deficit is due to Alu RNA accumulation and not miRNA dysregulation.

These data, taken together, support a model in which primary Alu transcripts are responsible for the observed RPE degeneration. Whether similar pathology can also result from upregulation of as yet undefined Pol II transcripts with embedded Alu sequences is an intriguing possibility that may be addressed in future studies. Importantly, the present inventors show here that primary Alu transcripts are elevated in human disease, that Alu transcripts recapitulate disease in relevant experimental models, and that targeted suppression of Alu transcripts successfully inhibits this pathology. These observations have direct relevance for clinical strategies to prevent and treat geographic atrophy.

Discussion

The findings elucidate a critical cell survival function for DICER1 by functional silencing of toxic Alu transcripts. This unexpected function suggests that RNAi-independent mechanisms should be considered in interpreting the phenotypes of systems in which Dicer1 is dysregulated. For example, it would be interesting to test the speculation that Dicer1 ablation induced cell death in mouse neural retina52 and heart53 might also involve B1/B2 RNA accumulation. More broadly, recognition of DICER1’s hitherto unidentified function as an important controller of transcripts derived from the most abundant repetitive elements in the human and mouse genomes can illuminate new functions for RNases in cytoprotective surveillance. DICER1 expression is reduced in geographic atrophy and partial loss of DICER promotes RPE degeneration; thus the present inventors could speculate that loss of heterozygosity in DICER1 may underlie the development of geographic atrophy, similar to its function as a haploinsufficient tumor suppressor9-36.

This also is, to our knowledge, the first example of how Alu could cause a human disease via direct RNA cytotoxicity rather than by inducing chromosomal DNA rearrangements or insertional mutagenesis through retrotransposition, which have been implicated in diseases such as α-thalassemia57, colon cancer38, hypercholesterolemia39,40, and neurofibromatosis41. Future studies can be employed to determine the precise chromosomal locus of the Alu RNA elements that accumulate in geographic atrophy and the nature of transcriptional and post-transcriptional machinery that enable their biogenesis.

In addition to processing miRNAs3, DICER1 has been implicated in heterochromatin assembly42-45. Since Alu repeat elements are abundant within heterochromatin46, it would be interesting to investigate whether perturbations in centromeric silencing also underlie the pathogenesis of geographic atrophy. Indeed, the finding that chromatin remodelling at Alu repeats can regulate miRNA expression45 raises the intriguing possibility of other types of regulatory interactions between DICER1 and Alu. It also remains to be investigated whether centromeric satellite repeats that have been described to accumulate in Dicer1-null mouse embryonic stem cells46,47 might be involved in the pathogenesis of geographic atrophy.

In the mouse germline, Dicer1 has been implicated in the generation of endogenous small interfering RNAs (endo-siRNAs) from repeat elements48,49. If this process is conserved in mammalian somatic tissues, it would be interesting to learn whether endo-siRNAs serve a homeostatic function in preventing the development of geographic atrophy. A recent study in nematodes demonstrated that caspases can cleave Dicer1 and convert it into a DNase that promotes apoptosis50. The finding that Alu RNA can induce caspase activation therefore introduces the possibility of bidirectional regulation between DICER1 and Alu that could trigger feed-forward loops that further amplify the disease state.

The inciting events that trigger an RPE-specific reduction of DICER1 in patients with geographic atrophy remain to be determined. Potential culprit could include oxidative stress, which is postulated to underlie AMD pathogenesis2, as the present inventors found that exposure to hydrogen peroxide downregulates DICER1 in human RPE cells (FIG. 30). While the upstream triggers of DICER1 dysregulation and the possible role of other DICER-dependent, DROSHA/DGCR8-independent small RNAs in geographic atrophy await clarification, the ability of Alu RNA antisense oligo-
nucleotides to inhibit RPE cell death induced by DICER1 depletion provides a rationale to investigate Alu RNA inhibition or DICER1 augmentation as potential therapies for geographic atrophy.

Additional Notes
DICER1 mRNA levels are not modulated in multiple mouse models of retinal degeneration including light damage, hyperoxia, 5\(^\text{th}\) retinal detachment, Crx\(^{-/-}\) mice, Rslha \(^{-/-}\), mice, rdl mice, cpfl1 mice, or Mif1 mice. DICER1 abundance also is not reduced in mouse models of cellular stress in the retina including exposure to advanced glycation endproducts or retinal detachment. Therefore, DICER1 downregulation is not a generic late-stage response in the retina.

Materials and Methods

Animals
All animal experiments were approved by institutional review committees and the Association for Research in Vision and Ophthalmology. C57BL/6J and Dicer\(^{−/−}\) mice were purchased from The Jackson Laboratory. Transgenic mice that express Cre recombinase in the retinal pigment epithelium under the control of the human bestrophin-1 promoter (BEST1 Cre mice), DGCR8\(^{−/−}\), Drosha\(^{−/−}\), Tarbp2\(^{−/−}\), Ccl2\(^{−/−}\), Ccr2\(^{−/−}\), and Cp\(^{−/−}\) Hep\(^{−/−}\) mice have been previously described.

Isolation of dsRNA.
Isolation of dsRNA from the RPE of a human eye with geographic atrophy that underwent fundus photography and subretinal injections was performed using the Leica SP-5 or Zeiss Axio Observer Z1 microscope.

Histology.
Mouse eyes were fixed with 4\(^\%\) paraformaldehyde and 3.5\(^\%\) glutaraldehyde, postfixed in 2\(^\%\) osmium tetroxide, and dehydrated in ethanol and embedded. Semi-thin (1 \(\mu\)m) sections were cut and stained with toluidine blue. Bright field images were obtained using the Zeiss Axio Observer Z1 microscope.

Subretinal Injection.
Subretinal injections (1 \(\mu\)L) in mice were performed using a Pico-Injector (PLI-100, Harvard Apparatus). In vivo transfection of plasmids coding for DICER1 (ref. 74), Alu Ya5 (ref. 75), Alu Yb9 (ref. 76), 7SL RNA (ref. 77), pri-miR29b1 (Addgene), or pri-miR26a2 (Addgene) and bovine tRNA (Sigma-Aldrich) (1:5 \(\mu\)g/mL) was achieved using 10\(^\%\) Neuroporter (Genelantis). AAV1-BEST1-Cre or AAV1-BEST1-GFP were injected at 1\(\times\)10\(^{12}\) pfu/mL and recombinant Alu RNAs (1: a single RNA strand of 281 nucleotides whose sequence is that of the eDNA clone TS 103 (ref 51) and folds into a defined secondary structure identical to a Pol III derived transcript; 2: a single RNA strand of 302 nucleotides whose sequence is identical to that of a clone isolated from the RPE of a human eye with geographic atrophy that folds into a defined secondary structure identical to a Pol III derived transcript; or 3: a fully complementary dsRNA version of this 302 nucleotide long sequence that mimics a Pol II derived transcript) was injected at 0.3 mg/mL. Cell-permeant cholesterol conjugated-B1/B2 antisense oligonucleotides (as) (5\(^\prime\)-TCAGATCTCGTACGCAUGITGCA-3\(^\prime\) or choline conjugated-control as (5\(^\prime\)-TTGCTACGCACTGTTGATGGCA-3\(^\prime\)), both from Integrated DNA Technologies) were injected (2 \(\mu\)g in 1 \(\mu\)L) 10 days after AAV1-BEST1-Cre was injected in Dicer\(^{−/−}\) mice.

Isolation of dsRNA.
Human eyes were stored in RNAlater (Ambion). Tissue extracts were prepared by lysis in buffer containing 50 mM Tris-HCl, pH 8, 150 mM NaCl, 1% Nonidet P-40, protease and phosphatase inhibitors (complete mini EDTA-free, protease inhibitor and phosphatase inhibitor cocktail tablets, Roche), and RNase inhibitor (SUPERase-In, Ambion). After homogenization using bullet blender (Nextadvance) and centrifugation, immunoprecipitations were performed by adding 40 \(\mu\)g of mouse antibody against dsRNA (clone J2) for 16 h at 4\(^\circ\) C. Immunocomplexes were collected on protein A/G agarose (ThermoScientific) and dsRNA species.
were separated and isolated using Trizol (Invitrogen) according to the manufacturer’s instructions.

Ligation of dsRNA and Anchor Primer.

An anchor primer (PC3-T7 loop, 5'-p-CCGGATCCCTACAATGTTT-3') was ligated to dsRNA (200-400 ng) in 50 mM HEPES/NaOH, pH 8 (vWR), 18 mM MgCl2 (Invitrogen), 0.01% BSA (Fisher Scientific), 1 mM ATP (Roche), 3 mM DTT (Fluka), 10% DMSO (Finnzymes), 20% PEG 6000 (Alfa Aesar), and 30U T4 RNA ligase (Ambion). Ligation was performed at 37° C. for 16 h, and ligated dsRNA was purified by MinElute Gel extraction columns (Qiagen).

Sequence-Independent cDNA Synthesis.

After denaturation, ligated dsRNA was reverse transcribed in a RT reaction containing 50 mM Tris-HCl, pH 8.3, 10 mM MgCl2, 70 mM KCl, 30 mM β-mercaptoethanol, 1 mM dNTPs and 15U cloned AMV reverse transcriptase (Invitrogen). The mixture was incubated in a thermal cycler (Eppendorf) at 42° C. for 45 min followed by 55° C. for 15 min. Polymerase Chain Reaction (PCR) Amplification.

Amplification of cDNA was performed using primer PC2 (5'-p-CCGGATCCCTACAATGTTT-3') in a RT reaction buffer containing 5 μL cDNA and 40 μL Platinum PCR SuperMix (Invitrogen). The PCR cycling parameters consisted of one step of 72° C. for 1 min to fill incomplete cDNA ends and produce intact DNA, followed by one step of initial denaturation (94° C., 2 min), 39 cycles of 94° C. for 30 s, 53° C. for 30 s, and 72° C. for 1 min, and a final extension step of 72° C. for 10 min. In vitro transcribed dsRNAs of varying lengths (325 bp, 1 and 2 kb) were used as positive controls.

Cloning and Sequencing.

The amplified cDNA products were incubated with 1 μL calf intestinal alkaline phosphatase (Invitrogen) at 37° C. for 5 min to remove the 5'-phosphate group, separated on a low-melting point agarose gel (1%) and purified using Qiagen gel extraction kit (Qiagen). The purified dephosphorylated cDNA fragments were cloned into pCRII TOPO vector (Invitrogen) and sequenced using M13 forward (5'-p-TTACTACGACTTCGAGCTAC-3') and M13 reverse primers at the University of Kentucky Advanced Genetic Technologies Center using multi-colour fluorescent based DNA sequencer (ABI 3730xl). Sequences were assembled using ContigExpress from vector NTI Advance. The homology of the isolated cDNA sequences to known Alu consensus sequences was determined using the CENSOR server35 (a WU-BLAST-powered database of repetitive elements (http://www.girinst.org/censor)). For each cDNA sequence, the homologous region of the query was aligned to the consensus Alu sequence using ClustalW2 (http://www.ebi.ac.uk/Tools/clustalw2). Multiple sequence alignment was performed using ClustalW2 (http://www.ebi.ac.uk/Tools/clustalw2). The consensus sequences have been deposited in GenBank under the accession numbers H1N17658 and H1N176585.

Alu RNA Synthesis.

The present inventors synthesized two Alu RNAs: a 281 nt Alu sequence originating from the cDNA clone T8103 which is known to be expressed in human cells33 and a 302 nt Alu sequence isolated from the RPE of a human eye with geographic atrophy. Both of these Alu RNAs were synthesized using a RNA polymerase T7 promoter and runoff transcription followed by gel purification as previously described35. This yields single stranded RNAs that fold into a defined secondary structure identical to Pol III derived transcripts. The present inventors also synthesized a fully complementary dsRNA form (resembling a Pol II derived transcript) of the 302 nt human geographic atrophy Alu using linearized pCRII TOPO plasmid templates using 17 or SP6 RNA polymerases (MegaScript, Ambion) according to the manufacturer’s recommendations. After purification, equal molar amount of each transcript were combined and heated at 95°C for 1 min, cooled and then annealed at room temperature for 24 h. The Alu dsRNA was precipitated, suspended in water and analyzed on 1.4% non-denaturing agarose gel using the single-stranded complementary strands as controls.

Real-Time PCR.

Total RNA was extracted from tissues or cells using Trizol reagent (Invitrogen) according to manufacturer’s recommendations and were treated with RNase free DNase (Ambion). Total RNA (1 μg) was reverse transcribed as previously described35 using gScript cDNA SuperMix (Quanta Biosciences). The RT products (cDNA) were amplified by real-time quantitative PCR (Applied Biosystems 7900 HT Fast Real-Time PCR system) with Power SYBR green Master Mix. Oligonucleotide primers specific for Dicer1 (forward 5'-CCCGGCTGAGAGAACTTACG-3' and reverse 5'-CTGTAACCTCTGACAAACACTTCTAAA-3'), DROSHA (forward 5'-GAACAGTTCAACCCGAATGTTG-3' and reverse 5'-CTCAACTGTGAGGCCGGTATC-3'), DGCR8 (forward 5'-TCTGTCTCCITATGCCTGCTAGT-3' and reverse 5'-CCAAACTCCGGCCAAAGAG-3'), EIF2C2 (forward 5'-GCACGGAGATTCATGAGTTCG-3' and reverse 5'-CCGGCGTCTCTCGAGATCT-3'), human Alu rRNA (forward 5'-CCGAGTTGTTGATGCATTGAGG-3' and reverse 5'-GCCTCACTTCCGAAAACCAA-3'), Alu (forward 5'-CAACATAATGGAACCCCGTCTACT-3' and reverse 5'-GCCTCACTTCCGAAAACCAA-3'), LINE L1.3 (ORF2) (forward 5'-CGTAGTTTCTGAACTTCCC-3' and reverse 5'-TGTCGGCTCAATTCCTGAGGA-3'), HERV-W1 (forward 5'-GCCGCTGTATGACCAGTAGCT-3' and reverse 5'-GGGAGGCCGCTCTCCATCC-3'), human R-associated Y3 (bY3) (forward 5'-CCCGAGTGCTGTTGTTACA-3' and reverse 5'-GGAGTGGAAAAGAAGAAATC-3'), 7SL (forward 5'-CCGGCATCAATGTTGACCT-3' and reverse 5'-CTGATCAGCACCGGAGTTT-3'), B1 (forward 5'-TGCCATTITATCCAGACTT-3' and reverse 5'-GGCTGCCTACAAGGGTTGAA-3'), B2 (forward 5'-GAATGCATATTCGGGTTCT-3' and reverse 5'-GGTAAGTGGCTGAGGAATGAC-3'), Dicer1 (forward 5'-CCCAACCAGGGAATGTTGTT-3' and reverse 5'-TATGTGAGAGGCCGGTGTAAAA-3'), mouse 18S rRNA (forward 5'-TTGTTGTTGATGGCTGCCTGAG-3' and reverse 5'-CCCTTGAATTTCCATTCTCTG-3'), 5S and 18S rRNA of mouse were used as templates in the QPCR and verified by the absence of gel-detected bands. Relative expressions of target genes were determined by the 2-ΔΔCT method.

miRNA PCR.

miRNA abundance was quantified using the All-in-One™ miRNA qRT-PCR Detection Kit (GeneCopoeia). Briefly, total RNA was polyadenylated and reverse transcribed using
a poly d(T)-adapter primer. Quantitative RT-PCR was carried out using a miRNA-specific forward primer and universal reverse primer. PCR products were subjected to dissociation curve and gel electrophoresis analyses to ensure that single, mature miRNA products were amplified. Data were normalized to ACTB levels. The forward primers for the miRNAs were as follows: miR-184 (5’-TGGAGGGAGAAGCTGGGTTGAGAGGGCGA-3’); miR-221/222 (5’-AGCATCATTGCATCGTACTGGGTTGACAGGCCGA-3’); miR-204/211 (5’-TCTCCCTTGTGCTCACTACTTGCCCTCGC-3’); miR-877 (5’-GTAGAGGAGATGGCGACACGAG-3’); miR-320a (5’-AAAAGCTGTTCTGAGAGGGCGA-3’); miR-484 (5’-TCAGGTCAGTCCCTCCCTGCAAT-3’); let-7a (5’-TGAGGATTGTTGATGATGTGTT-3’). The reverse primers were proprietary (GeneCopoeia). The primers for ACTB were forward (5’-TGGATCAGCAAGCAGGAGACTB-3’) and reverse (5’-TCAGGTCAGTCCCTCCCTGCAAT-3’).

