(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
27 April 2017 (27.04.2017)

WIPO | PCT

(10) International Publication Number
WO 2017/070623 A1

(51) International Patent Classification:
A61K 39/245 (2006.01) *A61K 31/7115* (2006.01)
A61K 31/7105 (2006.01) *A61P 31/22* (2006.01)

(21) International Application Number:
PCT/US2016/058322

(22) International Filing Date:
21 October 2016 (21.10.2016)

(25) Filing Language: English

(26) Publication Language: English

(30) Priority Data:

62/245,031	22 October 2015 (22.10.2015)	US
62/245,159	22 October 2015 (22.10.2015)	US
62/247,576	28 October 2015 (28.10.2015)	US
62/248,252	29 October 2015 (29.10.2015)	US

(71) Applicant: **MODERNATX, INC.** [US/US]; 320 Bent Street, Cambridge, MA 02141 (US).

(72) Inventors: **CIARAMELLA, Giuseppe**; 1 Hawes Road, Sudbury, MA 01776 (US). **JOHN, Shinu**; 318 Rindge Avenue, Unit 403, Cambridge, MA 02140 (US). **BETT, Andrew, J.**; 834 Collins Avenue, Lansdale, PA 19446 (US). **CASIMIRO, Danilo, R.**; 626 Westbourne Road, Harleysville, PA 19438 (US).

(74) Agents: **DIPIETRANTONIO, Heather, J.** et al.; Wolf, Greenfield & Sacks, P.C., 600 Atlantic Avenue, Boston, MA 02210-2206 (US).

(81) Designated States *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DJ, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KN, KP, KR, KW, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) Designated States *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

Published:

— *with international search report (Art. 21(3))*

— *with sequence listing part of description (Rule 5.2(a))*

WO 2017/070623 A1

(54) Title: HERPES SIMPLEX VIRUS VACCINE

(57) Abstract: The disclosure relates to herpes simplex virus (HSV) ribonucleic acid (RNA) vaccines, as well as methods of using the vaccines and compositions comprising the vaccines.

HERPES SIMPLEX VIRUS VACCINE

RELATED APPLICATIONS

This application claims the benefit under 35 U.S.C. § 119(e) of U.S. provisional application number 62/245,159, filed October 22, 2015, U.S. provisional application number 62/247,576, filed October 28, 2015, and U.S. provisional application number 62/248,252, filed October 29, 2015, each of which is incorporated by reference herein in its entirety. This application also claims the benefit under 35 U.S.C. § 119(e) of U.S. provisional application number 62/245,031, filed October 22, 2015, which is incorporated by reference herein in its entirety.

BACKGROUND

Herpes simplex viruses (HSV) are double-stranded linear DNA viruses in the *Herpesviridae* family. Two members of the herpes simplex virus family infect humans – known as HSV-1 and HSV-2. Symptoms of HSV infection include the formation of blisters in the skin or mucous membranes of the mouth, lips, and/or genitals. HSV is a neuroinvasive virus that can cause sporadic recurring episodes of viral reactivation in infected individuals. HSV is transmitted by contact with an infected area of the skin during a period of viral activation.

Deoxyribonucleic acid (DNA) vaccination is one technique used to stimulate humoral and cellular immune responses to foreign antigens, such as HSV antigens. The direct injection of genetically engineered DNA (*e.g.*, naked plasmid DNA) into a living host results in a small number of its cells directly producing an antigen, resulting in a protective immunological response. With this technique, however, come potential problems, including the possibility of insertional mutagenesis, which could lead to the activation of oncogenes or the inhibition of tumor suppressor genes.

SUMMARY

Provided herein are ribonucleic acid (RNA) vaccines that build on the knowledge that modified RNA (*e.g.*, messenger RNA (mRNA)) can safely direct the body's cellular machinery to produce nearly any protein of interest, from native proteins to antibodies and other entirely novel protein constructs that can have therapeutic activity inside and outside of cells. The RNA (*e.g.*, mRNA) vaccines of the present disclosure may be used to induce a

balanced immune response against herpes simplex virus (HSV), comprising both cellular and humoral immunity, without risking the possibility of insertional mutagenesis, for example.

The RNA (*e.g.*, mRNA) vaccines may be utilized in various settings depending on the prevalence of the infection or the degree or level of unmet medical need. The RNA vaccines may be utilized to treat and/or prevent a HSV of various genotypes, strains, and isolates. The RNA vaccines have superior properties in that they produce much larger antibody titers and produce responses earlier than commercially available anti-viral therapeutic treatments. While not wishing to be bound by theory, it is believed that the RNA vaccines, as mRNA polynucleotides, are better designed to produce the appropriate protein conformation upon translation as the RNA vaccines co-opt natural cellular machinery. Unlike traditional vaccines which are manufactured *ex vivo* and may trigger unwanted cellular responses, the RNA vaccines are presented to the cellular system in a more native fashion.

Some embodiments of the present disclosure provide herpes simplex virus (HSV) vaccines that include at least one ribonucleic acid (RNA) polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide or an immunogenic fragment thereof (*e.g.*, an immunogenic fragment capable of inducing an immune response to HSV).

Some embodiments of the present disclosure provide herpes simplex virus (HSV) vaccines that include (i) at least one ribonucleic acid (RNA) polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide or an immunogenic fragment thereof (*e.g.*, an immunogenic fragment capable of inducing an immune response to HSV) and (ii) a pharmaceutically-acceptable carrier.

In some embodiments, at least one antigenic polypeptide is HSV (HSV-1 or HSV-2) glycoprotein B, HSV (HSV-1 or HSV-2) glycoprotein C, HSV (HSV-1 or HSV-2) glycoprotein D, HSV (HSV-1 or HSV-2) glycoprotein E, HSV (HSV-1 or HSV-2) glycoprotein I. In some embodiments, at least one antigenic polypeptide has at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identity to HSV (HSV-1 or HSV-2) glycoprotein B, HSV (HSV-1 or HSV-2) glycoprotein C, HSV (HSV-1 or HSV-2) glycoprotein D, HSV (HSV-1 or HSV-2) glycoprotein E, HSV (HSV-1 or HSV-2) glycoprotein I or HSV (HSV-1 or HSV-2) ICP4 protein.

In some embodiments, at least one antigen polypeptide is a non-glycogenic polypeptide, for example, but not limited to, HSV (HSV-1 or HSV-2) ICP4 protein, HSV (HSV-1 or HSV-2) ICP0 protein, or an immunogenic fragment thereof.

In some embodiments, at least one antigenic polypeptide has at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identity to HSV (HSV-1 or HSV-2)

glycoprotein B, HSV (HSV-1 or HSV-2) glycoprotein C, HSV (HSV-1 or HSV-2) glycoprotein D, HSV (HSV-1 or HSV-2) glycoprotein E, HSV (HSV-1 or HSV-2) glycoprotein I or HSV (HSV-1 or HSV-2) ICP4 protein.

In some embodiments, at least one antigenic polypeptide is HSV (HSV-1 or HSV-2) glycoprotein C, HSV (HSV-1 or HSV-2) glycoprotein D, a combination of HSV (HSV-1 or HSV-2) glycoprotein C and HSV (HSV-1 or HSV-2) glycoprotein D, or an immunogenic fragment thereof.

In some embodiments, a HSV vaccine includes at least one RNA polynucleotide having an open reading frame encoding HSV (HSV-1 or HSV-2) glycoprotein D, formulated with aluminum hydroxide and a 3-O-deacylated form of monophosphoryl lipid A (MPL). In some embodiments, the HSV vaccine is formulated for intramuscular injection.

In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide having greater than 90% identity to an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and having membrane fusion activity. In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide having greater than 95% identity to an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and having membrane fusion activity. In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide having greater than 96% identity to an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and having membrane fusion activity. In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide having greater than 97% identity to an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and having membrane fusion activity. In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide having greater than 98% identity to an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and having membrane fusion activity. In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide having greater than 99% identity to an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and having membrane fusion activity. In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide having 95-99% identity to an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and having membrane fusion activity.

In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide having an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and is codon optimized mRNA.

In some embodiments, at least one mRNA polynucleotide encodes an antigenic polypeptide having an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and has less than 80% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide encodes an antigenic polypeptide having an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and has less than 75%, 85% or 95% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide encodes an antigenic polypeptide having an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and has 50-80%, 60- 80%, 40-80%, 30-80%, 70-80%, 75-80% or 78-80% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide encodes an antigenic polypeptide having an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and has 40-85%, 50- 85%, 60-85%, 30-85%, 70-85%, 75-85%, or 80-85% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide encodes an antigenic polypeptide having an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and has 40-90%, 50- 90%, 60-90%, 30-90%, 70-90%, 75-90%, 80-90%, or 85-90% identity to wild-type mRNA sequence.

In some embodiments, at least one RNA polynucleotide is encoded by a nucleic acid having greater than 90% identity to a nucleic acid sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3). In some embodiments, at least one RNA polynucleotide is encoded by a nucleic acid having greater than 95% identity to a nucleic acid sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3). In some embodiments, at least one RNA polynucleotide is encoded by a nucleic acid having greater than 96% identity to a nucleic acid sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3). In some embodiments, at least one RNA polynucleotide is encoded by a nucleic acid having greater than 97% identity to a nucleic acid sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3). In some embodiments, at least one RNA polynucleotide is encoded by a nucleic acid having greater than 98% identity to a nucleic acid sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3). In some embodiments, at least one RNA polynucleotide is encoded by a nucleic acid having greater than 99% identity to a nucleic acid sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3). In some embodiments, at least one RNA polynucleotide is encoded by a nucleic acid having 95-99% identity to a nucleic acid sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3).

In some embodiments, at least one mRNA polynucleotide is encoded by a nucleic acid having a sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3) and has less than 80% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide is encoded by a nucleic acid having a sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3) and has less than 75%, 85% or 95% identity to a wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide is encoded by a nucleic acid having a sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3) and has less than 50-80%, 60- 80%, 40-80%, 30-80%, 70-80%, 75-80% or 78-80% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide is encoded by a nucleic acid having a sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3) and has less than 40-85%, 50- 85%, 60-85%, 30-85%, 70-85%, 75-85%, or 80-85% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide is encoded by a nucleic acid having a sequence of any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, in Table 1 or 3) and has less than 40-90%, 50- 90%, 60-90%, 30-90%, 70-90%, 75-90%, 80-90%, or 85-90% identity to wild-type mRNA sequence.

In some embodiments, at least one RNA polynucleotide comprises a nucleic acid having greater than 90% identity to a nucleic acid sequence of any one of SEQ ID NO: 90-124. In some embodiments, at least one RNA polynucleotide comprises a nucleic acid having greater than 95% identity to a nucleic acid sequence of any one of SEQ ID NO: 90-124. In some embodiments, at least one RNA polynucleotide comprises a nucleic acid having greater than 96% identity to a nucleic acid sequence of any one of SEQ ID NO: 90-124. In some embodiments, at least one RNA polynucleotide comprises a nucleic acid having greater than 97% identity to a nucleic acid sequence of any one of SEQ ID NO: 90-124. In some embodiments, at least one RNA polynucleotide comprises a nucleic acid having greater than 98% identity to a nucleic acid sequence of any one of SEQ ID NO: 90-124. In some embodiments, at least one RNA polynucleotide comprises a nucleic acid having greater than 99% identity to a nucleic acid sequence of any one of SEQ ID NO: 90-124. In some embodiments, at least one RNA polynucleotide comprises a nucleic acid having 95-99% identity to a nucleic acid sequence of any one of SEQ ID NO: 90-124.

In some embodiments, at least one mRNA polynucleotide comprises a nucleic acid having a sequence of any one of SEQ ID NO: 90-124 and has less than 80% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide comprises a nucleic acid having a sequence of any one of SEQ ID NO: 90-124 and has less than 75%, 85% or 95% identity to a wild-type mRNA sequence. In some embodiments, at least one

mRNA polynucleotide comprises a nucleic acid having a sequence of any one of SEQ ID NO: 90-124 and has less than 50-80%, 60- 80%, 40-80%, 30-80%, 70-80%, 75-80% or 78-80% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide comprises a nucleic acid having a sequence of any one of SEQ ID NO: 90-124 and has less than 40-85%, 50- 85%, 60-85%, 30-85%, 70-85%, 75-85%, or 80-85% identity to wild-type mRNA sequence. In some embodiments, at least one mRNA polynucleotide comprises a nucleic acid having a sequence of any one of SEQ ID NO: 90-124 and has less than 40-90%, 50- 90%, 60-90%, 30-90%, 70-90%, 75-90%, 80-90%, or 85-90% identity to wild-type mRNA sequence.

Table 3 provides National Center for Biotechnology Information (NCBI) accession numbers of interest. It should be understood that the phrase "an amino acid sequence of Table 3" refers to an amino acid sequence identified by one or more NCBI accession numbers listed in Table 3. Each of the nucleic acid sequences, amino acid sequences, and variants having greater than 95% identity to each of the nucleic acid sequences and amino acid sequences encompassed by the Accession Numbers of Table 3 are included within the constructs of the present disclosure.

In some embodiments, at least one mRNA polynucleotide encodes an antigenic polypeptide having an amino acid sequence of any one of SEQ ID NO: 24-53 or 66-67 (*e.g.*, in Table 2 or 3) and has greater than 80% identity to wild-type mRNA sequence, but does not include wild-type mRNA sequence.

In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide that attaches to cell receptors.

In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide that causes fusion of viral and cellular membranes.

In some embodiments, at least one RNA polynucleotide encodes an antigenic polypeptide that is responsible for binding of the HSV to a cell being infected.