Dot Blot (Immuno-Dot Binding).

Increasing amounts of Alu RNA were spotted onto hybond-N+ positively charged nylon membrane (Amerham) and UV cross-linked. After blocking, the membranes were incubated with mouse antibody against dsRNA (1:1,000, clone J2) for 1 h at RT. The peroxidase-conjugated goat anti-mouse secondary antibody (1:5,000, Sigma) was used 25 min. Membranes were washed several times, the signals were visualized by enhanced chemiluminescence (ECL plus) and captured by VisionWorksLS Image Acquisition and Analysis software (Version 6.7.2, UVP, LLC). Densitometry analysis was performed using ImageJ (NIH).

The value of 1 was arbitrarily assigned for normal eye samples.

Dicer1 Cleavage.

The ability of Dicer1 to cleave Alu RNA was tested using Recombinant Human Dicer Enzyme Kit (Genlantis) according the manufacturer’s instructions. The products of the digestion were purified for the in vivo injection using RNA Purification Column (Genlantis).

Cell Culture.

All cell lines were cultured at 37°C and 5% CO₂. Primary mouse RPE cells were isolated as previously described and grown in Dulbecco Modified Eagle Medium (DMEM) supplemented with 10% FBS and standard antibiotics concentrations. Primary human RPE cells were isolated as previously described and maintained in DMEM supplemented with 20% FBS and antibiotics. Parental HCT116 and isogenic Dicer cells were cultured in McCoy’s 5A medium supplemented with 10% FBS.

Transient Transfection.

Human and mouse RPE cells were transfected with pUC19, pAlu, pCDNA3.1/Dicer1-FLAG, pCDNA3.1, Dicer1 antisense oligonucleotide (as) (5’-GCCGACGCATTGTTTCTGTCGGAAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGAGA...
Caspase-3 Activity.
Sub-confluent human RPE cells were treated with PBS or Alu RNA at different concentrations in 2% FBS medium for 8 h. The caspase-3 activity was measured using Caspase-3 Fluorometric Assay (R&D Systems) according to the manufacturer's instructions.

Confluent human RPE cells were exposed to hydrogen peroxide (0-2 mM, Fisher Scientific).

Statistics.
Results are expressed as mean±SEM, with P<0.05 considered statistically significant. Differences between groups were compared by using Mann-Whitney U test or Student t test, as appropriate, and 2-tailed P values are reported.

Throughout this document, various references are mentioned. All such references are incorporated herein by reference, including the references set forth in the following list:

REFERENCES


It will be understood that various details of the presently disclosed subject matter can be changed without departing from the scope of the subject matter disclosed herein. Furthermore, the foregoing description is for the purpose of illustration only, and not for the purpose of limitation.

---

**SEQUENCE LISTING**

<160> NUMBER OF SEQ ID NOS: 29

<210> SEQ ID NO 1
<211> LENGTH: 19
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: siRNA
<400> SEQUENCE: 1
cctagccctca cagtagct

<210> SEQ ID NO 2
<211> LENGTH: 19
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: siRNA
<400> SEQUENCE: 2
tgggacctaca gccgccgga

<210> SEQ ID NO 3
<211> LENGTH: 21
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: siRNA
<400> SEQUENCE: 3
gctcagcctc cagtagctgc t

<210> SEQ ID NO 4
<211> LENGTH: 21
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: siRNA
<400> SEQUENCE: 4

gcgggacgt caggccgcgc a 21

<210> SEQ ID NO 5
<211> LENGTH: 21
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: siRNA

<400> SEQUENCE: 5

gggactacag gcgccgaca c 21

<210> SEQ ID NO 6
<211> LENGTH: 21
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: siRNA

<400> SEQUENCE: 6

acaggccgcc gacacacatc c 21

<210> SEQ ID NO 7
<211> LENGTH: 10323
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 7
cggagggcccc gcgcgcgggc agttagaccc agcttggaccc gagccgcggc 60
gaggcgacgg cgacgcgggc gtcggcgcgc gcttggagcc ggggggccgg 120
ggcgggcgc gcgcggcggc ggcggggtct gccgacctgc gctgggaglc 180
cggcgcgc gcgcgcgcgc ccggcgcgac cggggtctgc gctgggaglc 240
gacaagcccc ccatctgccg ccatctgccg ccatctgccg ccatctgccg 300
tctcaccatct gcctgccctt ctggtggacc gaccaagcag atcagctacc 360
ttatacagcc agaataatacc atgatttacc cgggtgaccc cgggtgcacc 420
cgctctgctt ataataataat ccgggagcgc gcgcgctgct gcgcgctgct 480
gcgcgctgct gcgcgctgct gcgcgctgct gcgcgctgct gcgcgctgct 540
cgcgcgcgag ggcgcgcgcg gcgcgaggcc gcgcgaggcc gcgcgaggcc 600
tgggatacac ctagctcagcg ctagctcagcg ctagctcagcg ctagctcagcg 660
tttatctgct ctgctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga 720
tttatctgct ctgctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga 780
tgtctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga 840
tgtctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga 900
tgtctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga 960
tgtctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga 1020
tgtctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga 1080
cagtctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga 1140
cagtctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga gtcgctgtcga 1200
gcacttctca cctgctcacc ttgacctgaa atttgaact cttaaagtaa tccaatctgtc 1380
cgaatctta cgcacataa aaccatatga ggcacagcag tttgaaacgc tcagcttgta 1440
taataataga aatccaggtt attagtgtgc atggaggtat tctgagatatgagca 1500
tgaaagaatt gaaggaaag aagacgcgca gcaaaatatt cttatctcct ttacaaacat 1560
tttgtgagca attatctctc tgggaaaga agatcagcga ctgttgctaa cagagatgtg 1620
aaagagagtc ggcacagaca atccagagct gcgacagcag tttgaaagcg ttgagtggta 1680
acagggctt gggagactgc acgcctgcaac ccaacagagta aagacgaaat tccaaaca 1740
ggagaggtta ttgagccact ttcgagccca ctcgtatatttg ccacaaagtt 1800
tggaagagca ggtgtggtgata cccactatgc atctgtagatgatc 1860
agaaatacgg tgcctgtgcct tttcaacagtc aatccagaag gaccaagtctcta tagttagat 1920
aatgatgtagc gatccagaca aatataaagaa ttttgaagaa gacccatataa cttataaaac 1980
ataagagat gacggaactca aacagtagtgc caagggcttc tgggaaagcg ttgagtggta 2040
tgaatctggtg aatagatgtg atgtgtgtct cctactatgc actgtagatgagatc 2100
tggtctagca gtgcagcaac acacagccga tggacacatc atctgtagattgg 2160
acacagatgct ccagagctat tctacttctct aatgtgtagtt aagcttcatatc 2220
taatcactctt ctacttcatatc atctgtagatgatc 2280
tcccacacta agacagagca gatcactgtag ttttcctctct tgggaaagcg ggatgtgatg 2340
actgcacaa ccagagctca agcactgtgc cctgtaggct gcgacagcag tttgaaagcg 2400
aatgtgtagc gatccagaca aatataaagaa ttttgaagaa gacccatataa cttataaaac 2460
tttcagcagaa ggcagggcttt ggtccacgaac ccaacagagta aagacgaaat tccaaaca 2520
tcagcaggct tagcctagtt ccacaaagtgtg agttgtaaatc cacttcatct 2580
acacctgtaga ctctgcaggtg aacagagca gatcactgtag ttttcctctct tgggaaagcg 2640
ctgggataa ctaagcgccaa aaccataac ccacaaagtt cttacttctct tgggaaagcg 2700
cgcgctagct ctctgaggg gtaagagctg gatccagaca aatataaagaa ttttgaagaa 2760
aatgtgtagc gatccagaca aatataaagaa ttttgaagaa gacccatataa cttataaaac 2820
aacctgtcga ctactggtgc tgggaaagcg tgggaaagcg ggataggga 2880
aatgtgtagc gatccagaca aatataaagaa ttttgaagaa gacccatataa cttataaaac 2940
gagcttgcaag gctctttcct gcgacagcag tttgaaagcg ttgagtggta 3000
atatagatgg gcagagctgc ttcatactgt ccgacagcag tttgaaagcg ttgagtggta 3060
gttactgtag ttttatattgct tggaggtcat gaccaaggtct ggtacttctct tccatattg 3120
atccactgtag ctggtccact tttataactgt cttacttcatct tgggaaagcg ttgagtggta 3180
caactgtaac ccacacagct gcgacagcag tttgaaagcg ttgagtggta 3240
acgtcagcag ttcacactgc ggtcggtct ggcagaaaggc gttacttctct tccatattg 3300
agcacaagag gtagaggtgc tgggaaagcg ttgagtggta 3360
tcactctggtgc ttcatactgt ccgacagcag tttgaaagcg ttgagtggta 3420
tcactctggtgc ttcatactgt ccgacagcag tttgaaagcg ttgagtggta 3480
agcacaagag gtagaggtgc tgggaaagcg ttgagtggta 3540
tcactctggtgc ttcatactgt ccgacagcag tttgaaagcg ttgagtggta 3600
tcactctggtgc ttcatactgt ccgacagcag tttgaaagcg ttgagtggta 3660
<table>
<thead>
<tr>
<th>Location</th>
<th>DNA Sequence</th>
</tr>
</thead>
<tbody>
<tr>
<td>3720</td>
<td>ctctctagaa aatcatgacc aatgtcgtg gaactgcaaa acgttgctca g cgagtc cccc</td>
</tr>
<tr>
<td>3780</td>
<td>tggtaagtcgg tttcagcaca aat taaacctg gtaa ccatcttc t gctcaacaa</td>
</tr>
<tr>
<td>3840</td>
<td>tcacacagtc ctgataagta ccctgtggag tggatcctaa gctagctgaa aagatcctg</td>
</tr>
<tr>
<td>3900</td>
<td>ttatatcag tctcagcagc ccagagttaa agctactctc tcagtattca tggatataaa</td>
</tr>
<tr>
<td>3960</td>
<td>ataccttggt ggaatgctca aacatcttc ctacgagcag cagcagcagc cagcaggagc</td>
</tr>
<tr>
<td>4020</td>
<td>gctgtgagcc ccagagtctt gccagctctc gcctgctagc agctgtgtaa cccagcaggag</td>
</tr>
<tr>
<td>4080</td>
<td>tctctctggag gctactctgct gctgctcttg ctacgagcag cagcagcagc cagcaggagc</td>
</tr>
<tr>
<td>4140</td>
<td>ctctctggag gctactctgct gctgctcttg ctacgagcag cagcagcagc cagcaggagc</td>
</tr>
<tr>
<td>4200</td>
<td>gactctgca aacgtgagtc cctgaccttc acctgacttc cctgaccttc acctgacttc</td>
</tr>
<tr>
<td>4260</td>
<td>cctctctcca aatactctgga cctagctgga aatcttgctg gctctctgga cctagctgga</td>
</tr>
<tr>
<td>4320</td>
<td>aaagagagaga cctacgacgact agcatgccact gctgctactg gctgctactg gctgctactg</td>
</tr>
<tr>
<td>4380</td>
<td>tcctctctca gctgctactg gctgctactg gctgctactg gctgctactg gctgctactg</td>
</tr>
<tr>
<td>4440</td>
<td>aacgtgagtca aactgacttc cctgaccttc acctgacttc cctgaccttc acctgacttc</td>
</tr>
<tr>
<td>4500</td>
<td>tcactctcagc cctgctcttc acctgacttc cctgaccttc acctgacttc cctgaccttc</td>
</tr>
<tr>
<td>4560</td>
<td>tcagctcatt ctctgtcttc gctcagcttc cctgctcttc acctgacttc cctgaccttc</td>
</tr>
<tr>
<td>4620</td>
<td>tccctgtct cctgctcaga gctgacttc cctgaccttc acctgacttc cctgaccttc</td>
</tr>
<tr>
<td>4680</td>
<td>aacgtgagcct cctgctcaga gctgacttc cctgaccttc acctgacttc cctgaccttc</td>
</tr>
<tr>
<td>4740</td>
<td>tcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>4800</td>
<td>tggtaagtcgg tttcagcaca aat taaacctg gtaa ccatcttc t gctcaacaa</td>
</tr>
<tr>
<td>4860</td>
<td>ggaggggggg ggggggggg ggggggggg ggggggggg ggggggggg ggggggggg</td>
</tr>
<tr>
<td>4920</td>
<td>tcgcgttgga gctactctgct gctgctcttg ctacgagcag cagcagcagc cagcaggagc</td>
</tr>
<tr>
<td>4980</td>
<td>tccctctcagc cctgctcaga gctgacttc cctgaccttc acctgacttc cctgaccttc</td>
</tr>
<tr>
<td>5040</td>
<td>ggaggggggg ggggggggg ggggggggg ggggggggg ggggggggg ggggggggg</td>
</tr>
<tr>
<td>5100</td>
<td>tggtaagtcgg tttcagcaca aat taaacctg gtaa ccatcttc t gctcaacaa</td>
</tr>
<tr>
<td>5160</td>
<td>ggaggggggg ggggggggg ggggggggg ggggggggg ggggggggg ggggggggg</td>
</tr>
<tr>
<td>5220</td>
<td>tcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>5280</td>
<td>ggaggggggg ggggggggg ggggggggg ggggggggg ggggggggg ggggggggg</td>
</tr>
<tr>
<td>5340</td>
<td>tcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>5400</td>
<td>ggaggggggg ggggggggg ggggggggg ggggggggg ggggggggg ggggggggg</td>
</tr>
<tr>
<td>5460</td>
<td>tcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>5520</td>
<td>ggaggggggg ggggggggg ggggggggg ggggggggg ggggggggg ggggggggg</td>
</tr>
<tr>
<td>5580</td>
<td>gcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>5640</td>
<td>ggaggggggg ggggggggg ggggggggg ggggggggg ggggggggg ggggggggg</td>
</tr>
<tr>
<td>5700</td>
<td>gcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>5760</td>
<td>gcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>5820</td>
<td>gcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>5880</td>
<td>gcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>5940</td>
<td>gcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>6000</td>
<td>gcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
<tr>
<td>6060</td>
<td>gcctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc ctctctctcc</td>
</tr>
</tbody>
</table>
ttgactcaca gtttgagcga ttctgtgatc ccctggttac tgagttaaaa aataaaaaag 8460
tacagtttag acatattgaa ttgtttgatc tggcttgctt ttaaatgctgt 8520
aaagttttttc tctgttttgc tttgtgatat atggcaataa gataaaaaaa 8580
aatctactta tctgtggcaca ctttagagcc cttgtgtgcc cccctggttc ctgtgatgc 8640
aatgtagaac cgaatgtaga atggaaaccc taccaggtgg ggtgtgtgt gccttgagca 8700
cgtgtagaaa ggcgttggaga gcggctgctt gaaaaacgaa ctcgcaaaat tccatctgt 8760
gatgtgtagc aagttgatga acatgaacct tcattttcta aaaaatggttt 8820
taatattgtt ccagctgtgga taactatgtg gcgttttgatg ggaagtttta 8880
cgtctcttgt tgggtaatga agcagagaga aaaaaatgtt gttggcttgtt ggtttggaga 8940
aatgtagaaa agataatact aagatgtcgg cttgtagcct gtttttacctt atctcaagaa 9000
aatgtagaaa agataatact aagatgtcgg cttgtagcct gtttttacctt atctcaagaa 9060
gtcttctttt ttctttgttg gggatatgat ttaaatgctgt 9120
aatgtagaaa ggcagagaaa aaaaaatgtt gttggcttgtt ggtttggaga 9180
gatgtgtagc aagttgatga acatgaacct tcattttcta aaaaatggttt 9240
gatgtgtagc aagttgatga acatgaacct tcattttcta aaaaatggttt 9300
aacagagaaa tggggctgtt tgggtaatga agcagagaaa aaaaaatgtt gttggcttgtt ggtttggaga 9360
ttttggttggaa cggaaactctg aaagaactta gaatcagcat tttgagagca gaagcttggg catgctgtga 9420
tttggttggaa cggaaactctg aaagaactta gaatcagcat tttgagagca gaagcttggg catgctgtga 9480
ttttggtagtt ttctttggtt gggatatgat ttaaatgctgt 9540
gcttcttttt ttttttctttt ttcttttctttt ttcttttctttt 9600
aatgtagaaa agataatact aagatgtcgg cttgtagcct gtttttacctt atctcaagaa 9660
aatgtagaaa agataatact aagatgtcgg cttgtagcct gtttttacctt atctcaagaa 9720
ttttggttggaa cggaaactctg aaagaactta gaatcagcat tttgagagca gaagcttggg catgctgtga 9780
ttttggttggaa cggaaactctg aaagaactta gaatcagcat tttgagagca gaagcttggg catgctgtga 9840	agacagagaaa aacaaaaatg caatgaaaga aacactggat 9900
aatgtagaaa agataatact aagatgtcgg cttgtagcct gtttttacctt atctcaagaa 9960
aatgtagaaa agataatact aagatgtcgg cttgtagcct gtttttacctt atctcaagaa 10020
ttttggttggaa cggaaactctg aaagaactta gaatcagcat tttgagagca gaagcttggg catgctgtga 10080
ttttggttggaa cggaaactctg aaagaactta gaatcagcat tttgagagca gaagcttggg catgctgtga 10140
ttttggttggaa cggaaactctg aaagaactta gaatcagcat tttgagagca gaagcttggg catgctgtga 10200
ttttggttggaa cggaaactctg aaagaactta gaatcagcat tttgagagca gaagcttggg catgctgtga 10260
ttttggttggaa cggaaactctg aaagaactta gaatcagcat tttgagagca gaagcttggg catgctgtga 10320
aaa 10323
gaatgaaag cctggtttgc caacccctca gcatgagcagg cttgcagctc atgacccttg

cctccctacc aatgggctct ttctttggac tgccatggca acaagaagca attcatgata
acatttatac gcacaaaaaa tataatatgg aacctgcctgg gataataataa
ccatgctttg ttttaaactct gctcaggag aagacatata cctcactaagg
agctgcttc acaagcctgg tgtcagcttg cagaactcct ctaagcttc
aggttgggga atctctcaca ctgagaagtg atgcagctttg gcacaaaaag aagaggaacc
aagagtttac taagacccag gttoctattt gttcgccttg aatgttttga
aaatgttta cttatcaact tgcagacatta accttttggt gtttgatgag tgtcatcttg
caatcctaga caacccctat cgaagaatga tgaagctctg ttgtaaatata
tgtaaatata tagaaactata ctaatttctt cctttcttca
aacagactgct gtagcgctct ttagactttg gcagctcttg tgtggaagtc
aagaaaaatg tcaagactca gagaatatcg ttaagatagaa agctgcttc

tggtgtctt tcgacaggtct tcttcgtgag tgtgcttggt gaagtgggct
attcctgaga ctgccacagt ctggccctg tgcagacgtc
tgcagctgat gccagctctg ccactggcacc gcaggttctg
ttccttgacc aacttgctgg ttccttcgct ctgcgtctc
ctgcgtttct tggagctggag ttccttgctg ccacgctttg
tgcagctgct gttcagacct ggcttccttg ttcagccata
tcttcgggct gcagcttcat tcacatctag aacagctttg
tttgctgttg gttcttctcc cattgctttg gttcctgttct
tctctcattc gctcttccag tggagccagct tttccgcaag
tacctgcttt cccatctgct gtttctcttg aagagtggct
tcccaagaaaaa atgcagctttg gcagctcttg
tggtgacggt cctggtttgc caacccctca gcatgagcagg cttgcagctc atgacccttg

cctccctacc aatgggctct ttctttggac tgccatggca acaagaagca attcatgata
acatttatac gcacaaaaaa tataatatgg aacctgcctgg gataataataa
ccatgctttg ttttaaactct gctcaggag aagacatata cctcactaagg
agctgcttc acaagcctgg tgtcagcttg cagaactcct ctaagcttc
aggttgggga atctctcaca ctgagaagtg atgcagctttg gcacaaaaag aagaggaacc
aagagtttac taagacccag gttoctattt gttcgccttg aatgttttga
aaatgttta cttatcaact tgcagacatta accttttggt gtttgatgag tgtcatcttg
caatcctaga caacccctat cgaagaatga tgaagctctg ttgtaaatata
tgtaaatata tagaaactata ctaatttctt cctttcttca
aacagactgct gtagcgctct ttagactttg gcagctcttg tgtggaagtc
aagaaaaatg tcaagactca gagaatatcg ttaagatagaa agctgcttc

tggtgtctt tcgacaggtct tcttcgtgag tgtgcttggt gaagtgggct
attcctgaga ctgccacagt ctggccctg tgcagacgtc
tgcagctgat gccagctctg ccactggcacc gcaggttctg
ttccttgacc aacttgctgg ttccttcgct ctgcgtctc
tcttcgggct gcagcttcat tcacatctag aacagctttg
tttgctgttg gttcttctcc cattgctttg gttcctgttct
tctctcattc gctcttccag tggagccagct tttccgcaag
tacctgcttt cccatctgct gtttctcttg aagagtggct
tcccaagaaaaa atgcagctttg gcagctcttg
ctttacctga tgcaacctac tttagaaggg ggaagctcta tcctcctgaa gataccacaa
gatgctttgg aatactgacg gccaaaccca tacctcagat tccacacttt cctgtgtaca
ccgctctgg agaggttacc atatccattg agttgaagaa gtctggtttc atgttgtctc
tcaaaattgt tgaatgccat tccagcatg ttttatttttatttataa ctgtatatag
tttagcttc atacaagcat ctgtagactg tgcagctgaa tttaaaccag
tttctcccc tcgagtatgaa ctcttgcag aatattataa aacaaagtac aaccttgacc
taccaatct ctaaccagct tggatgctag tagagccacac atctcactaa tttaaatcag
ttttctccac atactgttctg tctcttcaag ctgactttttc tcttttttttcttttttca
tttttctcg ctctttcagc tggagtcgct ggtcttttcg aatattgtagc atctctctgaa
tattcctgt gtagcactgtg ctcggtggag tggagtgctg tggaggggag
atgaagcctg acattgagttg cgaaggtgct taaacagcag tctcttacctg gattttctaa
ttttacctga tttagaaggg ggaagctcta tcctcctgaa gataccacaa
agcttttccct gtgttcactg gggtcgaagg tgctcgcggt aatataaagg actgatcggg 4980
aaaagccctt gtgcctctcct cgggagaaatt tcaacagcca acaaaagaaac ctccagagg 5040
getgtgcctg gcgtttctcgg cacctgctgt acttgaagac tggatatag 5100
gttgtaagca cttcggccca agatgttcttt tggctacatt cagatcagat aacaactgtg 5160
atacacttat atggtggttg gaaatatttgg aaaaagaaat ccaactcaca ttcacagata 5220
agccctactcc ttctccaggct ttttacacatg cctcctacca ctacaatact atcactgatt 5280
gctacaggct ttagacattg ctcgagagtg ctgatttagc tccactatag aacagcacc 5340
atatgagact acctccgcag cacacccgag gggtcctcag gttcgccttg 5400
tcaacacacat ctcctttcga tctgtggtctg taaagtaagac ctacacaaag tacccttaag 5460
cctcgccttc ctagtccttgc atagaccttttg cacatcgcag tctggaagaag 5520
agtataagct agctcataaag gcacatgggg aataatatttgga gttgcttctt ggtgctttttt 5580
acaggtatat tggtagatcga cttgagacac cttggcgcgg ctgtacatcc atgacgctgc 5640
cactaatagc caaagttctct gcacagctccct cggctcggaga gggttcgacc 5700
tgggacacag aacatccag ttatccggcag ttagacagag aaggtctcag 5760
tcactctgga atgtagagga aaggggaaata ttaaagatttg ccagttgagaagaa 5820
tcccatctgctg ccacagcagc gagaagcctcc gaaagctcctg 5880
ccaatgccg aagcactttgg aagctaatttttg gaatcctgaaagat tcatctgcgtc 5940
caatattgtg aaacgggcct ttaaattcaca acaacacaac aaaaactaag 6000
aggggtttat ttagatcggg aagggagtta taaaattgcctg agaagaggtt 6060
ggcggcgcttt aacagttctcg gttctaaacc gatattagaatt gattacgacc 6120
gtataattt tgggagctaa aagattaatcgg tgaatctcgg 6180
tttccctcacc ttcgctgtaatt tttaaatcaca agagttctagtt atgattttggcctct 6240
gctcaataa attacagcag cttgttctttt tttttgtcag tttttgctg 6300
ttgtgagcag ccatcagctg ctaaccctcag gcacgtcctgt cagtaattgtg gttctttttc 6360
catctctcgtg tggcatcaggt ctgtaggttg tgaatctcgg atatagagac 6420
taaggtgtttc gagggagtag gcggcgctatt gccgctcagtt tattctcgtt 6480
aaggtgtatt tggcagtcag cttggtcctt tggagacttc gacagattttt taaggtgtttc 6540
tggtgtggcat ttggtgtaatt cacatatttgg tttcgttatt gcacacagctt ctcagaaaa 6600
aaggggttttt gctttaaag tgaacctccc atagatactc ttttaatcttg gatctttttg 6660
gaaacaactt ttttacattct ctttccatttt atatgcattt agacgttgagg atacgctgat 6720
actacactac cactcttgtt gttgtaacat tttacagggat cattacatatt tttttttgctc 6780
atgtacagtc aagatctgctt aaaaaaccag atcgatcagtc taaggtcttt tttttttttc 6840
aaaaactgtt tcggattctc aacccatttt actaaagttct cttcgcagct gagttctttcc 6900
tgattagacact tttatttggc aataattttg ctctctgagg tcggctctta aacaaatataa 6960
cctgacacct atcacacctc aataatgcctg cagatttttat aatggtggttg ttaccattttt 7020
aagaagcaca aacagcacctt ttatcataatt gttctctcac ataattttttt tttatatctt 7080
cttcataatgt tggctacatt ctttcactac caaaacgttg cttgtaattgc cggtaaagttt 7140
taacggttttgct gataacaggct ctaattattttg atacactcgg tattaggggctatatattag 7200
ataaacagct cttctattttt ctccttttga aagaggaaaaa aaaaaaact tctcttcgcca 7260
ttgcttaag ttctttcaat tagacctgta ggcactcttc acttaaatac ttcagttcctt
7320
cttttctttt gcattgaact ttcocctgtt tgggtctatg tttatgtatt atgcttgaaa
7380
rttaaatattttttttt cactgtaaat atataaccc tttataaaccc ttttttaaaaag
7440
cctgctggcta gttgcatcct ccctatgcc ggcttgctgc aatttggccc
7500
gtttaatttg cttgaagttt aagaaagctg aggagaggtg tttctatatt cccagcaccat
7560
gattctgacg ttgtgctcct gttgaaatgt gcattttatg gcattggaca ctttgatact
7620
gtcttctcttg cttatcctgc atctacccca cccagagaaa tgcctctgtg cgagtgcacc
7680
gacacaaaac tgcagctctt gtttcttaag gcacccctag tggagggggt attaagcttc
7740
tectgttatt ttttgtgtct atcaatctaa acttaaatg agatcttaat tataaaagca
7800
gtttttgagc aaattaggtg acttgtttta aaaaatattta attgcatttt ggaaccttag
7860
tagctctatt gattgtaatt tttattgttt ggtatgctct tttgttgttc ccaatcttaa
7920
tcctttctttttt gattttgacg gctttgttgc gcctttgatg atttgtgatt atgttttgttt
7980
gaggtgatg tttgatcaca gttttaatctg atatattataa aactctggtg atagtgtaaa
8040
ggcagctgtg ttggtagttt ccctadcttt ctccctgtct gatcggctct gcccttttca
8100
tgctagttt ttttgtgttt ccaatcttaa acttaaatg agatcttaat tataaaagca
8160
tgcagctatg atatactgct aaaaagcattg aagatcctgtt gtttctttaa ttttcttttttt
8220
ttcctttctttttt gcccttttctt gtttctttttt gcttttggagt aatattggtt ctcttttcttt
8280
tttctttttgg aacagtattg gacttttccc ggcttttttt tttcttttctt ttttcttttttt
8340
tgagtattc tcctctttct tttcttattt tcttttctttt ctttttctttt ctttttctttt
8400
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
8460
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
8520
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
8580
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
8640
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
8700
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
8760
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
8820
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
8880
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
8940
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9000
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9060
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9120
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9180
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9240
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9300
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9360
tttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9420
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9480
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9540
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9600
ntttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt ttttttttttt
9660
US 9,464,289 B2

-continued

attgtaagtg aaaaagttta aagatatgtt taagaccaag actatattaa tgaattttaa
9720
agttggtga gacgcaata gcaatatcta ggaatttgc atttgacca ttgttatatttcc
9780
cactagcgt gaaattggatt tttccaaact aacttttggaa tatatttttttt tcataacttc
9840
ttttttttttt gctcctatttt atttggagat caaccacaga caatttttaaat tttatagatg
9900
cactaagat tcactgcaag acgagttac atagcgaatttt tgcacaagtta acaggaagt
9960
aaattttgct cttttctgct gtaaatagtg aagaaasaatt actaaataatca agtaaaacta
10020
atcactatta ttctattgac aaataaatat ttccactcacc atcgctgcaag ttttatttttta
10080
ggaacatgat gtcattcatt catacagtaa tcatgctgca gaaatttttgca gtctgcaacct
10140
tatggatcac aattacccatg ttgtttttttt ttgtaataaattgtagcgaattttttt atcttc
10200
ataaaagttta ttggtcgtgcc
10220
<210> SEQ ID NO 9
<211> LENGTH: 1922
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<400> SEQUENCE: 9