In some embodiments, the vaccines further comprise an adjuvant.

Some embodiments of the present disclosure provide a herpes simplex virus (HSV) vaccine that includes at least one ribonucleic acid (RNA) polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide.

In some embodiments, the HSV vaccine includes at least one RNA polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide having at least one modification.

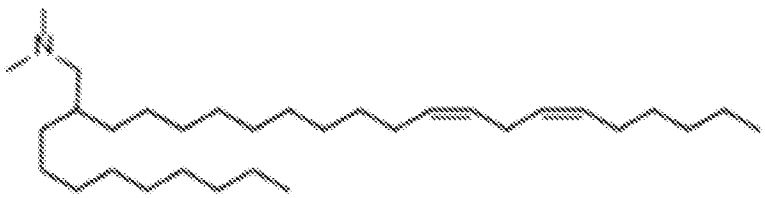
In some embodiments, the HSV vaccine includes at least one RNA polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide having at least one modification, at least one 5′ terminal cap, and is formulated within a lipid nanoparticle.

In some embodiments, a 5' terminal cap is 7mG(5')ppp(5')NlmpNp.

In some embodiments, at least one chemical modification is selected from the group consisting of pseudouridine, N1-methylpseudouridine, N1-ethylpseudouridine, 2-thiouridine, 4'-thiouridine, 5-methylcytosine, 2-thio-1-methyl-1-deaza-pseudouridine, 2-thio-1-methyl-pseudouridine, 2-thio-5-aza-uridine , 2-thio-dihydropseudouridine, 2-thio-dihydrouridine, 2-thio-pseudouridine, 4-methoxy-2-thio-pseudouridine, 4-methoxy-pseudouridine, 4-thio-1-methyl-pseudouridine, 4-thio-pseudouridine, 5-aza-uridine, dihydropseudouridine, 5-methoxyuridine, and 2'-O-methyl uridine.

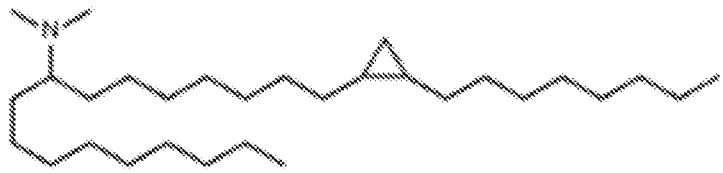
In some embodiments, a lipid nanoparticle comprises a cationic lipid, a PEG-modified lipid, a sterol, and a non-cationic lipid. In some embodiments, a cationic lipid is an ionizable cationic lipid and the non-cationic lipid is a neutral lipid, and the sterol is a cholesterol. In some embodiments, a cationic lipid is selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), (12Z,15Z)-N,N-dimethyl-2-nonylhenicosa-12,15-dien-1-amine (L608), and N,N-dimethyl-1-[(1S,2R)-2-octylcyclopropyl]heptadecan-8-amine (L530).

In some embodiments, the lipid is



(L608).

In some embodiments, the lipid is



(L530).

Some embodiments of the present disclosure provide a herpes simplex virus (HSV) vaccine that includes at least one ribonucleic acid (RNA) polynucleotide having an open

reading frame encoding at least one HSV antigenic polypeptide, wherein at least 80% of the uracil in the open reading frame have a chemical modification, optionally wherein the HSV vaccine is formulated in a lipid nanoparticle.

In some embodiments, 100% of the uracil in the open reading frame have a chemical modification. In some embodiments, a chemical modification is in the 5-position of the uracil. In some embodiments, a chemical modification is a N1-methyl pseudouridine. In some embodiments, 100% of the uracil in the open reading frame have a N1-methyl pseudouridine in the 5-position of the uracil.

Some embodiments of the present disclosure provide methods of inducing an antigen specific immune response in a subject, comprising administering to the subject a HSV vaccine in an amount effective to produce an antigen specific immune response.

In some embodiments, an antigen specific immune response comprises a T cell response or a B cell response.

In some embodiments, a method of producing an antigen specific immune response involves a single administration of the HSV vaccine. In some embodiments, a method further includes administering to the subject a booster dose of the HSV vaccine. A booster vaccine according to this invention may comprise any HSV vaccine disclosed herein.

In some embodiments, a HSV vaccine is administered to the subject by intradermal or intramuscular injection.

Also provided herein are HSV vaccines for use in a method of inducing an antigen specific immune response in a subject, the method comprising administering the HSV vaccine to the subject in an amount effective to produce an antigen specific immune response in the subject.

Further provided herein are uses of HSV vaccines in the manufacture of a medicament for use in a method of inducing an antigen specific immune response in a subject, the method comprising administering the HSV vaccine to the subject in an amount effective to produce an antigen specific immune response.

In some embodiments, an anti-HSV antigenic polypeptide antibody titer produced in the subject is increased by at least 1 log relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased by 1-3 log relative to a control.

In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased at least 2 times relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased at least 5 times

relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased at least 10 times relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased 2-10 times relative to a control.

In some embodiments, the control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has not been administered HSV vaccine. In some embodiments, the control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has been administered a live attenuated or inactivated HSV vaccine. In some embodiments, the control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has been administered a recombinant or purified HSV protein vaccine. In some embodiments, the control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has been administered an HSV virus-like particle (VLP) vaccine.

In some embodiments, the effective amount is a dose equivalent to at least a 2-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount is a dose equivalent to at least a 4-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount is a dose equivalent to at least a 10-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount is a dose equivalent to at least a 100-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the

standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount is a dose equivalent to at least a 1000-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount is a dose equivalent to a 2-fold to 1000-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount is a total dose of 25 μg to 1000 μg, or 50 μg to 1000 μg, or 25 to 200 μg. In some embodiments, the effective amount is a total dose of 100 μg. In some embodiments, the effective amount is a dose of 25 μg administered to the subject a total of two times. In some embodiments, the effective amount is a dose of 100 μg administered to the subject a total of two times. In some embodiments, the effective amount is a dose of 400 μg administered to the subject a total of two times. In some embodiments, the effective amount is a dose of 500 μg administered to the subject a total of two times.

Other aspects of the present disclosure provide methods of inducing an antigen specific immune response in a subject, the method comprising administering to a subject the HSV RNA (*e.g.*, mRNA) vaccine described herein in an effective amount to produce an antigen specific immune response in a subject.

In some embodiments, an antigen specific immune response comprises (an increase in) antigenic polypeptide antibody production. In some embodiments, an anti-HSV antigenic polypeptide antibody titer produced in the subject is increased by at least 1 log relative to a control. In some embodiments, an anti-HSV antigenic polypeptide antibody titer produced in the subject is increased by 1 log to 3 log relative to a control.

In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased at least 2 times relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased at least 5 times

relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased at least 10 times relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased 2 times to 10 times relative to a control.

In some embodiments, the control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has not been administered HSV vaccine. In some embodiments, the control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has been administered a live attenuated or inactivated HSV vaccine. In some embodiments, the control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has been administered a recombinant or purified HSV protein vaccine. In some embodiments, the control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has been administered a HSV VLP vaccine.

In some embodiments, the effective amount administered to a subject is a dose (of HSV RNA, *e.g.*, mRNA, vaccine) equivalent to at least a 2-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant HSV protein vaccine, a live attenuated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount administered to a subject is a dose (of HSV RNA, *e.g.*, mRNA, vaccine) equivalent to at least a 4-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount administered to a subject is a dose (of HSV RNA, *e.g.*, mRNA, vaccine) equivalent to at least a 10-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, and wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount is a dose (of HSV RNA, *e.g.*, mRNA, vaccine) administered to a subject equivalent to at least a 100-fold reduction in the standard

of care dose of a recombinant HSV protein vaccine, wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount administered to a subject is a dose (of HSV RNA, *e.g.*, mRNA, vaccine) equivalent to at least a 1000-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, and wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount administered to a subject is a dose (of HSV RNA, *e.g.*, mRNA, vaccine) equivalent to a 2-fold to 1000-fold reduction in the standard of care dose of a recombinant HSV protein vaccine, and wherein an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount administered to a subject is a total dose (of HSV RNA, *e.g.*, mRNA, vaccine) of 50 μg to 1000 μg. In some embodiments, the effective amount is a total dose of 50 μg, 100 μg, 200 μg, 400 μg, 800 μg, or 1000 μg. In some embodiments, the effective amount is a dose of 25 μg administered to the subject a total of two times. In some embodiments, the effective amount is a dose of 50 μg administered to the subject a total of two times. In some embodiments, the effective amount is a dose of 100 μg administered to the subject a total of two times. In some embodiments, the effective amount is a dose of 200 μg administered to the subject a total of two times. In some embodiments, the effective amount is a dose of 400 μg administered to the subject a total of two times. In some embodiments, the effective amount is a dose of 500 μg administered to the subject a total of two times.

In some embodiments, the efficacy (or effectiveness) of the HSV RNA (*e.g.*, mRNA) vaccine against HSV is greater than 60%.

Vaccine efficacy may be assessed using standard analyses (*see*, *e.g.*, Weinberg *et al.*, *J Infect Dis*. 2010 Jun 1;201(11):1607-10). For example, vaccine efficacy may be measured

by double-blind, randomized, clinical controlled trials. Vaccine efficacy may be expressed as a proportionate reduction in disease attack rate (AR) between the unvaccinated (ARU) and vaccinated (ARV) study cohorts and can be calculated from the relative risk (RR) of disease among the vaccinated group with use of the following formulas:

Efficacy = (ARU – ARV)/ARU x 100; and

Efficacy = (1-RR) x 100.

Likewise, vaccine effectiveness may be assessed using standard analyses (*see, e.g.*, Weinberg *et al., J Infect Dis.* 2010 Jun 1;201(11):1607-10). Vaccine effectiveness is an assessment of how a vaccine (which may have already proven to have high vaccine efficacy) reduces disease in a population. This measure can assess the net balance of benefits and adverse effects of a vaccination program, not just the vaccine itself, under natural field conditions rather than in a controlled clinical trial. Vaccine effectiveness is proportional to vaccine efficacy (potency) but is also affected by how well target groups in the population are immunized, as well as by other non-vaccine-related factors that influence the 'real-world' outcomes of hospitalizations, ambulatory visits, or costs. For example, a retrospective case control analysis may be used, in which the rates of vaccination among a set of infected cases and appropriate controls are compared. Vaccine effectiveness may be expressed as a rate difference, with use of the odds ratio (OR) for developing infection despite vaccination:

Effectiveness = (1 – OR) x 100.

In some embodiments, the efficacy (or effectiveness) of the HSV RNA (*e.g.*, mRNA) vaccine against HSV is greater than 65%. In some embodiments, the efficacy (or effectiveness) of the vaccine against HSV is greater than 70%. In some embodiments, the efficacy (or effectiveness) of the vaccine against HSV is greater than 75%. In some embodiments, the efficacy (or effectiveness) of the vaccine against HSV is greater than 80%. In some embodiments, the efficacy (or effectiveness) of the vaccine against HSV is greater than 85%. In some embodiments, the efficacy (or effectiveness) of the vaccine against HSV is greater than 90%.

In some embodiments, the vaccine immunizes the subject against HSV up to 1 year (*e.g.* for a single HSV season). In some embodiments, the vaccine immunizes the subject against HSV for up to 2 years. In some embodiments, the vaccine immunizes the subject against HSV for more than 2 years. In some embodiments, the vaccine immunizes the subject against HSV for more than 3 years. In some embodiments, the vaccine immunizes the subject against HSV for more than 4 years. In some embodiments, the vaccine immunizes the subject against HSV for 5-10 years.

In some embodiments, the subject has been exposed to HSV, is infected with (has) HSV, or is at risk of infection by HSV.

In some embodiments, the subject is immunocompromised (has an impaired immune system, *e.g.*, has an immune disorder or autoimmune disorder).

In some embodiments, the subject is a subject about 10 years old, about 20 years old, or older (*e.g.*, about 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, or 20 years old).

In some embodiments, the subject is an adult between the ages of about 20 years and about 50 years (*e.g.*, about 20, 25, 30, 35, 40, 45 or 50 years old).

Some aspects of the present disclosure provide herpes simplex virus (HSV) RNA (*e.g.*, mRNA) vaccines containing a signal peptide linked to a HSV antigenic polypeptide. Thus, in some embodiments, the HSV RNA (*e.g.*, mRNA) vaccines contain at least one ribonucleic acid (RNA) polynucleotide having an open reading frame encoding a signal peptide linked to a HSV antigenic peptide. Also provided herein are nucleic acids encoding the HSV RNA (*e.g.*, mRNA) vaccines disclosed herein.

In some embodiments, the signal peptide is a IgE signal peptide. In some embodiments, the signal peptide is an IgE HC (Ig heavy chain epsilon-1) signal peptide. In some embodiments, the signal peptide has the sequence MDWTWILFLVAAATRVHS (SEQ ID NO: 78). In some embodiments, the signal peptide is an IgGκ signal peptide. In some embodiments, the signal peptide has the sequence METPAQLLFLLLLWLPDTTG (SEQ ID NO: 79). In some embodiments, the signal peptide is selected from: a Japanese encephalitis PRM signal sequence (MLGSNSGQRVVFTILLLLVAPAYS; SEQ ID NO: 80), VSVg protein signal sequence (MKCLLYLAFLFIGVNCA; SEQ ID NO: 81), and Japanese encephalitis JEV signal sequence (MWLVSLAIVTACAGA; SEQ ID NO: 82).