Met Lys Ser Pro Ala Leu Gln Pro Leu Ser Met Ala Gly Leu Gly Leu Met Thr Pro Ala Ser Ser Pro Met Gly Pro Phe Phe Gly Leu Pro Trp 20 25 30
Gln Gln Glu Ala Ile His Asp Asn Ile Tyr Thr Pro Arg Arg Tyr Gln Val Glu Leu Glu Ala Leu Asp His Asn Thr Ile Val Cys Leu 35 40 45
Val Glu Leu Leu Glu Ala Leu Asp His Asn Thr Ile Val Cys Leu 50 55 60
Asn Thr Gly Ser Gly Lys Thr Phe Ile Ala Val Leu Leu Thr Lys Glu 65 70 75 80
Leu Ser Tyr Gln Ile Arg Gly Asp Phe Ser Arg Asn Gly Lys Arg Thr 85 90 95
Val Phe Leu Val Asn Ser Ala Asn Val Ala Gln Gln Val Ser Val Arg Thr His Ser Asp Leu Lys Val Gly Glu Tyr Ser Asn Leu Glu 100 105 110
Val Phe Leu Val Asn Ser Ala Asn Val Ala Glu Val Ser Ser Val Ala Ser Phe Leu Arg Lys Glu Arg Phe Thr Lys His Gln Val Leu Ile Met Thr Cys Tyr Val Ala Leu Asn Val Leu Lys 115 120 125
His Gln Val Leu Ile Met Thr Cys Tyr Val Ala Leu Asn Val Leu Lys 130 135 140
Asn Gly Tyr Leu Ser Leu Ser Asp Ile Asn Leu Leu Val Phe Asp Glu 145 150 155 160
Asn Gly Tyr Leu Ser Leu Ser Asp Ile Asn Leu Leu Val Phe Asp Glu 165 170 175
Cys His Leu Ala Ile Leu Asp His Pro Tyr Arg Glu Ile Met Lys Leu 180 185 190
Cys Glu Asn Cys Pro Ser Cys Pro Arg Ile Leu Gly Leu Thr Ala Ser 195 200 205
Ile Leu Asn Gly Lys Cys Asp Pro Glu Glu Leu Glu Gly Leu Gly Leu Lys Ile Gln 210 215 220
Lys Leu Glu Lys Ile Leu Lys Ser Asn Ala Glu Thr Ala Thr Aep Leu 225 230 235 240
Val Val Leu Asp Arg Tyr Thr Ser Gln Pro Cys Glu Ile Val Val Asp 245 250 255
Cys Gly Pro Phe Thr Aep Arg Ser Gly Leu Tyr Glu Arg Leu Leu Met 260 265 270
<table>
<thead>
<tr>
<th>Residue</th>
<th>Sequence</th>
</tr>
</thead>
<tbody>
<tr>
<td>275</td>
<td>Glu Leu Glu Glu Ala Leu Asn Phe Ile Asn Asp Cys Asn Ile Ser Val</td>
</tr>
<tr>
<td>280</td>
<td></td>
</tr>
<tr>
<td>285</td>
<td></td>
</tr>
<tr>
<td>290</td>
<td>His Ser Lys Glu Arg Asp Ser Thr Leu Ile Ser Lys Gln Ile Leu Ser</td>
</tr>
<tr>
<td>295</td>
<td></td>
</tr>
<tr>
<td>300</td>
<td>Asp Cys Arg Ala Val Leu Val Leu Gly Pro Trp Cys Ala Asp Lys</td>
</tr>
<tr>
<td>305</td>
<td></td>
</tr>
<tr>
<td>310</td>
<td>Val Ala Gly Met Met Val Arg Leu Gln Lys Tyr Ile Lys His Glu</td>
</tr>
<tr>
<td>315</td>
<td></td>
</tr>
<tr>
<td>320</td>
<td>Gln Glu Glu Leu His Arg Lys Phe Leu Leu Phe Thr Asp Thr Phe Leu</td>
</tr>
<tr>
<td>325</td>
<td></td>
</tr>
<tr>
<td>330</td>
<td>Arg Lys Ile His Ala Leu Cys Glu Glu His Phe Ser Pro Ala Ser Leu</td>
</tr>
<tr>
<td>335</td>
<td></td>
</tr>
<tr>
<td>340</td>
<td>Asp Leu Lys Phe Val Thr Pro Lys Val Ile Lys Leu Leu Glu Ile Leu</td>
</tr>
<tr>
<td>345</td>
<td></td>
</tr>
<tr>
<td>350</td>
<td>Arg Lys Tyr Lys Pro Tyr Glu Arg Gln Gln Phe Glu Ser Val Glu Trp</td>
</tr>
<tr>
<td>355</td>
<td></td>
</tr>
<tr>
<td>360</td>
<td>Tyr Asn Asn Arg Asn Gln Asp Tyr Val Ser Trp Ser Asp Ser Glu</td>
</tr>
<tr>
<td>365</td>
<td></td>
</tr>
<tr>
<td>370</td>
<td>Asp Asp Asp Glu Asp Glu Glu Ile Glu Glu Lys Glu Lys Pro Glu Thr</td>
</tr>
<tr>
<td>375</td>
<td></td>
</tr>
<tr>
<td>380</td>
<td>Asn Phe Pro Ser Pro Phe Thr Asn Ile Leu Cys Gly Ile Ile Phe Val</td>
</tr>
<tr>
<td>385</td>
<td></td>
</tr>
<tr>
<td>390</td>
<td>Glu Arg Arg Tyr Thr Ala Val Val Leu Asn Arg Leu Ile Lys Glu Ala</td>
</tr>
<tr>
<td>395</td>
<td></td>
</tr>
<tr>
<td>400</td>
<td>Gly Lys Gln Asp Pro Glu Leu Ala Tyr Ile Ser Ser Asn Phe Ile Thr</td>
</tr>
<tr>
<td>405</td>
<td></td>
</tr>
<tr>
<td>410</td>
<td>Gly His Gly Ile Gly Lys Asn Gln Pro Arg Asn Lys Gln Met Glu Ala</td>
</tr>
<tr>
<td>415</td>
<td></td>
</tr>
<tr>
<td>420</td>
<td>Glu Phe Arg Lys Gln Glu Glu Val Leu Arg Lys Phe Arg Ala His Glu</td>
</tr>
<tr>
<td>425</td>
<td></td>
</tr>
<tr>
<td>430</td>
<td>Thr Asn Leu Leu Ile Ala Thr Ser Ile Val Glu Glu Gly Val Asp Ile</td>
</tr>
<tr>
<td>435</td>
<td></td>
</tr>
<tr>
<td>440</td>
<td>Pro Lys Cys Asn Leu Val Val Arg Phe Asp Leu Pro Thr Glu Tyr Arg</td>
</tr>
<tr>
<td>445</td>
<td></td>
</tr>
<tr>
<td>450</td>
<td>Ser Tyr Val Gln Ser Lys Gly Arg Ala Arg Ala Pro Ile Ser Asn Tyr</td>
</tr>
<tr>
<td>455</td>
<td></td>
</tr>
<tr>
<td>460</td>
<td>Ile Met Leu Ala Asp Thr Asp Lys Ile Lys Ser Phe Glu Glu Asp Leu</td>
</tr>
<tr>
<td>465</td>
<td></td>
</tr>
<tr>
<td>470</td>
<td>Lys Thr Tyr Lys Ala Ile Glu Lys Ile Leu Arg Asn Lys Cys Ser Lys</td>
</tr>
<tr>
<td>475</td>
<td></td>
</tr>
<tr>
<td>480</td>
<td>Ser Val Asp Thr Gly Glu Thr Asp Ile Asp Pro Val Met Asp Asp Asp</td>
</tr>
<tr>
<td>485</td>
<td></td>
</tr>
<tr>
<td>490</td>
<td>Asp Val Phe Pro Pro Tyr Val Leu Arg Pro Asp Gly Gly Gly Pro Arg</td>
</tr>
<tr>
<td>495</td>
<td></td>
</tr>
<tr>
<td>500</td>
<td>Val Thr Ile Asn Thr Ala Ile Gly His Ile Asn Arg Tyr Cys Ala Arg</td>
</tr>
<tr>
<td>505</td>
<td></td>
</tr>
<tr>
<td>510</td>
<td>Leu Pro Ser Asp Pro Phe Thr His Leu Ala Pro Lys Cys Arg Thr Arg</td>
</tr>
<tr>
<td>515</td>
<td></td>
</tr>
<tr>
<td>520</td>
<td>Glu Leu Pro Asp Gly Thr Phe Tyr Ser Thr Leu Tyr Leu Pro Ile Asn</td>
</tr>
<tr>
<td>525</td>
<td></td>
</tr>
<tr>
<td>530</td>
<td>Ser Pro Leu Arg Ala Ser Ile Val Gly Pro Met Ser Cys Val Arg</td>
</tr>
<tr>
<td>535</td>
<td></td>
</tr>
<tr>
<td>540</td>
<td></td>
</tr>
</tbody>
</table>
Leu Ala Glu Arg Val Val Ala Leu Ile Cys Cys Glu Lys Leu His Lys 690 628 700
Ile Gly Glu Leu Asp Asp His Leu Met Pro Val Gly Lys Glu Thr Val 705 710 715 620 720
Lys Tyr Glu Glu Leu Asp Leu His Asp Glu Glu Glu Thr Ser Val 725 730 735
Pro Gly Arg Pro Gly Ser Thr Lys Arg Arg Glu Cys Tyr Pro Lys Ala 740 745 750
Ile Pro Glu Cys Leu Arg Asp Ser Tyr Pro Arg Pro Asp Glu Pro Cys 755 760 765
Tyr Leu Tyr Val Ile Gly Met Val Leu Thr Thr Pro Leu Pro Asp Glu 770 775 780
Leu Asn Phe Arg Arg Arg Lys Leu Tyr Pro Glu Asp Thr Thr Arg 785 790 795 800
Cys Phe Gly Ile Leu Thr Ala Lys Pro Ile Pro Glu Ile Pro His Phe 805 810 815
Pro Val Tyr Thr Arg Ser Gly Glu Val Thr Ile Ser Ile Glu Leu Lys 820 825 830
Lys Ser Gly Phe Met Leu Ser Leu Gln Met Leu Glu Leu Ile Thr Arg 835 840 845
Leu His Glu Tyr Ile Phe Ser His Ile Leu Arg Leu Glu Lys Pro Ala 850 855 860
Leu Glu Phe Lys Pro Thr Asp Ala Asp Ser Ala Tyr Cys Val Leu Pro 865 870 875 880
Leu Asn Val Val Asn Asp Ser Thr Leu Asp Ile Asp Phe Lys Phe 885 890 895
Met Glu Asp Ile Glu Lys Ser Glu Ala Arg Ile Gly Ile Pro Ser Thr 900 905 910
Lys Tyr Thr Lys Glu Thr Pro Phe Val Phe Lys Leu Glu Asp Tyr Gln 915 920 925
Asp Ala Val Ile Ile Pro Arg Tyr Arg Asn Phe Asp Glu Pro His Arg 930 935 940
Phe Tyr Val Ala Asp Val Tyr Thr Asp Leu Thr Pro Leu Ser Lys Phe 945 950 955 960
Pro Ser Pro Glu Tyr Glu Thr Phe Ala Glu Tyr Tyr Lys Thr Tyr 965 970 975
Asn Leu Asp Leu Thr Asn Leu Asn Gln Pro Leu Leu Asp Val Asp His 980 985 990
Thr Ser Ser Arg Leu Asn Leu Leu Thr Pro Arg His Leu Asn Gln Lys 995 1000 1005
Gly Lys Ala Leu Pro Leu Ser Ser Ala Glu Lys Arg Lys Ala Lys 1010 1015 1020
Trp Glu Ser Leu Gln Asn Lys Gln Ile Leu Val Pro Glu Leu Cys 1025 1030 1035
Ala Ile His Pro Ile Pro Ala Ser Leu Thr Arg Lys Ala Val Cys 1040 1045 1050
Leu Pro Ser Ile Leu Tyr Arg Leu His Cys Leu Leu Thr Ala Glu 1055 1060 1065
Glu Leu Arg Ala Gln Thr Ala Ser Asp Ala Gly Val Gly Val Arg 1070 1075 1080
Ser Leu Pro Ala Asp Phe Arg Tyr Pro Asn Leu Asp Phe Gly Thr 1085 1090 1095
Lys Lys Ser Ile Asp Ser Lys Ser Phe Ile Ser Ile Ser Asn Ser
Ser Ser Ala Glu Asn Asp Asn Tyr Cys His Ser Thr Ile Val
Glu Asn Ala Ala His Gln Gly Ala Asn Arg Thr Ser Ser Leu
Glu Asp Glu Met Ser Val Asn Cys Arg Thr Leu Leu Ser
Glu Ser Pro Gly Lys Leu His Val Glu Val Ser Ala Asp Leu Thr
Ala Ile Asn Gly Leu Ser Tyr Asn Gln Asn Leu Ala Asn Gly Ser
Tyr Asp Leu Ala Asn Arg Asp Phe Cys Gln Gly Asn Gln Leu Asn
Tyr Tyr Lys Gln Glu Ile Pro Val Gln Pro Thr Thr Ser Tyr Ser
Ile Gln Asn Leu Tyr Ser Tyr Glu Asn Gln Pro Gln Pro Ser Asp
Glu Cys Thr Leu Leu Ser Asn Lys Tyr Leu Asp Gly Asn Ala Asn
Lys Ser Thr Ser Asp Gly Ser Pro Val Met Ala Val Met Pro Gly
Thr Thr Asp Thr Ile Gln Val Leu Lys Gly Arg Met Asp Ser Glu
Gln Ser Pro Ser Ile Gly Tyr Ser Ser Arg Thr Leu Gly Pro Asn
Pro Gly Leu Ile Leu Gln Ala Leu Thr Leu Ser Asn Ala Ser Asp
Gly Phe Asn Leu Glu Arg Leu Glu Met Leu Gly Asp Ser Phe Leu
Lys His Ala Ile Thr Thr Tyr Leu Phe Cys Thr Tyr Pro Asp Ala
His Glu Gly Arg Leu Ser Tyr Met Arg Ser Lys Lys Val Ser Asn
Cys Asn Leu Tyr Arg Leu Gln Lys Lys Gln Leu Pro Ser Arg
Met Val Val Ser Ile Phe Asp Pro Pro Val Asp Trp Leu Pro Pro
Gly Tyr Val Val Asn Glu Asp Lys Ser Asn Thr Asp Lys Trp Glu
Lys Asp Glu Met Thr Lys Asp Cys Met Leu Ala Asn Gly Lys Leu
Asp Glu Asp Tyr Glu Glu Asp Glu Glu Glu Ser Leu Met
Trp Arg Ala Pro Lys Glu Ala Asp Tyr Glu Asp Phe Leu
Glu Tyr Asp Gln Glu His Ile Arg Phe Ile Asp Asn Met Leu Met
Gly Ser Gly Ala Phe Val Lys Lys Ile Ser Leu Ser Pro Phe Ser
Thr Thr Asp Ser Ala Tyr Glu Trp Lys Met Pro Lys Lys Ser Ser
Leu Gly Ser Met Pro Phe Ser Ser Asp Phe Glu Asp Phe Asp Tyr
Ser Ser Trp Asp Ala Met Cys Tyr Leu Asp Pro Ser Lys Ala Val
1505 1510 1515
Glu Glu Asp Asp Phe Val Val Gly Phe Trp Asn Pro Ser Glu Glu
1520 1525 1530
Asn Cys Gly Val Asp Thr Gly Lys Gln Ser Ile Ser Tyr Asp Leu
1535 1540 1545
His Thr Glu Gln Cys Ile Ala Asp Lys Ser Ile Ala Asp Cys Val
1550 1555 1560
Glu Ala Leu Leu Gly Cys Tyr Leu Thr Ser Cys Gln Glu Arg Ala
1565 1570 1575
 Ala Gln Leu Phe Leu Cys Ser Leu Gly Leu Lys Val Leu Pro Val
1580 1585 1590
Ile Lys Arg Thr Asp Arg Glu Lys Ala Leu Cys Pro Thr Arg Glu
1595 1600 1605
Asn Phe Asn Ser Glu Gln Lys Asn Leu Ser Val Ser Cys Ala Ala
1610 1615 1620
 Ala Ser Val Ala Ser Arg Ser Ser Val Leu Lys Asp Ser Glu
1625 1630 1635
Tyr Gly Cys Leu Lys Ile Pro Pro Arg Cys Met Phe Asp His Pro
1640 1645 1650
Asp Ala Asp Lys Thr Leu Asn His Leu Ile Ser Gln Phe Glu Asn
1655 1660 1665
Phe Glu Lys Lys Ile Asn Tyr Arg Phe Lys Asn Lys Ala Tyr Leu
1670 1675 1680
Leu Gln Ala Phe Thr His Ala Ser Tyr His Tyr Asn Thr Ile Thr
1685 1690 1695
Asp Cys Tyr Gln Arg Leu Glu Phe Leu Gly Asp Ala Ile Leu Asp
1700 1705 1710
Tyr Leu Ile Thr Lys His Leu Tyr Glu Asp Pro Arg Glu Asn Ser
1715 1720 1725
Pro Gly Val Leu Thr Asp Leu Arg Ser Ala Leu Val Asn Asn Thr
1730 1735 1740
Ile Phe Ala Ser Leu Ala Val Lys Tyr Asp Tyr His Lys Tyr Phe
1745 1750 1755
Lys Ala Val Ser Pro Glu Leu Phe His Val Ile Asp Asp Phe Val
1760 1765 1770
Gln Phe Gln Leu Glu Lys Asn Glu Met Gln Gly Met Asp Ser Glu
1775 1780 1785
Leu Arg Arg Ser Glu Glu Glu Glu Lys Glu Glu Asp Ile Glu
1790 1795 1800
Val Pro Lys Ala Met Gly Asp Ile Phe Glu Ser Leu Ala Gly Ala
1805 1810 1815
Ile Tyr Met Asp Ser Gly Met Ser Leu Glu Thr Val Trp Gln Val
1820 1825 1830
Tyr Tyr Pro Met Met Arg Pro Leu Ile Glu Lys Phe Ser Ala Asn
1835 1840 1845
Val Pro Arg Ser Pro Val Arg Glu Leu Leu Glu Met Glu Pro Glu
1850 1855 1860
Thr Ala Lys Phe Ser Pro Ala Glu Arg Thr Tyr Asp Gly Lys Val
1865 1870 1875
Arg Val Thr Val Glu Val Val Gly Lys Gly Lys Phe Lys Gly Val
1880 1885 1890
<table>
<thead>
<tr>
<th>Gly Arg Ser Tyr Arg Ile Ala Lys Ser Ala Ala Ala Arg Arg Ala</th>
<th>Gly Arg Ser Tyr Arg Ile Ala Lys Ser Ala Ala Ala Arg Arg Ala</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leu Arg Ser Leu Lys Ala Asn Gln Pro Gln Val Pro Asn Ser</td>
<td>Leu Arg Ser Leu Lys Ala Asn Gln Pro Gln Val Pro Asn Ser</td>
</tr>
<tr>
<td>1895</td>
<td>1895</td>
</tr>
<tr>
<td>1900</td>
<td>1900</td>
</tr>
<tr>
<td>1905</td>
<td>1905</td>
</tr>
<tr>
<td>1910</td>
<td>1910</td>
</tr>
<tr>
<td>1915</td>
<td>1915</td>
</tr>
<tr>
<td>1920</td>
<td>1920</td>
</tr>
</tbody>
</table>

-continued

| <210> SEQ ID NO 10<br/> <211> LENGTH: 647<br/> <212> TYPE: PRT<br/> <213> ORGANISM: Homo sapiens<br/> <400> SEQUENCE: 10<br/> Asp Ser Glu Gln Ser Pro Ser Ile Gly Tyr Ser Ser Arg Thr Leu Gly<br/> Pro Asn Pro Gly Leu Ile Leu Gln Ala Leu Thr Leu Ser Asn Ala Ser<br/> 1 5 10 15
| Pro Asn Pro Gly Leu Ile Leu Gln Ala Leu Thr Leu Ser Asn Ala Ser |

| 20 25 30 |
| Lys His Ala Ile Thr Thr Tyr Leu Phe Cys Thr Tyr Pro Asp Ala His<br/> Glu Gly Arg Leu Ser Tyr Met Arg Ser Lys Lys Val Ser Asn Cys Asn<br/> Leu Tyr Arg Leu Gly Lys Lys Lys Leu Pro Ser Arg Met Val Val<br/> Ser Ile Phe Asp Pro Pro Val Asn Trp Leu Pro Pro Gly Tyr Val Val<br/> Asn Gln Asp Lys Ser Asn Thr Asp Tyr Trp Glu Lys Asp Glu Met Thr<br/> Lys Asp Cys Met Leu Ala Asn Gln Leu Asp Glu Tyr Glu Glu<br/> Glu Asp Glu Glu Glu Ser Leu Met Trp Arg Ala Pro Lys Glu Glu<br/> Ala Asp Tyr Glu Asp Asp Ala Leu Phe Gln Glu His Ile Arg<br/> Phe Ile Asp Asn Met Leu Met Gly Ser Gly Ala Phe Val Lys Lys Ile<br/> Ser Leu Ser Pro Phe Ser Thr Asp Ser Ala Tyr Glu Trp Lys Met<br/> Pro Lys Lys Ser Ser Leu Gly Ser Met Pro Phe Ser Ser Asp Phe Glu<br/> Asp Phe Asp Tyr Ser Ser Trp Asp Ala Met Cys Tyr Leu Asp Pro Ser<br/> Lys Ala Val Glu Asp Phe Val Val Gln Phe Pro Met Ala Val<br/> Glu Glu Asp Cys Gly Val Asp Thr Gly Lys Gln Ser Ile Ser Tyr Asp<br/> Leu His Thr Glu Gln Cys Ile Ala Asp Lys Ser Ile Ala Asp Cys Val<br/> Glu Ala Leu Leu Gly Cys Tyr Leu Thr Ser Cys Gly Glu Arg Ala Ala<br/> Gin Leu Phe Leu Cys Ser Leu Gly Leu Lys Val Leu Pro Val Ile Lys<br/> Arg Thr Asp Arg Glu Lys Ala Leu Cys Pro Thr Arg Glu Asn Phe Asn<br/> Ser Gin Gin Lys Gin Leu Ser Val Ser Cys Ala Ala Ala Ser Val Ala<br/> 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |

| 63 64 |
Ser Ser Arg Ser Ser Val Leu Lys Asp Ser Glu Tyr Gly Cys Leu Lys 355 360 365
Ile Pro Pro Arg Cys Met Phe Asp His Pro Asp Ala Asp Lys Thr Leu 370 375 380
Asn His Leu Ile Ser Gly Phe Glu Asn Phe Glu Lys Ile Asn Tyr 385 390 395 400
Arg Phe Lys Asn Lys Ala Tyr Leu Gln Ala Phe Thr His Ala Ser 405 410 415
Tyr His Tyr Asn Thr Ile Thr Asp Cys Tyr Glu Arg Leu Glu Phe Leu 420 425 430
Gly Asp Ala Ile Leu Asp Tyr Leu Ile Thr Lys His Leu Tyr Glu Asp 435 440 445
Pro Arg Gln His Ser Pro Gly Val Leu Thr Asp Leu Arg Ser Ala Leu 450 455 460
Val Asn Thr Ile Phe Ala Ser Leu Ala Val Lys Tyr Asp Tyr His 465 470 475 480
Lys Tyr Phe Lys Ala Val Ser Pro Glu Leu Phe His Val Ile Asp Asp 485 490 495
Phe Val Gln Phe Glu Leu Glu Met Gln Gly Met Asp Ser 500 505 510
Glu Leu Arg Arg Ser Glu Glu Glu Lys Glu Glu Asp Ile Glu 515 520 525
Val Pro Lys Ala Met Gly Asp Ile Phe Glu Ser Leu Ala Gly Ala Ile 530 535 540
Tyr Met Asp Ser Gly Met Ser Leu Glu Thr Val Trp Gln Val Tyr Tyr 545 550 555 560
Pro Met Met Arg Pro Leu Ile Glu Phe Ser Ala Asn Val Pro Arg 565 570 575
Ser Pro Val Arg Glu Leu Leu Glu Met Glu Pro Glu Thr Ala Lys Phe 580 585 590
Ser Pro Ala Glu Arg Thr Tyr Asp Gly Val Arg Val Arg Val Glu 595 600 605
Val Val Gly Lys Gly Lys Gly Phe Gly Val Gly Arg Ser Tyr Arg Ile 610 615 620
Asp Lys Ser Ala Ala Ala Arg Ala Leu Arg Ser Leu Lys Ala Arg 625 630 635 640
Gln Pro Gln Val Pro Asn Ser 645

<210> SEQ ID NO: 11
<211> LENGTH: 1318
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<400> SEQUENCE: 11
Met Asp Asp Asp Arg Val Phe Pro Pro Tyr Val Leu Arg Pro Asp Asp 1 5 10 15
Gly Gly Pro Arg Val Thr Ile Asn Thr Ala Ile Gly His Ile Asn Arg 20 25 30
Tyr Cys Ala Arg Leu Pro Ser Asp Pro Phe Thr His Leu Ala Pro Lys 35 40 45
Cys Arg Thr Arg Glu Leu Pro Asp Gly Thr Phe Tyr Ser Thr Leu Tyr 50 55 60
Leu Pro Ile Asn Ser Pro Leu Arg Ala Ser Ile Val Gly Pro Pro Met 66
<p>| | | | | |</p>
<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>66</td>
<td>67</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ser Cys Val Arg Leu Ala Glu Arg Val Val Ala Leu Ile Cys Cys Glu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>85</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lys Leu His Lys Ile Gly Glu Leu Asp Asp His Leu Met Pro Val Gly</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>100</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lys Glu Thr Val Lys Tyr Glu Glu Leu Asp Leu His Asp Glu Glu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>115</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glu Thr Ser Val Pro Gly Arg Pro Gly Ser Thr Lys Arg Arg Glu Cys</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>130</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tyr Pro Lys Ala Ile Pro Glu Cys Leu Arg Asp Ser Tyr Pro Arg Pro</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>145</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Pro Asp Glu Leu Asn Phe Arg Arg Asp Lys Leu Tyr Pro Pro Glu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>160</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Asp Thr Thr Arg Cys Phe Gly Ile Leu Thr Ala Lys Pro Ile Pro Glu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>175</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ile Pro His Phe Pro Val Tyr Thr Arg Ser Gly Glu Val Thr Ile Ser</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>190</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ile Glu Leu Lys Lys Ser Gly Phe Met Leu Ser Leu Glu Gln Met Leu Glu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>205</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Ile Thr Arg Leu His Glu Tyr Ile Phe Ser His Ile Leu Arg Leu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>220</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glu Lys Pro Ala Leu Glu Phe Lys Pro Thr Asp Ala Asp Ser Ala Tyr</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>235</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cys Val Leu Pro Leu Asn Val Val Asn Asp Ser Thr Leu Asp Ile</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>250</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Asp Phe Lys Phe Met Glu Asp Ile Glu Lys Ser Glu Ala Arg Ile Gly</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>265</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ile Pro Ser Thr Lys Tyr Thr Lys Glu Thr Pro Phe Val Phe Lys Leu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>280</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glu Asp Tyr Gln Asp Ala Val Ile Ile Pro Arg Tyr Arg Asn Phe Asp</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>295</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gln Pro His Arg Phe Tyr Val Ala Asp Val Tyr Thr Asp Leu Thr Pro</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>310</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Ser Lys Phe Pro Ser Pro Glu Tyr Glu Thr Phe Ala Glu Tyr Ty r</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>325</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lys Thr Lys Tyr Asn Leu Asp Leu Thr Asn Leu Asp Glu Pro Leu Leu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>340</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Asp Val Asp His Thr Ser Ser Arg Leu Asn Leu Leu Thr Pro Arg His</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>355</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Asn Gln Lys Gly Lys Ala Leu Pro Leu Ser Ser Ala Glu Lys Arg</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>370</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lys Ala Lys Trp Glu Ser Leu Gln Asn Lys Gln Ile Leu Val Pro Glu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>385</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Cys Ala Ile His Pro Ile Pro Ala Ser Leu Trp Arg Lys Ala Val</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>400</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cys Leu Pro Ser Ile Leu Tyr Arg Leu His Cys Leu Thr Ala Glu</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>415</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glu Leu Arg Ala Gln Thr Ala Ser Asp Ala Gly Val Val Glu Val Arg</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>430</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Pro Ala Asp Phe Arg Tyr Pro Asn Leu Asp Phe Gly Trp Lys Lys</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>445</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Pro Ala Asp Phe Arg Tyr Pro Asn Leu Asp Phe Gly Trp Lys Lys</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>460</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Pro Ala Asp Phe Arg Tyr Pro Asn Leu Asp Phe Gly Trp Lys Lys</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>475</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Pro Ala Asp Phe Arg Tyr Pro Asn Leu Asp Phe Gly Trp Lys Lys</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>490</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Leu Pro Ala Asp Phe Arg Tyr Pro Asn Leu Asp Phe Gly Trp Lys Lys</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>495</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Ser Ile Asp Ser Lys Ser Phe Ile Ser Ile Ser Asn Ser Ser Ser Ser Ala
  500 505 510
Glu Asn Asp Tyr Cys Lys His Ser Thr Ile Val Pro Glu Asn Ala
  515 520 525
Ala His Glu Gly Ala Asn Arg Thr Ser Ser Leu Glu Asn His Asp Gln
  530 535 540
Met Ser Val Asn Cys Arg Thr Leu Ser Glu Ser Pro Gly Lys Leu
  545 550 555 560
His Val Glu Val Ser Ala Asp Leu Thr Ala Ile Asn Gly Leu Ser Tyr
  565 570 575
Asn Gln Asn Leu Ala Asn Gly Ser Tyr Asp Leu Ala Asn Arg Asp Phe
  580 585 590
Cys Gln Gly Asn Phe Asp Tyr Lys Gln Glu Ile Pro Val Gln
  595 600 605
Pro Thr Thr Ser Tyr Ser Ile Gln Asn Leu Tyr Ser Tyr Glu Asn Gln
  610 615 620
Pro Gln Pro Ser Asp Gly Cys Thr Leu Leu Ser Asn Lys Tyr Leu Asp
  625 630 635 640
Gly Asn Ala Asn Lys Ser Thr Ser Asp Gly Ser Pro Val Met Ala Val
  645 650 655
Met Pro Gly Thr Thr Asp Thr Ile Gln Val Leu Lys Gly Arg Met Asp
  660 665 670
Ser Glu Gln Ser Pro Ser Ile Gly Tyr Ser Ser Arg Thr Leu Gly Pro
  675 680 685
Asn Pro Gly Leu Ile Leu Gln Ala Leu Thr Leu Ser Asn Ala Ser Asp
  690 695 700
Gly Phe Asn Leu Glu Arg Leu Glu Met Leu Gly Asp Ser Phe Leu Lys
  705 710 715 720
His Ala Ile Thr Thr Tyr Leu Phe Cys Thr Tyr Pro Asp Ala His Gln
  725 730 735
Gly Arg Leu Ser Tyr Met Arg Ser Lys Lys Val Ser Asn Cys Asn Leu
  740 745 750
Tyr Arg Leu Gly Lys Lys Gly Leu Pro Ser Arg Met Val Val Ser
  755 760 765
Ile Phe Asp Pro Pro Val Arg Trp Leu Pro Pro Gly Tyr Val Val Asn
  770 775 780
Gln Asp Lys Ser Asn Thr Asp Lys Trp Glu Lys Asp Glu Met Thr Lys
  785 790 795 800
Asp Cys Met Leu Ala Asn Gly Leu Asp Glu Asp Tyr Glu Glu Glu
  805 810 815
Asp Glu Glu Glu Ser Leu Met Trp Arg Ala Pro Lys Glu Glu Ala
  820 825 830
Asp Tyr Glu Asp Phe Leu Tyr Asp Glu His Ile Arg Phe
  835 840 845
Ile Asp Asn Met Leu Met Gly Ser Gly Ala Phe Val Lys Ile Ser
  850 855 860
Leu Ser Pro Phe Ser Thr Asp Ser Ala Tyr Glu Trp Lys Met Pro
  865 870 875 880
Lys Lys Ser Ser Leu Gly Ser Met Pro Phe Ser Ser Asp Phe Glu Asp
  885 890 895
Phe Asp Tyr Ser Ser Trp Asp Ala Met Cys Tyr Leu Asp Pro Ser Lys
  900 905 910
<table>
<thead>
<tr>
<th>Sequence</th>
<th>Position</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ala Val Glu Glu Asp Asp Phe Val Val Gly Phe Trp Aen Pro Ser Glu</td>
<td>910</td>
</tr>
<tr>
<td>Glu Asn Cys Gly Val Asp Thr Gly Lys Gln Ser Ile Ser Tyr Asp Leu</td>
<td>915</td>
</tr>
<tr>
<td>His Thr Glu Gln Cys Ile Ala Asp Lys Ser Ile Ala Asp Cys Val Glu</td>
<td>920</td>
</tr>
<tr>
<td>Ala Leu Leu Gly Cys Tyr Leu Thr Ser Cys Gly Glu Arg Ala Ala Gln</td>
<td>925</td>
</tr>
<tr>
<td>Leu Phe Leu Cys Ser Leu Gly Leu Lys Val Leu Pro Val Ile Lys Arg</td>
<td>930</td>
</tr>
<tr>
<td>Thr Asp Arg Glu Lys Ala Leu Cys Pro Thr Arg Glu Asn Phe Asn Ser</td>
<td>935</td>
</tr>
<tr>
<td>Glu Gln Lys Asn Leu Ser Val Ser Cys Ala Ala Ala Ser Val Ala</td>
<td>940</td>
</tr>
<tr>
<td>Ser Ser Arg Ser Ser Val Leu Lys Asp Ser Glu Tyr Gly Cys Leu</td>
<td>945</td>
</tr>
<tr>
<td>Lys Ile Pro Pro Arg Cys Met Phe Asp His Pro Asp Ala Asp Lys</td>
<td>950</td>
</tr>
<tr>
<td>Thr Leu Aen His Leu Ile Ser Gly Phe Glu Asn Phe Glu Lys Lys</td>
<td>955</td>
</tr>
<tr>
<td>Ile Asn Tyr Arg Phe Lys Asn Lys Ala Tyr Leu Leu Gln Ala Phe</td>
<td>960</td>
</tr>
<tr>
<td>Thr His Ala Ser Tyr His Tyr Aen Thr Ile Thr Asp Cys Tyr Gln</td>
<td>965</td>
</tr>
<tr>
<td>Arg Leu Glu Phe Leu Gly Asp Ala Ile Leu Asp Tyr Leu Ile Thr</td>
<td>970</td>
</tr>
<tr>
<td>Lys His Leu Tyr Glu Asp Pro Arg Gln His Ser Pro Gly Val Leu</td>
<td>975</td>
</tr>
<tr>
<td>Thr Asp Leu Arg Ser Ala Leu Val Asn Amn Thr Ile Phe Ala Ser</td>
<td>980</td>
</tr>
<tr>
<td>Leu Ala Val Lys Tyr Asp Tyr His Lys Tyr Phe Lys Ala Val Ser</td>
<td>985</td>
</tr>
<tr>
<td>Pro Glu Leu Phe His Val Ile Asp Asp Phe Val Gln Phe Gln Leu</td>
<td>990</td>
</tr>
<tr>
<td>Glu Lys Aen Glu Met Gln Gly Met Asp Ser Glu Leu Arg Arg Ser</td>
<td>995</td>
</tr>
<tr>
<td>Glu Glu Asp Glu Glu Lys Glu Glu Asp Ile Glu Val Pro Lys Ala</td>
<td>1000</td>
</tr>
<tr>
<td>Met Gly Asp Ile Phe Glu Ser Leu Ala Gly Ala Ile Tyr Met Asp</td>
<td>1005</td>
</tr>
<tr>
<td>Ser Gly Met Ser Leu Glu Thr Val Trp Gln Val Tyr Tyr Pro Met</td>
<td>1010</td>
</tr>
<tr>
<td>Met Arg Pro Leu Ile Glu Lys Phe Ser Ala Asn Val Pro Arg Ser</td>
<td>1015</td>
</tr>
<tr>
<td>Pro Val Arg Glu Leu Leu Glu Met Glu Pro Glu Thr Ala Lys Phe</td>
<td>1020</td>
</tr>
<tr>
<td>Ser Pro Ala Glu Arg Thr Tyr Asp Gly Lys Val Arg Val Thr Val</td>
<td>1025</td>
</tr>
<tr>
<td>Glu Val Val Gly Lys Gly Lys Phe Lys Gly Val Gln Gly Arg Ser Tyr</td>
<td>1030</td>
</tr>
<tr>
<td>Arg Ile Ala Lys Ser Ala Ala Ala Arg Arg Ala Leu Arg Ser Leu</td>
<td>1035</td>
</tr>
<tr>
<td>Lys Ala Asn Glu Pro Gln Val Pro Asn Ser</td>
<td>1040</td>
</tr>
</tbody>
</table>
SEQ ID NO: 12
LENGTH: 257
TYPE: PRT
ORGANISM: Homo sapiens