In some embodiments, an effective amount of an HSV RNA (*e.g.*, mRNA) vaccine (*e.g.*, a single dose of the HSV vaccine) results in a 2-fold to 200-fold (*e.g.*, about 2-, 3-, 4-, 5-, 6-, 7-, 8-, 9-, 10-, 20-, 30-, 40-, 50-, 60-, 70-, 80-, 90-, 100-, 110-, 120-, 130-, 140-, 150-, 160-, 170-, 180-, 190- or 200-fold) increase in serum neutralizing antibodies against HSV, relative to a control. In some embodiments, a single dose of the HSV RNA (*e.g.*, mRNA) vaccine results in an about 5-fold, 50-fold, or 150-fold increase in serum neutralizing antibodies against HSV, relative to a control. In some embodiments, a single dose of the HSV RNA (*e.g.*, mRNA) vaccine results in an about 2-fold to 10 fold, or an about 40 to 60 fold increase in serum neutralizing antibodies against HSV, relative to a control.

In some embodiments, the serum neutralizing antibodies are against HSV A and/or HSV B.

In some embodiments, the HSV vaccine is formulated in a MC3 lipid nanoparticle or a L-608 lipid nanoparticle.

In some embodiments, the methods further comprise administering a booster dose of the HSV RNA (*e.g.*, mRNA) vaccine. In some embodiments, the methods further comprise administering a second booster dose of the HSV vaccine.

In some embodiments, efficacy of RNA vaccines RNA (*e.g.*, mRNA) can be significantly enhanced when combined with a flagellin adjuvant, in particular, when one or more antigen-encoding mRNAs is combined with an mRNA encoding flagellin.

RNA (*e.g.*, mRNA) vaccines combined with the flagellin adjuvant (*e.g.*, mRNA-encoded flagellin adjuvant) have superior properties in that they may produce much larger antibody titers and produce responses earlier than commercially available vaccine formulations. While not wishing to be bound by theory, it is believed that the RNA vaccines, for example, as mRNA polynucleotides, are better designed to produce the appropriate protein conformation upon translation, for both the antigen and the adjuvant, as the RNA (*e.g.*, mRNA) vaccines co-opt natural cellular machinery. Unlike traditional vaccines, which are manufactured *ex vivo* and may trigger unwanted cellular responses, RNA (*e.g.*, mRNA) vaccines are presented to the cellular system in a more native fashion.

Some embodiments of the present disclosure provide RNA (*e.g.*, mRNA) vaccines that include at least one RNA (*e.g.*, mRNA) polynucleotide having an open reading frame encoding at least one antigenic polypeptide or an immunogenic fragment thereof (*e.g.*, an immunogenic fragment capable of inducing an immune response to the antigenic polypeptide) and at least one RNA (*e.g.*, mRNA polynucleotide) having an open reading frame encoding a flagellin adjuvant.

In some embodiments, at least one flagellin polypeptide (*e.g.*, encoded flagellin polypeptide) is a flagellin protein. In some embodiments, at least one flagellin polypeptide (*e.g.*, encoded flagellin polypeptide) is an immunogenic flagellin fragment. In some embodiments, at least one flagellin polypeptide and at least one antigenic polypeptide are encoded by a single RNA (*e.g.*, mRNA) polynucleotide. In other embodiments, at least one flagellin polypeptide and at least one antigenic polypeptide are each encoded by a different RNA polynucleotide.

In some embodiments, at least one flagellin polypeptide has at least 80%, at least 85%, at least 90%, or at least 95% identity to a flagellin polypeptide having a sequence of SEQ ID NO: 89, 125, or 126.

In some embodiments the nucleic acid vaccines described herein are chemically modified. In other embodiments the nucleic acid vaccines are unmodified.

Yet other aspects provide compositions for and methods of vaccinating a subject comprising administering to the subject a nucleic acid vaccine comprising one or more RNA polynucleotides having an open reading frame encoding a first virus antigenic polypeptide, wherein the RNA polynucleotide does not include a stabilization element, and wherein an adjuvant is not coformulated or co-administered with the vaccine.

In other aspects the invention is a composition for or method of vaccinating a subject comprising administering to the subject a nucleic acid vaccine comprising one or more RNA polynucleotides having an open reading frame encoding a first antigenic polypeptide wherein a dosage of between 10 μg/kg and 400 μg/kg of the nucleic acid vaccine is administered to the subject. In some embodiments the dosage of the RNA polynucleotide is 1-5 μg, 5-10 μg, 10-15 μg, 15-20 μg, 10-25 μg, 20-25 μg, 20-50 μg, 30-50 μg, 40-50 μg, 40-60 μg, 60-80 μg, 60-100 μg, 50-100 μg, 80-120 μg, 40-120 μg, 40-150 μg, 50-150 μg, 50-200 μg, 80-200 μg, 100-200 μg, 120-250 μg, 150-250 μg, 180-280 μg, 200-300 μg, 50-300 μg, 80-300 μg, 100-300 μg, 40-300 μg, 50-350 μg, 100-350 μg, 200-350 μg, 300-350 μg, 320-400 μg, 40-380 μg, 40-100 μg, 100-400 μg, 200-400 μg, or 300-400 μg per dose. In some embodiments, the nucleic acid vaccine is administered to the subject by intradermal or intramuscular injection. In some embodiments, the nucleic acid vaccine is administered to the subject on day zero. In some embodiments, a second dose of the nucleic acid vaccine is administered to the subject on day twenty one.

In some embodiments, a dosage of 25 micrograms of the RNA polynucleotide is included in the nucleic acid vaccine administered to the subject. In some embodiments, a dosage of 100 micrograms of the RNA polynucleotide is included in the nucleic acid vaccine administered to the subject. In some embodiments, a dosage of 50 micrograms of the RNA polynucleotide is included in the nucleic acid vaccine administered to the subject. In some embodiments, a dosage of 75 micrograms of the RNA polynucleotide is included in the nucleic acid vaccine administered to the subject. In some embodiments, a dosage of 150 micrograms of the RNA polynucleotide is included in the nucleic acid vaccine administered to the subject. In some embodiments, a dosage of 400 micrograms of the RNA polynucleotide is included in the nucleic acid vaccine administered to the subject. In some embodiments, a dosage of 200 micrograms of the RNA polynucleotide is included in the nucleic acid vaccine administered to the subject. In some embodiments, the RNA polynucleotide accumulates at a 100 fold higher level in the local lymph node in comparison with the distal lymph node. In

other embodiments the nucleic acid vaccine is chemically modified and in other embodiments the nucleic acid vaccine is not chemically modified.

Aspects of the invention provide a nucleic acid vaccine comprising one or more RNA polynucleotides having an open reading frame encoding a first antigenic polypeptide, wherein the RNA polynucleotide does not include a stabilization element, and a pharmaceutically acceptable carrier or excipient, wherein an adjuvant is not included in the vaccine. In some embodiments, the stabilization element is a histone stem-loop. In some embodiments, the stabilization element is a nucleic acid sequence having increased GC content relative to wild type sequence.