SEQUENCE: 12

Phe Glu Asn Phe Glu Lys Ile Asn Tyr Arg Phe Lys Asn Lys Ala 1 5 10 15
Tyr Leu Leu Glu Ala Phe Thr His Ala Ser Tyr His Tyr Asn Thr Ile 20 25 30
Thr Asp Cys Tyr Glu Arg Leu Glu Phe Leu Gly Asp Ala Ile Leu Asp 35 40 45
Tyr Leu Ile Thr Lys His Leu Tyr Glu Asp Pro Arg Glu His Ser Pro 50 55 60
Gly Val Leu Thr Asp Leu Arg Ser Ala Leu Val Asn Thr Ile Phe 65 70 75 80
Ala Ser Leu Ala Val Lys Tyr Asp Tyr His Lys Tyr Phe Lys Ala Val 95 99 95
Ser Pro Glu Leu Phe His Val Ile Asp Asp Phe Val Glu Phe Glu Leu 100 105 110
Glu Lys Asn Glu Met Glu Gly Met Asp Ser Glu Leu Arg Arg Ser Glu 115 120 125
Glu Asp Glu Glu Lys Glu Glu Asp Ile Glu Val Pro Lys Ala Met Gly 130 135 140
Asp Ile Phe Glu Ser Leu Ala Gly Ala Ile Tyr Met Asp Ser Gly Met 145 150 155 160
Ser Leu Glu Thr Val Tyr Glu Val Tyr Tyr Pro Met Met Arg Pro Leu 165 170 175
Ile Glu Lys Phe Ser Ala Asn Val Pro Arg Ser Pro Val Arg Glu Leu 180 185 190
Leu Glu Met Glu Pro Glu Thr Ala Lys Phe Ser Pro Ala Glu Arg Thr 195 200 205
Tyr Asp Gly Lys Val Arg Val Thr Val Glu Val Val Gly Lys Gly Lys 210 215 220
Phe Lys Gly Val Gly Arg Ser Tyr Arg Ile Ala Lys Ser Ala Ala 225 230 235 240
Arg Arg Ala Leu Arg Ser Leu Lys Ala Asn Glu Pro Glu Glu Val Pro 245 250 255
Ser
US 9,464,289 B2

-continued

Gly Val Leu Thr Asp Leu Arg Ser Ala Leu Val Asn Asn Thr Ile Phe 65 70 75 80
Ala Ser Leu Ala Val Lys Tyr Tyr His Lys Tyr Phe Lys Ala Val 85 90 95
Ser Pro Glu Leu Phe His Val Ile Asp Asp Phe Val Gln Phe Gln Leu 100 105 110
Glu Lys Asn Glu Met Gln Gly Met Asp Ser Leu Arg Arg Ser Glu 115 120 125
Glu Asp Glu Lys Glu Glu Asp Ile Glu Val Pro Lys Ala Met Gly 130 135 140
Asp Ile Phe Glu Ser Leu Ala Gly Ala Ile Tyr Met Asp Ser Gly Met 145 150 155 160
Ser Leu Glu Thr Val Trp Gln Val Tyr Tyr Pro Met Met Arg Pro Leu 165 170 175
Ile Glu Lys Phe Ser Ala Asn Val Pro Arg Ser Pro Val Arg Glu Leu 180 185 190
Leu Glu Met Glu Pro Glu Thr Ala Lys Phe Ser Pro Ala Glu Arg Thr 195 200 205
Tyr Asp Gly Lys Val Arg Val Thr Val Glu Val Gly Lys Gly Lys 210 215 220
Phe Lys Gly Val Gly Arg Ser Tyr Arg Ile Ala Lys Ser Ala Ala 225 230 235 240
Arg Arg Ala Leu Arg Ser Leu 245

<210> SEQ ID NO 14
<211> LENGTH: 234
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 14
Phe Glu Asn Phe Glu Lys Lys Ile Asn Tyr Arg Phe Lys Asn Lys Ala 1 5 10 15
Tyr Leu Leu Gln Ala Phe Thr His Ala Ser Tyr His Tyr Asn Thr Ile 20 25 30
Thr Asp Cys Tyr Gln Arg Leu Glu Phe Leu Gly Asp Ala Ile Leu Asp 35 40 45
Tyr Leu Ile Thr Lys His Leu Tyr Glu Asp Pro Arg Glu Asn His Ser Pro 50 55 60
Gly Val Leu Thr Asp Leu Arg Ser Ala Leu Val Asn Asn Thr Ile Phe 65 70 75 80
Ala Ser Leu Ala Val Lys Tyr Tyr His Lys Tyr Phe Lys Ala Val 85 90 95
Ser Pro Glu Leu Phe His Val Ile Asp Asp Phe Val Gln Phe Gln Leu 100 105 110
Glu Lys Asn Glu Met Gln Gly Met Asp Glu Asp Ile Glu Val Pro Lys 115 120 125
Ala Met Gly Asp Ile Phe Glu Ser Leu Ala Gly Ala Ile Tyr Met Asp 130 135 140
Ser Gly Met Ser Leu Glu Thr Val Trp Gln Val Tyr Tyr Pro Met Met 145 150 155 160
Arg Pro Leu Ile Glu Lys Phe Ser Ala Asn Val Pro Arg Ser Pro Val 165 170 175
Arg Glu Leu Leu Glu Met Glu Pro Glu Thr Ala Lys Phe Ser Pro Ala
<table>
<thead>
<tr>
<th>180</th>
<th>185</th>
<th>190</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glu Arg Thr Tyr Asp Gly Lys Val Arg Val Thr Val Glu Val Val Gly</td>
<td></td>
<td></td>
</tr>
<tr>
<td>195</td>
<td>200</td>
<td>205</td>
</tr>
<tr>
<td>Lys Gly Lys Phe Lys Gly Val Gly Arg Ser Tyr Arg Ile Ala Lys Ser</td>
<td></td>
<td></td>
</tr>
<tr>
<td>210</td>
<td>215</td>
<td>220</td>
</tr>
<tr>
<td>Ala Ala Ala Arg Arg Ala Leu Arg Ser Leu</td>
<td></td>
<td></td>
</tr>
<tr>
<td>225</td>
<td>230</td>
<td></td>
</tr>
</tbody>
</table>

<210> SEQ ID NO 15
<211> LENGTH: 550
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens

<400> SEQUENCE: 15

Met Asp Ser Glu Gln Ser Pro Ser Ile Gly Tyr Ser SerArg Thr Leu
1  5  10  15
Gly Pro Asn Pro Gly Leu Ile Leu Gln Ala Leu Thr Leu Ser Asn Ala
20  25 | 30
Ser Asp Gly Phe Asn Leu Glu Arg Leu Glu Met Leu Gly Asp Ser Phe
35  40 | 45
Leu Lys His Ala Ile Thr Thr Tyr Leu Phe Cys Thr Tyr Pro Asp Ala
50  55 | 60
His Glu Gly Arg Leu Ser Tyr Met Arg Ser Lys Val Ser Asn Cys
65  70  75  80
Asn Leu Tyr Arg Leu Gly Lys Lys Gly Leu Pro Ser Arg Met Val
95  99 | 100
Val Ser Ile Phe Asp Pro Pro Val Asn Trp Leu Pro Pro Gly Tyr Val
105 110
Val Asn Gln Asp Lys Ser Asn Thr Asp Lys Trp Glu Lys Asp Glu Met
115 120 125
Thr Lys Asp Cys Met Leu Ala Asn Gly Lys Leu Asp Glu Asp Tyr Glu
130 135 | 140
Glu Glu Asp Gly Glu Glu Met Ser Leu Met Trp Arg Ala Pro Lys Glu
145 150 155 | 160
Glu Ala Asp Tyr Glu Asp Asp Phe Leu Glu Tyr Asp Glu Glu His Ile
165 170 | 175
Arg Phe Ile Asp Arg Met Leu Met Gly Ser Gly Ala Phe Val Lys Lys
180 185 190
Ile Ser Leu Ser Pro Phe Ser Thr Thr Asp Ser Ala Tyr Glu Trp Lys
195 200 205
Met Pro Lys Ser Ser Leu Gly Ser Met Pro Phe Ser Ser Asp Phe
210 215 | 220
Glu Asp Phe Tyr Ser Ser Trp Asp Ala Met Cys Tyr Leu Asp Pro
225 230 235 | 240
Ser Lys Ala Val Gly Asp Lys Phe Val Val Gly Tyr Pro Asp Ala
245 250 | 255
Ser Glu Glu Asn Cys Gly Val Asp Thr Gly Lys Gln Ser Ile Ser Tyr
260 265 | 270
Asp Leu His Thr Glu Glu Cys Ile Ala Asp Lys Ser Ile Ala Asp Cys
275 280 | 285
Val Glu Ala Leu Leu Gly Cys Tyr Leu Thr Ser Cys Gly Glu Arg Ala
290 295 | 300
Ala Glu Leu Phe Leu Cys Ser Leu Gly Leu Lys Val Leu Pro Val Ile
305 310 315 320
Lys Arg Thr Asp Arg Glu Lys Ala Leu Cys Pro Thr Arg Glu Asn Phe 325 330 335
Asn Ser Gln Gln Lys Asn Leu Ser Val Ser Cys Ala Ala Ala Ser Val 340 345 350
 Ala Ser Ser Arg Ser Ser Val Leu Lys Asp Ser Glu Tyr Gly Cys Leu 355 360 365
Lys Ile Pro Pro Arg Cys Met Phe Asp His Pro Asp Ala Asp Lys Thr 370 375 380
Leu Asn His Leu Ile Ser Gly Phe Glu Asn Phe Glu Lys Lys Ile Asn 385 390 395 400
Tyr Arg Phe Lys Asn Lys Ala Tyr Leu Leu Glu Ala Ala Phe Thr His Ala 405 410 415
Ser Tyr His Tyr Asn Thr Ile Thr Asp Cys Tyr Gln Arg Leu Glu Phe 420 425 430
Leu Gly Asp Ala Ile Leu Asp Tyr Leu Ile Thr His Leu Tyr Glu 435 440 445
Asp Pro Arg Gln His Ser Pro Gly Val Leu Thr Asp Leu Arg Ser Ala 450 455 460
Leu Val Asn Asn Thr Ile Phe Ala Ser Leu Ala Val Lys Tyr Asp Tyr 465 470 475 480
His Lys Tyr Phe Lys Ala Val Ser Pro Glu Leu Phe His Val Ile Asp 485 490 495
Asp Phe Val Gln Phe Gln Leu Glu Lys Asn Glu Met Gln Gly Met Asp 500 505 510
515
Ser Glu Leu Arg Arg Ser Glu Glu Asp Glu Lys Gly Glu Asp Ile 520 525
Glu Val Pro Lys Ala Met Gly Asp Ile Phe Glu Ser Leu Ala Gly Ala 530 535 540
Ile Tyr Met Asp Ser Gly 545 550

<210> SEQ ID NO 16
<211> LENGTH: 549
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<400> SEQUENCE: 16
Asp Ser Glu Gln Ser Pro Ser Ile Gly Tyr Ser Ser Arg Thr Leu Gly 5 10 15
Pro Asn Pro Gly Leu Ile Leu Gln Ala Leu Thr Leu Ser Asn Ala Ser 20 25 30
Asp Gly Phe Asn Leu Glu Arg Leu Glu Met Leu Gly Asp Ser Phe Leu 35 40 45
Lys His Ala Ile Thr Thr Tyr Leu Phe Cys Thr Tyr Pro Asp Ala His 50 55 60
Glu Gly Arg Leu Ser Tyr Met Arg Ser Lys Val Ser Asn Cys Asn 65 70 75 80
Leu Tyr Arg Leu Gly Lys Lys Gly Leu Pro Ser Arg Met Val Val 85 90 95
Ser Ile Phe Asp Pro Pro Val Asn Trp Leu Pro Pro Gly Tyr Val Val 100 105 110
Asn Gln Asp Lys Ser Asn Thr Asp Lys Trp Glu Lys Asp Glu Met Thr 115 120 125
Lys Asp Cys Met Leu Ala Asn Gly Lys Leu Asp Glu Asp Tyr Glu Glu 130 135 140
<table>
<thead>
<tr>
<th></th>
<th>Glu</th>
<th>Asp</th>
<th>Glu</th>
<th>Glu</th>
<th>Glu</th>
<th>Ser</th>
<th>Leu</th>
<th>Met</th>
<th>Trp</th>
<th>Arg</th>
<th>Ala</th>
<th>Pro</th>
<th>Lys</th>
<th>Glu</th>
</tr>
</thead>
<tbody>
<tr>
<td>145</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>150</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>155</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>160</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>165</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>170</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>175</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>180</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>185</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>190</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>195</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>200</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>205</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>210</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>215</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>220</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>225</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>230</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>235</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>240</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>245</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>250</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>255</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>260</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>265</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>270</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>275</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>280</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>285</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>290</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>295</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>300</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>305</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>310</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>315</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>320</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>325</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>330</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>335</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>340</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>345</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>350</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>355</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>360</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>365</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>370</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>375</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>380</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>385</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>390</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>395</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>400</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>405</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>410</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>415</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>420</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>425</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>430</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>435</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>440</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>445</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>450</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>455</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>460</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>465</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>470</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>475</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>480</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>485</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>490</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>495</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>500</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>505</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>510</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>515</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>520</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>525</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>530</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>535</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>540</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>545</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
SEQ ID NO 17
LENGTH: 10323
TYPE: DNA
ORGANISM: Homo sapiens