Aspects of the invention provide nucleic acid vaccines comprising one or more RNA polynucleotides having an open reading frame encoding a first antigenic polypeptide, wherein the RNA polynucleotide is present in the formulation for in vivo administration to a host, which confers an antibody titer superior to the criterion for seroprotection for the first antigen for an acceptable percentage of human subjects. In some embodiments, the antibody titer produced by the mRNA vaccines of the invention is a neutralizing antibody titer. In some embodiments the neutralizing antibody titer is greater than a protein vaccine. In other embodiments the neutralizing antibody titer produced by the mRNA vaccines of the invention is greater than an adjuvanted protein vaccine. In yet other embodiments the neutralizing antibody titer produced by the mRNA vaccines of the invention is 1,000-10,000, 1,200-10,000, 1,400-10,000, 1,500-10,000, 1,000-5,000, 1,000-4,000, 1,800-10,000, 2000-10,000, 2,000-5,000, 2,000-3,000, 2,000-4,000, 3,000-5,000, 3,000-4,000, or 2,000-2,500. A neutralization titer is typically expressed as the highest serum dilution required to achieve a 50% reduction in the number of plaques.

Also provided are nucleic acid vaccines comprising one or more RNA polynucleotides having an open reading frame encoding a first antigenic polypeptide, wherein the RNA polynucleotide is present in a formulation for in vivo administration to a host for eliciting a longer lasting high antibody titer than an antibody titer elicited by an mRNA vaccine having a stabilizing element or formulated with an adjuvant and encoding the first antigenic polypeptide. In some embodiments, the RNA polynucleotide is formulated to produce a neutralizing antibodies within one week of a single administration. In some embodiments, the adjuvant is selected from a cationic peptide and an immunostimulatory nucleic acid. In some embodiments, the cationic peptide is protamine.

Aspects provide nucleic acid vaccines comprising one or more RNA polynucleotides having an open reading frame comprising at least one chemical modification or optionally no

chemical modification, the open reading frame encoding a first antigenic polypeptide, wherein the RNA polynucleotide is present in the formulation for in vivo administration to a host such that the level of antigen expression in the host significantly exceeds a level of antigen expression produced by an mRNA vaccine having a stabilizing element or formulated with an adjuvant and encoding the first antigenic polypeptide.

Other aspects provide nucleic acid vaccines comprising one or more RNA polynucleotides having an open reading frame comprising at least one chemical modification or optionally no chemical modification, the open reading frame encoding a first antigenic polypeptide, wherein the vaccine has at least 10 fold less RNA polynucleotide than is required for an unmodified mRNA vaccine to produce an equivalent antibody titer. In some embodiments, the RNA polynucleotide is present in a dosage of 25-100 micrograms.

Aspects of the invention also provide a unit of use vaccine, comprising between 10ug and 400 ug of one or more RNA polynucleotides having an open reading frame comprising at least one chemical modification or optionally no chemical modification, the open reading frame encoding a first antigenic polypeptide, and a pharmaceutically acceptable carrier or excipient, formulated for delivery to a human subject. In some embodiments, the vaccine further comprises a cationic lipid nanoparticle.

Aspects of the invention provide methods of creating, maintaining or restoring antigenic memory to a virus strain in an individual or population of individuals comprising administering to said individual or population an antigenic memory booster nucleic acid vaccine comprising (a) at least one RNA polynucleotide, said polynucleotide comprising at least one chemical modification or optionally no chemical modification and two or more codon-optimized open reading frames, said open reading frames encoding a set of reference antigenic polypeptides, and (b) optionally a pharmaceutically acceptable carrier or excipient. In some embodiments, the vaccine is administered to the individual via a route selected from the group consisting of intramuscular administration, intradermal administration and subcutaneous administration. In some embodiments, the administering step comprises contacting a muscle tissue of the subject with a device suitable for injection of the composition. In some embodiments, the administering step comprises contacting a muscle tissue of the subject with a device suitable for injection of the composition in combination with electroporation.

Aspects of the invention provide methods of vaccinating a subject comprising administering to the subject a single dosage of between 25 ug/kg and 400 ug/kg of a nucleic

acid vaccine comprising one or more RNA polynucleotides having an open reading frame encoding a first antigenic polypeptide in an effective amount to vaccinate the subject.

Other aspects provide nucleic acid vaccines comprising one or more RNA polynucleotides having an open reading frame comprising at least one chemical modification, the open reading frame encoding a first antigenic polypeptide, wherein the vaccine has at least 10 fold less RNA polynucleotide than is required for an unmodified mRNA vaccine to produce an equivalent antibody titer. In some embodiments, the RNA polynucleotide is present in a dosage of 25-100 micrograms.

Other aspects provide nucleic acid vaccines comprising an LNP formulated RNA polynucleotide having an open reading frame comprising no modified nucleotides (unmodified), the open reading frame encoding a first antigenic polypeptide, wherein the vaccine has at least 10 fold less RNA polynucleotide than is required for an unmodified mRNA vaccine not formulated in a LNP to produce an equivalent antibody titer. In some embodiments, the RNA polynucleotide is present in a dosage of 25-100 micrograms.

The data presented in the Examples demonstrate significant enhanced immune responses using the formulations of the invention. Both chemically modified and unmodified RNA vaccines are useful in the invention. Surprisingly, in contrast to prior art reports that it was preferable to use chemically unmodified mRNA formulated in a carrier for the production of vaccines, it is described herein that chemically modified mRNA-LNP vaccines required a much lower effective mRNA dose than unmodified mRNA, i.e., tenfold less than unmodified mRNA when formulated in carriers other than LNP. Both the chemically modified and unmodified RNA vaccines of the invention produce better immune responses than mRNA vaccines formulated in a different lipid carrier.

In other aspects the invention encompasses a method of treating an elderly subject age 60 years or older comprising administering to the subject a nucleic acid vaccine comprising one or more RNA polynucleotides having an open reading frame encoding an virus antigenic polypeptide in an effective amount to vaccinate the subject.

In other aspects the invention encompasses a method of treating a young subject age 17 years or younger comprising administering to the subject a nucleic acid vaccine comprising one or more RNA polynucleotides having an open reading frame encoding an virus antigenic polypeptide in an effective amount to vaccinate the subject.

In other aspects the invention encompasses a method of treating an adult subject comprising administering to the subject a nucleic acid vaccine comprising one or more RNA

polynucleotides having an open reading frame encoding a virus antigenic polypeptide in an effective amount to vaccinate the subject.