SEQUENCE:

17

cggagcgcc gcgcagcttg cgtcagcgc cggagcagct cggagctgct cggagctgct cggagctgct cggagctgct 60
gagcgcgccc gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc 120
ggcctggtgg gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc gcgcagcggc 180
cgcctggtgc cagtaaagg cgcgtaggaa aaaaatggct gtcgtagaag aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct 240
gaaagctcct ggttggcact ccccttagct ggcgcgtgttg cctctctgcc cttagaggtg ggcgcgtgttg cctctctgcc cttagaggtg 300
cctcagcgtg ggcgctcttc ttgactattg agaggtgcaag aaaaatggct cgcctatttt cttagaggtg ggcgcgtgttg cctctctgcc 360
ttaagccac aagaaatagt gttgagctga gtcgtagaag aaaaatggct 420
cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 480
gttcgctgttt ttcagctcgc aacccttggc aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 540
cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 600
tgctccgata cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 660
gtttaacta acaatgggaac cgaagagaag cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 720
tggtaatact tccctgggct aacccttggc aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 780
cctagacac ccaatccttga cggagcgcc aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 840
catttttgga ctcacgctggc caaaaagaga aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 900
aacagtcttc cccctgggct aacccttggc aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 960
gttctcttc agctgtctct cccctgggct aacccttggc aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 1020
tgcagacaag gggctttcag cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1080
taataactcgc cccctgggct aacccttggc aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 1140
gatccttttt cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1200
caagacagct aacccttggc aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 1260
cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1320
gatccttttt cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1380
cgaaattttc gggctgttca aacccttggc aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa aaaaatggct cagtaaagg cgcgtaggaa 1440
ttttttttttt cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1500
caagacagct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1560
ttttttttttt cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1620
caagacagct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1680
caagacagct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1740
gagaggttgg gggctttcag cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1800
ttttttttttt cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1860
caagacagct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1920
caagacagct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 1980
ttttttttttt cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 2040
ttttttttttt cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 2100
ttttttttttt cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct cggcagccct 2160
accaagtga tcggttaactt actctagctec taaatgcaga acccgagagt tgcctgatgg
2220
tacattttat tcacactctt attctgccaat tctcagcctt ccgtttactc atctagctcc taaatgcaga acccgagagt tgcctgatgg
2280
tccacagtga atcggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
2340
tcgcacaaa atggatgtgg tctcagcctt ccgtttactc atctagctcc taaatgcaga acccgagagt tgcctgatgg
2400
atatgaagag gagcttgatt tgcagatgga agaagaagcc atgtgtctag gagacagtcc
2460
ttccacagaaa caagcgctag gctacccaaa agcaatcaca gagaagtccg gggatagtta
ttaataattac ccgttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
2520
taacagcc acaatcaactt gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
2580
tggcagtcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
2640
ttgagctcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
2700
ttcgctgag tcggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
2760
atattgctag tgcagatgga agaagaagcc atgtgtctag gagacagtcc
2820
caatcacttg ctcgctcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
2880
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
2940
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3000
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3060
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3120
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3180
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3240
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3300
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3360
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3420
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3480
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3540
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3600
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3660
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3720
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3780
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3840
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3900
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
3960
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
4020
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
4080
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
4140
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
4200
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
4260
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
4320
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
4380
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
4440
tctcagcag gctacccaaa agcaatccaa gacggttaacctt actctagctec taaatgcaga acccgagagt tgcctgatgg
4500
<table>
<thead>
<tr>
<th>Sequence</th>
<th>Continued</th>
</tr>
</thead>
<tbody>
<tr>
<td>tgaggaggag gagaagctga tggtggagggc tccgaaaggaa gagggtcact atgaagatga</td>
<td>4560</td>
</tr>
<tr>
<td>ttctcgtggg atatatcagc atatatagat atatatattt ataatgtgtga</td>
<td>4620</td>
</tr>
<tr>
<td>gcgtctgtgc gagaagcctg ggtggtggtg ggtggtggtg ggtggtggtg</td>
<td>4680</td>
</tr>
<tr>
<td>gaaatgccc aaaaaatcct cccagaatttt ccccaaaaag ccccaaaaag</td>
<td>4740</td>
</tr>
<tr>
<td>tggaggggtg caggtgtact ctcgtgcag ctcgtgcag ctcgt gcgtgc</td>
<td>4800</td>
</tr>
<tr>
<td>tcttggggag cagggagaag gaaaatgccc cccagaatttt ccccaaaaag ccccaaaaag</td>
<td>4860</td>
</tr>
<tr>
<td>cgagggagaag gaaaatgccc cccagaatttt ccccaaaaag ccccaaaaag</td>
<td>4920</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>4980</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5040</td>
</tr>
<tr>
<td>ggtcattctgc tcttggggag cagggagaag gaaaatgccc cccagaatttt ccccaaaaag ccccaaaaag</td>
<td>5100</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5160</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5220</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5280</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5340</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5400</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5460</td>
</tr>
<tr>
<td>ggtcattctgc tcttggggag cagggagaag gaaaatgccc cccagaatttt ccccaaaaag ccccaaaaag</td>
<td>5520</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5580</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5640</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5700</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5760</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5820</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5880</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>5940</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6000</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6060</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6120</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6180</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6240</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6300</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6360</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6420</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6480</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6540</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6600</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6660</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6720</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6780</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6840</td>
</tr>
<tr>
<td>ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag ccccaaaaag</td>
<td>6900</td>
</tr>
</tbody>
</table>
tgatcctttgg caaaaccc cattttaca aagctctctt ggcaggttagt tccccactgat

6960

ggaaatgttt atggcaatata attttgcctt ctaggctgtt gctctaacaa aataaacctt

7020

agacataca caacttaaa atgctcaga ttttttaaat gatttgtaac tttattaaga

7080

agcaaaacac agcactttta cctcagttct cctccataaa attttctact atacttttca

7140

taagtgcga tcgatatttac acctacaaaa gctggtgctt tatacgccg aagttttaa

7200

gttggcaga aagtgcgta attttgatac atctgtgatt taggctatta atttagataa

7260

aactgctct tattttcact tttggaaaaag gaaaaaattt aaaactcttt taggccttgg

7320

cctaatgttc tttaattaga ctttgagcct cttttacact aaatataacct ctgtttttt

7380

ttttttgtc gcattttttcc cctggtgctt gctttctagt tatttttttt

7440

aatatttttt tdttttgtca gtaatcatatag ttttttttta aataactctt taaacgattg

7500

gggttcagtt ccgctcctcc tcaaccatac agtaagagtt tgcgctcaatt tcggccgatta

7560

attatgttgg aagtttaaga aagtgcgacta cgggctcctt atatcttccca gcacatgatt

7620

cgtaacgtgg tggccctctc ctttacttga atttctgtaa ctctgcacat taaaagttca

7680

ttcgctggttt ctggccctcc ctaaaccaca agaaagaccc ctgctgagag tggcctgaaaa

7740

gaaaaacgtc agctcctggtctct gaaaagctaag ctgctctttt ttaattcaac

7800

gttttttttttttgcacca tcttttattt atttgaagta attgtttttt

7860

ttgagcgaat tagggtcatt gttttttttta attttaattc gatggtggaattgcaagtgtt

7920

catatttttatat ttttattttttt atatatatat tttttttttttt

7980

taatatttttac aagtaaaat gtagttttttt gatggttatttt ggtatataga

8040

gggtgattttg gtcgagttttat cggatccag cttcctttttt attttttttttt

8100

atttgctgcct ccctatatcg atggtttttata cctcttttta ctttcttccc

8160

ctttatat accatgtcctttt cctttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttttt
US 9,464,289 B2

-continued

**SEQ ID NO 18**

**LENGTH:** 1374

**TYPE:** PRT

**ORGANISM:** Schizosaccharomyces pombe

**SEQUENCE:**

```
gatattttta tttgttaata atactttgcc ctcagaaaga ttctgatacc ctgccttgac
aacatgaaac ttgaggctgc tttggttcat gaatccaggt gttcccccgg cagtcggctt
cttcagtcgc tccctggagg caggtgggca ctgcagagga tcactggaat ccagatcgag
cgcagttcat gcacaaggcc ccgttgattt aaaatattgg atcttgctct gttagggtgt
taatccctt tacacaagat tgaagccacc aaactgagac cttgatacct ttttttaact
gcatctgaaa ttatgttaag agtctttaac ccatttgcat tatctgcaga agagaaactc
atgtcatgtt tattacctat atggttgttt taattacatt tgaataatta tattttttca
acacoaggt acttggcaag aatatttaa tttccagata aatattttta aatattttg
tacagaaaa gtatataagg atcaactctta tttaaaggt gttaaagtt
gttggagagc ttaatggaag atatgcaagg agaactagtgt aatattttcaact
gagcagaaaa atgtatttttt caacaactt tattttatata ttttttcat attttttttc
ttttattttg gcaactcaac ccagagct ttaattttta tattttttgtcat
aaattttgaa cggcagcaga ggtttatatag caaattgtgca aaagttaaatg
tttgtaacgcc cttggttaag aatatttttattg aattttaatattttcaact
gatcacaatt acctttagtt gttttttttg taataattgt agccaagtaa atctccaata
aagattcgt ctgtttcaaa aaaaaaaaaa aaaaaaaaaa aaaaaaaaaa aaaaaaaaaa
```

Met Asp Ile Ser Ser Phe Leu Leu Pro Gln Leu Leu Arg Lys Tyr Gln
1  5  10  15
Gln Asp Val Tyr Asp Ile Ala Ser Lys Glu Gln Asn Thr Leu Leu Val Met
20  25  30
Arg Thr Gly Ala Gly Lys Thr Leu Ala Val Lys Leu Ile Lys Glu
35  40  45
Lys Leu Glu Glu Gln Ile Leu Ile Gln Glu Ser Asn Leu Glu His Lys
50  55  60
Lys Ile Ser Val Phe Leu Val Asn Lys Val Pro Leu Val Phe Gln Glu
65  70  75  80
Ala Glu Tyr Ile Arg Ser Gln Leu Pro Ala Lys Val Gly Met Phe Tyr
85  90  95
Gly Glu Leu Ser Ile Glu Met Ser Glu Gln Leu Thr Asn Ile Ile
100 105 110
Leu Lys Tyr Asn Val Ile Val Ile Thr Ala Asp Leu Phe Tyr Leu Phe
115 120 125
Leu Ala Arg Gly Phe Leu Ser Ile Asn Asp Leu Asn Leu Ile Ile Phe
130 135 140
Asp Glu Cys His Asp Ala Ile Gly Asn Asp Ala Tyr Ala Arg Ile Met
145 150 155 160
Asn Asp Phe Tyr His Asp Ala Val Leu Ser Lys Lys His Phe
165 170 175
Ile Arg Met Gly Leu Ile Asp His Leu Lys Pro Leu Asp Phe Arg
Arg Lys Ile Ala Asp Leu Glu Thr Leu Glu Glu Asp Glu Leu Lys Asp
Glu Gly Tyr Ile Glu Thr Tyr Glu Arg Tyr Val Pro Lys Ser Trp Met
Lys Val Pro Glu Asp Ile Thr Arg Cys Phe Val Ser Leu Leu Tyr Thr
Asp Ala Asn Gly Asp Asn His Ile Phe His Pro Leu Val Phe Val
Gln Ala His Ser Phe Pro Lys Ile Asp Ser Phe Ile Leu Asn Ser Thr
Val Gly Pro Arg Val Lys Ile Val Leu Glu Thr Ile Glu Asp Ser Phe
Lys Ile Asp Ser His Leu Leu Glu Leu Leu Lys Lys Ser Thr Arg Tyr
Leu Leu Gln Phe Gly Leu Ser Thr Ser Leu Glu Gin Gin Ile Pro Thr
Pro Tyr Trp Leu Ala Pro Leu Asn Leu Ser Cys Thr Asp Tyr Arg Phe
Leu Gln Leu Ile Asp Val Asp Thr Ile Gin Asp Phe Lys Leu
Pro Glu Pro Val Gin Asn Val Thr Asp Leu Gin Ser Asp Thr Val Leu
Leu Val Asn Pro Gin Ser Ile Tyr Glu Gin Tyr Ala Phe Glu Gly Phe
Val Asn Ser Glu Phe Met Ile Pro Ala Lys Lys Asp Lys Ala Pro
Ser Ala Leu Cys Lys Lys Leu Pro Leu Arg Leu Asn Tyr Ser Leu Trp
Gly Asn Arg Ala Lys Ser Ile Pro Lys Ser Gin Gin Val Arg Ser Phe
Tyr Ile Asn Asp Leu Tyr Ile Leu Pro Val Ser Arg His Leu Lys Asn
Ser Ala Leu Ile Pro Ser Ile Leu Tyr His Ile Gin Leu Asn Leu Leu
Val Ala Ser Ser Phe Ile Glu His Phe Arg Leu Asp Cys Lys Ile Asp
Thr Ala Cys Gin Ala Leu Thr Ser Ala Glu Ser Gin Leu Asn Phe Asp
Tyr Asp Arg Leu Glu Phe Tyr Gly Asp Cys Phe Leu Lys Leu Gly Ala
Ser Ile Thr Val Phe Leu Lys Phe Pro Asp Thr Gin Glu Tyr Gin Leu
His Phe Asn Arg Lys Ile Ile Ser Asn Cys Asn Leu Tyr Lys Val
Ala Ile Asp Cys Glu Leu Pro Lys Tyr Ala Leu Ser Thr Pro Leu Glu
Ile Arg His Thr Cys Pro Tyr Gly Phe Gin Lys Ser Thr Ser Asp Lys
Cys Arg Tyr Ala Val Leu Gin Lys Leu Ser Val Lys Arg Ile Ala
Asp Met  Val Glu Ala Ser Ile  Gly Ala Cys Leu Leu  Asp Ser Gly  1025  1030  1035
Leu Asp  Ser Ala Leu Lys Ile  Cys Lys Ser Leu Ser  Val Gly Leu  1040  1045  1050
Leu Asp  Ile Ser Asn Trp Asp  Glu Trp Asn Tyr  Phe Asp Leu  1055  1060  1065
Asn Thr  Tyr Ala Asp Ser Leu  Arg Asn Val Gln Phe  Pro Tyr Ser  1070  1075  1080
Ser Tyr  Ile Glu Glu Thr Ile  Gly Tyr Ser Phe Lys  Asn Lys Lys  1085  1090  1095
Leu Leu  His Leu Ala Phe Ile  His Pro Ser Met Met  Ser Gln Gln  1100  1105  1110
Gly Ile  Tyr Glu Asn Tyr Gln  Gln Leu Glu Phe Leu  Gly Asp Ala  1115  1120  1125
Val Leu  Asp Tyr Ile Val  Gln Tyr Leu Tyr Lys  Lys Tyr Pro  1130  1135  1140
Asn Ala  Thr Ser Gly Glu Leu  Thr Asp Tyr Lys Ser  Phe Tyr Val  1145  1150  1155
Cys Asn  Lys Ser Leu Ser Tyr  Ile Gly Phe Val Leu  Asn Leu His  1160  1165  1170
Lys Tyr  Ile Gln His Glu Ser  Ala Ala Met Cys  Asp Ala Ile Phe  1175  1180  1185
Glu Tyr  Gln Glu Leu Ile Glu  Ala Phe Arg Glu Thr  Ala Ser Glu  1190  1195  1200
Asn Pro  Trp Phe Trp Phe Glu  Ile Asp Ser Pro Lys  Phe Ile Ser  1205  1210  1215
Asp Thr  Leu Glu Ala Met Ile  Cys Ala Ile Phe Leu  Asp Ser Gly  1220  1225  1230
Phe Ser  Leu Gln Ser Leu Gln  Phe Val Leu Pro Leu  Phe Leu Asn  1235  1240  1245
Ser Leu  Gly Asp Ala Thr His  Thr Lys Ala Lys Gly Ile Glu  1250  1255  1260
His Lys  Val Tyr Gln Leu Leu  Lys Asp Gln Gly Cys  Glu Asp Phe  1265  1270  1275
Gly Thr  Lys Cys Val Ile Glu  Glu Val Lys Ser Ser  His Lys Thr  1280  1285  1290
Leu Leu  Asn Thr Glu Leu His  Leu Thr Lys Tyr Tyr  Gly Phe Ser  1295  1300  1305
Phe Phe  Arg His Gly Asn Ile  Val Ala Tyr Gly Lys  Ser Arg Lys  1310  1315  1320
Val Ala  Asn Ala Lys Tyr Ile  Met Lys Gln Arg Leu  Leu Lys Leu  1325  1330  1335
Leu Glu  Asp Lys Ser Asn Leu  Leu Leu Tyr Ser Cys  Asn Cys Lys  1340  1345  1350
Phe Ser  Lys Lys Pro Ser  Asp Glu Gln Ile Lys  Gly Asp Gly  1355  1360  1365
Lys Val  Lys Ser Leu Thr  1370