In some aspects the invention is a method of vaccinating a subject with a combination vaccine including at least two nucleic acid sequences encoding antigens wherein the dosage for the vaccine is a combined therapeutic dosage wherein the dosage of each individual nucleic acid encoding an antigen is a sub therapeutic dosage. In some embodiments, the combined dosage is 25 micrograms of the RNA polynucleotide in the nucleic acid vaccine administered to the subject. In some embodiments, the combined dosage is 100 micrograms of the RNA polynucleotide in the nucleic acid vaccine administered to the subject. In some embodiments the combined dosage is 50 micrograms of the RNA polynucleotide in the nucleic acid vaccine administered to the subject. In some embodiments, the combined dosage is 75 micrograms of the RNA polynucleotide in the nucleic acid vaccine administered to the subject. In some embodiments, the combined dosage is 150 micrograms of the RNA polynucleotide in the nucleic acid vaccine administered to the subject. In some embodiments, the combined dosage is 400 micrograms of the RNA polynucleotide in the nucleic acid vaccine administered to the subject. In some embodiments, the sub therapeutic dosage of each individual nucleic acid encoding an antigen is 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, or 20 micrograms. In other embodiments the nucleic acid vaccine is chemically modified and in other embodiments the nucleic acid vaccine is not chemically modified.

The RNA polynucleotide is one of SEQ ID NO: 1-23, 54-64, and 90-124 and includes at least one chemical modification. In other embodiments the RNA polynucleotide is one of SEQ ID NO: 1-23, 54-64, and 90-124 and does not include any nucleotide modifications, or is unmodified. In yet other embodiments the at least one RNA polynucleotide encodes an antigenic protein of any of SEQ ID NO: 24-53 and 66-67 and includes at least one chemical modification. In other embodiments the RNA polynucleotide encodes an antigenic protein of any of SEQ ID NO: 24-53 and 66-67 and does not include any nucleotide modifications, or is unmodified.

In preferred aspects, vaccines of the invention (e.g., LNP-encapsulated mRNA vaccines) produce prophylactically- and/or therapeutically- efficacious levels, concentrations and/or titers of antigen-specific antibodies in the blood or serum of a vaccinated subject. As defined herein, the term antibody titer refers to the amount of antigen-specific antibody produces in s subject, e.g., a human subject. In exemplary embodiments, antibody titer is expressed as the inverse of the greatest dilution (in a serial dilution) that still gives a positive

result. In exemplary embodiments, antibody titer is determined or measured by enzyme-linked immunosorbent assay (ELISA). In exemplary embodiments, antibody titer is determined or measured by neutralization assay, e.g., by microneutralization assay. In certain aspects, antibody titer measurement is expressed as a ratio, such as 1:40, 1:100, etc.

In exemplary embodiments of the invention, an efficacious vaccine produces an antibody titer of greater than 1:40, greater that 1:100, greater than 1:400, greater than 1:1000, greater than 1:2000, greater than 1:3000, greater than 1:4000, greater than 1:500, greater than 1:6000, greater than 1:7500, greater than 1:10000. In exemplary embodiments, the antibody titer is produced or reached by 10 days following vaccination, by 20 days following vaccination, by 30 days following vaccination, by 40 days following vaccination, or by 50 or more days following vaccination. In exemplary embodiments, the titer is produced or reached following a single dose of vaccine administered to the subject. In other embodiments, the titer is produced or reached following multiple doses, e.g., following a first and a second dose (e.g., a booster dose.)

In exemplary aspects of the invention, antigen-specific antibodies are measured in units of μg/ml or are measured in units of IU/L (International Units per liter) or mIU/ml (milli International Units per ml). In exemplary embodiments of the invention, an efficacious vaccine produces >0.5 μg/ml, >0.1 μg/ml, >0.2 μg/ml, >0.35 μg/ml, >0.5 μg/ml, >1 μg/ml, >2 μg/ml, >5 μg/ml or >10 μg/ml. In exemplary embodiments of the invention, an efficacious vaccine produces >10 mIU/ml, >20 mIU/ml, >50 mIU/ml, >100 mIU/ml, >200 mIU/ml, >500 mIU/ml or > 1000 mIU/ml. In exemplary embodiments, the antibody level or concentration is produced or reached by 10 days following vaccination, by 20 days following vaccination, by 30 days following vaccination, by 40 days following vaccination, or by 50 or more days following vaccination. In exemplary embodiments, the level or concentration is produced or reached following a single dose of vaccine administered to the subject. In other embodiments, the level or concentration is produced or reached following multiple doses, e.g., following a first and a second dose (e.g., a booster dose.) In exemplary embodiments, antibody level or concentration is determined or measured by enzyme-linked immunosorbent assay (ELISA). In exemplary embodiments, antibody level or concentration is determined or measured by neutralization assay, e.g., by microneutralization assay.

The details of various embodiments of the invention are set forth in the description below. Other features, objects, and advantages of the invention will be apparent from the description and the drawings, and from the claims.

DETAILED DESCRIPTION

Embodiments of the present disclosure provide RNA (*e.g.*, mRNA) vaccines that include polynucleotide encoding a herpes simplex virus (HSV) antigen. HSV is a double-stranded, linear DNA virus in the *Herpesviridae*. Two members of the herpes simplex virus family infect humans – known as HSV-1 and HSV-2. Symptoms of HSV infection include the formation of blisters in the skin or mucous membranes of the mouth, lips and/or genitals. HSV is a neuroinvasive virus that can cause sporadic recurring episodes of viral reactivation in infected individuals. HSV is transmitted by contact with an infected area of the skin during a period of viral activation. HSV most commonly infects via the oral or genital mucosa and replicates in the stratified squamous epithelium, followed by uptake into ramifying unmyelinated sensory nerve fibers within the stratified squamous epithelium. The virus is then transported to the cell body of the neuron in the dorsal root ganglion, where it persists in a latent cellular infection (Cunningham AL *et al. J Infect Dis.* (2006) 194 (Supplement 1): S11-S18).

The genome of Herpes Simplex Viruses (HSV-1 and HSV-2) contains about 85 open reading frames, such that HSV can generate at least 85 unique proteins. These genes encode 4 major classes of proteins: (1) those associated with the outermost external lipid bilayer of HSV (the envelope), (2) the internal protein coat (the capsid), (3) an intermediate complex connecting the envelope with the capsid coat (the tegument), and (4) proteins responsible for replication and infection.

Examples of envelope proteins include UL1 (gL), UL10 (gM), UL20, UL22, UL27 (gB), UL43, UL44 (gC), UL45, UL49A, UL53 (gK), US4 (gG), US5 (gJ), US6 (gD), US7 (gI), US8 (gE), and US10. Examples of capsid proteins include UL6, UL18, UL19, UL35, and UL38. Tegument proteins include UL11, UL13, UL21, UL36, UL37, UL41, UL45, UL46, UL47, UL48, UL49, US9, and US10. Other HSV proteins include UL2, UL3, UL4, UL5, UL7, UL8, UL9, UL12, UL14, UL15, UL16, UL17, UL23, UL24, UL25, UL26, UL26.5, UL28, UL29, UL30, UL31, UL32, UL33, UL34, UL39, UL40, UL42, UL50, UL51, UL52, UL54, UL55, UL56, US1, US2, US3, US81, US11, US12, ICP0, and ICP4.