<210> SEQ ID NO 19
<211> LENGTH: 4125
<212> TYPE: DNA
<213> ORGANISM: Schizosaccharomyces pombe
atgcatattt caagtgtttct actctctcaa cttttaagta aatatcaaca agatgtgtat 60
aatatcgcga gcagacgaaaa tacctttactt tttgatagaa cgggcgctgg taagacatta 120
cctgtgtga agttggtaaaa ccaagaagtc ggagagcga tttatacga agatacataa 180
c tgtggacaata aaaaaatatac agttttttct gcaacaaag tgtctttgtg atttcaaca 240
gcggatataa tctcgtctca actacgggtc aaggttgccg tggtttttga cgattattct 300
atagaataag gcagacgagtt gttgactaat attatattga agtataatgt gttggtaat 360
actgcaagtatt gtgtcctgtca agagtttccc tttttaataaa tttttaaatg 420
ttaattatat ctcacgaatag tcatactgca atttgaaatg atgtgattatg tgcatacag 480
aatatgattt atcagagaga caagcgataa tgttaaaaata aacatcctac cttacacaaga 540
atttttggta tgcactgcoc aaccactccg gaaaaaaaga gaaacctata caacctgact 600
tatcaaatgg acaaatatt tggatcattaa gcaccgggttg cttgccagaa cagacgcc 660
gattaacttt tctttccgga agaagagat attagatatttg gtttggctca gaaaaaaca 720
cacctcgatt catatcaac gaaatcgccg aagacatctt gtttataatc 780
ggggtcgtta agonctgtttt ggcagaaaaa cctctctctc gttttgctca gaaaaaca 840
gttggtgtat attttttgtga ttttttgaga aagaaaaatg gtttttggtt 900
aacgcgttgct cagatgacatgg ggaacagggta atggagcgc ttaaaaattt ctgtaagag 960
tggaaaaaata caaataatag agacacatggc cctaggacag cgattttttc ttccagct 1020
gtacgtaa aagttttttg attcttgaag tttttaattt cttcagctca gtaaatctc 1080
agttacgctt atggagatctc atcattctca ttgctctctt ttaatgctaa tggacgcct 1140
tgaaacatct tttactgccc ttaaccttcg 1200
gactgcggtg aattttctttt cactccggag cggaaagaaag atacacccca taagtttaag 1260
actgcaaacat tatattgattt aattgctact gcgattgacg aagaagatct caagagctca 1320
tcattcact tagtattcact ttcataattt ttttggtgcgt tcacagctgta tcgctaatct 1380
cagggtagag cagagaaagat ggcgttcaag ttctcttgtt ttttaaaacag agaagctggtggt 1440
ttaatttattg atagccctcg caaataataa tttgagctt ccctggcagc 1500
agcacttcca atctttttga ttctcaggta ttgtggaaaga gaaacaggtg gcgagctttg 1560
atcgctcatg aagttggccga gacttggtcg tttccagcag gttttgtctg aggtttgctg 1620
ccttatcaacct ttttcaccac accctcctac gacgtatac taaccatatca caagatatattctaatctat 1680
acagccaaact cctgctttctt aaggtgttaga ttttttttgg tagaattggc cctgagcctg 1740
aataattcggc gggcttcagc atttccagcc attatcctgag ataaccatca gggagacgaa acagaaagctg 1800
gctgtcataa tggattttctg cttgtcttcg atgggtagta aagagacatc aaaaaccc 1860
cagtattta gaaatattta gtcgagccttc ctaagcttctgacttcttttc gaaattcttagagagctgc 1920
gaggtttataa ctagagttca cttgagctga attcataagaa agggcagct 1980
agatattcagg tagttcatct ttttcttttc attttgagtt agatctgtagtagttagtag 2040
aataactactt cttcctacttt tataatcgttag ctattagattg gaaactactatc 2100
cattaactc cttgtgtaaa gtttgtgcgg ctctgttattt gtttttattt gtttttattt 2160
aagatcatttt ccgttcttgc tggagctttt gtttggctgg atggagttatc acagcttgct 2220
gtgatatcct cttgcttgtgca gagaatac cttctatcctc ctggtgctgc cccctatatc 2280
ttgctctgaa gcagttgagaatctgtatag aaggtgtagc atgtgagcact tatccaaaaat 2340
ttttttaaat taacggaaacc tggctcaaat gttactgttt tcgaatcgcga tactgtatta 2400

<210> SEQ ID NO 20
<211> LENGTH: 754
<212> TYPE: PRT
<213> ORGANISM: Giardia lamblia virus

<400> SEQUENCE: 20

Met His Ala Leu Gly His Cys Thr Val Val Thr Thr Arg Gly Pro
1       5       10      15

Ser His Trp Leu Leu Leu Leu Asp Thr His Leu Gly Thr Leu Pro Gly
20     25     30

Phe Lys Val Ser Ala Gly Arg Gly Leu Pro Ala Ala Glu Val Tyr Phe
35     40     45

Glu Ala Gly Pro Arg Val Ser Leu Ser Arg Thr Asp Ala Thr Ile Val

Ala Val Tyr Gln Ser Ile Leu Phe Gln Leu Leu Leu Gly Pro Thr Phe Pro
65 70 75 80
Ala Ser Trp Thr Glu Ile Gly Ala Thr Met Pro His Asn Glu Tyr Thr
85 90 95
Phe Pro Arg Phe Ile Ser Asn Pro Pro Glu Phe Ala Thr Leu Ala Phe
100 105 110
Leu Pro Leu Leu Ser Pro Thr Ser Pro Leu Asp Leu Arg Ala Leu Met
115 120 125
Val Thr Ala Gln Leu Met Cys Asp Ala Lys Arg Leu Ser Asp Glu Tyr
130 135 140
Thr Asp Tyr Ser Thr Leu Ser Ala Ser Leu His Gly Arg Met Val Ala
145 150 155 160
Thr Pro Glu Ile Ser Trp Ser Leu Tyr Val Val Leu Gly Ile Asp Ser
165 170 175
Thr Glu Thr Ser Leu Ser Tyr Phe Thr Arg Ala Asn Glu Ser Ile Thr
180 185 190
Tyr Met Arg Tyr Tyr Ala Thr Ala His Asn Ile His Leu Arg Ala Ala
195 200 205
Asp Leu Pro Leu Val Ala Val Arg Leu Asp Leu Lys Asp His
210 215 220
Gln Ile Pro Ala Pro Gly Ser Trp Asp Ala Ala Pro Lys Leu Arg
225 230 235 240
Phe Leu Pro Pro Glu Leu Cys Leu Leu Leu Pro Asp Glu Phe Asp Leu
245 250 255
Ile Arg Val Gln Ala Leu Gln Phe Leu Pro Glu Ile Ala Lys His Ile
260 265 270
Cys Asp Ile Gln Asn Thr Ile Cys Ala Leu Asp Lys Ser Phe Pro Asp
275 280 285
Cys Gly Asp Ile Gly Gly Arg Tyr Phe Ala Ile Thr Ala Gly Leu
290 295 300
Arg Leu Asp Gin Gly Arg Gly Arg Leu Ala Gly Trp Arg Thr Pro
305 310 315 320
Phe Gly Pro Phe Gly Val Ser His Thr Asp Val Phe Gin Arg Leu Glu
325 330 335
Leu Leu Gly Asp Ala Val Leu Gly Phe Ile Val Thr Ala Arg Leu Leu
340 345 350
Cys Leu Phe Pro Asp Ala Ser Val Gly Thr Leu Val Glu Leu Lys Met
355 360 365
Glu Leu Val Arg Asn Glu Ala Leu Asn Tyr Leu Val Gin Thr Leu Gly
370 375 380
Leu Pro Gln Leu Ala Gln Phe Ser Asn Asn Leu Val Ala Lys Ser Lys
385 390 395 400
Thr Trp Ala Asp Met Tyr Glu Ile Val Gly Ser Ile Phe Thr Gly
405 410 415
Pro Asn Gly Ile Tyr Gly Cys Glu Glu Phe Leu Ala Lys Thr Leu Met
420 425 430
Ser Pro Glu His Ser Lys Thr Val Gly Ser Ala Cys Pro Asp Ala Val
435 440 445
Thr Lys Ala Ser Lys Arg Val Cys Met Gly Glu Ala Gly Ala His Glu
450 455 460
Phe Arg Ser Leu Val Asp Tyr Ala Cys Glu Gin Gly Ile Ser Val Phe
465 470 475 480
Cys Ser Ser Arg Val Ser Thr Met Phe Leu Glu Arg Leu Arg Asp Ile 485 490 495
Pro Ala Glu Asp Met Leu Asp Trp Tyr Leu Gly Ile Gln Phe Ser 500 505 510
His Arg Ser Gly Leu Ser Gly Pro Gly Gly Val Ser Val Ile Asp 515 520 525
Ile Met Thr His Leu Ala Arg Gly Leu Trp Leu Gly Ser Pro Gly Phe 530 535 540
Tyr Val Glu Gln Gln Thr Asp Lys Asn Glu Ser Ala Cys Pro Pro Thr 545 550 555 560 (truncated)
Ile Pro Val Leu Tyr Ile Tyr His Arg Ser Val Gln Cys Pro Val Leu 565 570 575
Tyr Gly Ser Leu Thr Glu Thr Pro Thr Gly Pro Val Ala Ser Lys Val 580 585 590
Leu Ala Leu Tyr Glu Ile Leu Ala Tyr Glu Ser Gly Ser Gly Ser 595 600 605
Lys His Ile Ala Ala Gln Thr Val Ser Arg Ser Leu Ala Val Pro Ile 610 615 620
Pro Ser Gly Thr Ile Pro Phe Leu Ile Arg Leu Gln Ile Ala Leu 625 630 635 640
Thr Pro His Val Tyr Gln Leu Leu Gly Leu Gly Asp Ala Phe Leu 645 650 655
Lys Cys Ser Leu Ala Leu His His Ala Leu His Pro Thr Leu Thr 660 665 670
Glu Gly Ala Leu Thr Arg Met Arg Gln Ser Ala Glu Thr Arg Ser Val 675 680 685
Leu Gly Arg Leu Thr Lys Arg Phe Pro Ser Val Ser Gly Val Ile 690 695 700
Ile Glu Ser His Pro Lys Ile Gln Pro Asp Ser Lys Tyr Gly Asp 705 710 715 720
Thr Phe Glu Ala Ile Leu Ala Ile Leu Ala Cys Gly Gly Glu 725 730 735
Ala Ala Gly Ala Phe Val Arg Glu His Val Leu Pro Glu Val Val Ala 740 745 750
Asp Ala

<210> SEQ ID NO: 21
<211> LENGTH: 2265
<212> TYPE: DNA
<213> ORGANISM: Giardia lamblia virus
<400> SEQUENCE: 21
atgcatgctt tgggacactg ttgcacagtt gtgactacta gaggaccatc ccactggttg 60
tcaacctag acactcaacct gggaaccttg ccaggggtta aagttactgc aggcgcaggg 120
ttcctcgcgc cagaggtgta ctttgaagcg ggtccgaggg tgtctctctc tcgaactgat 180
gcaactatag tagccgtgta tcagtccatt ctctttcagc tgcctgggacc cacatttcct 240
gcttcatgga ctgagattgg acgaacatg ccctcgattt atatccaatc caccacaatt 300
gcccaccctg gcatttttac ccttactatc tcctaccagc cctctggact tgcgtgcatt 360
aatagctggtc tctttatgtc gttcttggga ttcgattctac ccaaactagc actcccgaaa 420
tgcataactgc tgggacagtt gtgactacta gaggaccatc ccactggttg 480
atgcatgctt tgggacactg ttgcacagtt gtgactacta gaggaccatc ccactggttg 540
ccccgggttc a cgccattctc c tgcctcagc c tcaacgatca gctgggacta c aggcgcccg  60  
acaccactct cgcctcagc ctcacgagta gctgggacta c aggcgcccg  2265  
<210> SEQ ID NO: 22
<211> LENGTH: 91
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: oligonucleotide
<400> SEQUENCE: 22
ccccgggttca cgccattctc c tgcctcagc c tcaacgatca gctgggacta c aggcgcccg  60  
acaccactct cgcctcagc ctcacgagta gctgggacta c aggcgcccg  91
<210> SEQ ID NO: 23
<211> LENGTH: 29
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: Oligonucleotide

<400> SEQUENCE: 23
tgagtcagg agaatcagac cattccggc

<210> SEQ ID NO 24
<211> LENGTH: 29
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: oligonucleotide

<400> SEQUENCE: 24
tgagtcagg agaatcagac cattccggc

<210> SEQ ID NO 25
<211> LENGTH: 29
<212> TYPE: DNA
<213> ORGANISM: Artificial Sequence
<220> FEATURE:
<223> OTHER INFORMATION: oligonucleotide

<400> SEQUENCE: 25
tgagtcagg agaatcagac cattccggc

<210> SEQ ID NO 26
<211> LENGTH: 29
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens
<400> SEQUENCE: 26
tgagtcagg agaatcagac cattccggc

<210> SEQ ID NO 27
<211> LENGTH: 29
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens
<400> SEQUENCE: 27
ggccgggcgc ggtggtctac gggtgctcac ccgacacttt gggagggcga ggcgggtgga 60
tcactgtgg tcggaggtct gagagcagcc tggcccaacat ggtgaamccc cgcttcctact 120
aaaaatacga aatagcgcg aagtaatggg cggcgctctg tgtcccacc tattctggag 180
gttaggcgg gagaatctct tgaaccgggg agggcagact tgcagtgcag cgggtagcgc 240
cactgcac ccggctcggg caacaagagc gcgacttggt ctcacaaaaa a 291

<210> SEQ ID NO 28
<211> LENGTH: 302
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens
<400> SEQUENCE: 28
ggccgggcgc aatggtctag acctctact acggacacttt ggcagcctga ggcgggcaaa 60
tcactgtgg tcggaggtct gaaacactcc ttggtgctca ggtgaamccc cgcttcctact 120
aaaaatacga aatagcgcg aagtaatggg cggcgctctg tgtcccacc tattctggag 180
gggaatggcgtgaacccctg gggccgctctg ttacgaggtcg ccggagttgc gcacagtgcac 240
tcactgcac ccggctcggg caacaagagc gcgacttggt ctcacaaaaa a 300
aa 302
What is claimed is:

1. An isolated nucleotide molecule selected from:
   a double-stranded RNA molecule that inhibits expression of Alu RNA, wherein a first strand of the double-stranded RNA comprises a sequence selected from SEQ ID NO: 1, 2, 3, 4, 5, and 6 and including about 19 to 25 nucleotides; and
   a vector comprising an oligonucleotide that inhibits the expression of Alu RNA, comprising a sequence selected from SEQ ID NO: 22, 23, 24, and 25 and including about 29 to 100 nucleotides; and
   a vector comprising an oligonucleotide that inhibits the expression of Alu RNA, consisting of the sequence of SEQ ID NO: 26.