Since the envelope (most external portion of an HSV particle) is the first to encounter target cells, the present disclosure encompasses antigenic polypeptides associated with the envelope as immunogenic agents. In brief, surface and membrane proteins--glycoprotein D (gD), glycoprotein B (gB), glycoprotein H (gH), glycoprotein L (gL)--as single antigens or in combination with or without adjuvants may be used as HSV vaccine antigens.

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding HSV (HSV-1 or HSV-2) glycoprotein D.

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding HSV (HSV-1 or HSV-2) glycoprotein B.

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding HSV (HSV-1 or HSV-2) glycoprotein D and glycoprotein C.

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding HSV (HSV-1 or HSV-2) glycoprotein D and glycoprotein E (or glycoprotein I).

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding HSV (HSV-1 or HSV-2) glycoprotein B and glycoprotein C.

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding HSV (HSV-1 or HSV-2) glycoprotein B and glycoprotein E (or glycoprotein I).

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding a HSV (HSV-1 or HSV-2) antigenic polypeptide having at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identity with HSV (HSV-1 or HSV-2) glycoprotein D and has HSV (HSV-1 or HSV-2) glycoprotein D activity.

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding a HSV (HSV-1 or HSV-2) antigenic polypeptide having at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identity with HSV (HSV-1 or HSV-2) glycoprotein C and has HSV (HSV-1 or HSV-2) glycoprotein C activity.

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding a HSV (HSV-1 or HSV-2) antigenic polypeptide having at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identity with HSV (HSV-1 or HSV-2) glycoprotein B and has HSV (HSV-1 or HSV-2) glycoprotein B activity.

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding a HSV (HSV-1 or HSV-2) antigenic polypeptide having at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identity with HSV (HSV-1 or HSV-2) glycoprotein E and has HSV (HSV-1 or HSV-2) glycoprotein E activity.

In some embodiments, HSV vaccines comprise RNA (*e.g.*, mRNA) encoding a HSV (HSV-1 or HSV-2) antigenic polypeptide having at least 95%, at least 96%, at least 97%, at least 98%, or at least 99% identity with HSV (HSV-1 or HSV-2) glycoprotein I and has HSV (HSV-1 or HSV-2) glycoprotein I activity.

Glycoprotein "activity" of the present disclosure is described below.

Glycoprotein C (gC) is a glycoprotein involved in viral attachment to host cells; *e.g.*, it acts as an attachment protein that mediates binding of the HSV-2 virus to host adhesion receptors, namely cell surface heparan sulfate and/or chondroitin sulfate. gC plays a role in host immune evasion (aka viral immunoevasion) by inhibiting the host complement cascade activation. In particular, gC binds to and/or interacts with host complement component C3b; this interaction then inhibits the host immune response by disregulating the complement cascade (*e.g.*, binds host complement C3b to block neutralization of virus).

Glycoprotein D (gD) is an envelope glycoprotein that binds to cell surface receptors and/or is involved in cell attachment via poliovirus receptor-related protein and/or herpesvirus entry mediator, facilitating virus entry. gD binds to the potential host cell entry receptors (tumor necrosis factor receptor superfamily, member 14(TNFRSF14)/herpesvirus entry mediator (HVEM), poliovirus receptor-related protein 1 (PVRL1) and or poliovirus receptor-related protein 2 (PVRL2), and is proposed to trigger fusion with host membrane by recruiting the fusion machinery composed of, for example, gB and gH/gL. gD interacts with host cell receptors TNFRSF14 and/or PVRL1 and/or PVRL2 and (1) interacts (via profusion domain) with gB; an interaction which can occur in the absence of related HSV glycoproteins, *e.g.*, gH and/or gL; and (2) gD interacts (via profusion domain) with gH/gL heterodimer, an interaction which can occur in the absence of gB. As such, gD associates with the gB-gH/gL-gD complex. gD also interacts (via C-terminus) with UL11 tegument protein.

Glycoprotein B (gB) is a viral glycoprotein involved in the viral cell activity of herpes simplex virus (HSV) and is required for the fusion of the HSV's envelope with the cellular membrane. It is the most highly conserved of all surface glycoproteins and primarily acts as a fusion protein, constituting the core fusion machinery. gB, a class III membrane fusion glycoprotein, is a type-1 transmembrane protein trimer of five structural domains. Domain I includes two internal fusion loops and is thought to insert into the cellular membrane during virus-cell fusion. Domain II appears to interact with gH/gL during the fusion process, domain III contains an elongated alpha helix, and domain IV interacts with cellular receptors.

In epithelial cells, the heterodimer glycoprotein E/glycoproteinI (gE/gI) is required for the cell-to-cell spread of the virus, by sorting nascent virions to cell junctions. Once the virus reaches the cell junctions, virus particles can spread to adjacent cells extremely rapidly through interactions with cellular receptors that accumulate at these junctions. By similarity, it is implicated in basolateral spread in polarized cells. In neuronal cells, gE/gI is essential for the anterograde spread of the infection throughout the host nervous system. Together with

US9, the heterodimer gE/gI is involved in the sorting and transport of viral structural components toward axon tips. The heterodimer gE/gI serves as a receptor for the Fc part of host IgG. Dissociation of gE/gI from IgG occurs at acidic pH, thus may be involved in anti-HSV antibodies bipolar bridging, followed by intracellular endocytosis and degradation, thereby interfering with host IgG-mediated immune responses. gE/gI interacts (via C-terminus) with VP22 tegument protein; this interaction is necessary for the recruitment of VP22 to the Golgi and its packaging into virions.

In any of the embodiments described herein, the RNA may have at least one modification, including at least one chemical modification.

HSV RNA (*e.g.*, mRNA) vaccines, as provided herein may be used to induce a balanced immune response, comprising both cellular and humoral immunity, without many of the risks associated with DNA vaccination.

The entire contents of International Application No. PCT/US2015/02740 are incorporated herein by reference.

It has been discovered that the mRNA vaccines described herein are superior to current vaccines in several ways. First, the lipid nanoparticle (LNP) delivery is superior to other formulations including a protamine base approach described in the literature and no additional adjuvants are to be necessary. The use of LNPs enables the effective delivery of chemically modified or unmodified mRNA vaccines. Additionally it has been demonstrated herein that both modified and unmodified LNP formulated mRNA vaccines were superior to conventional vaccines by a significant degree. In some embodiments the mRNA vaccines of the invention are superior to conventional vaccines by a factor of at least 10 fold, 20 fold, 40 fold, 50 fold, 100 fold, 500 fold or 1,000 fold.

Although attempts have been made to produce functional RNA vaccines, including mRNA vaccines and self-replicating RNA vaccines, the therapeutic efficacy of these RNA vaccines have not yet been fully established. Quite surprisingly, the inventors have discovered, according to aspects of the invention a class of formulations for delivering mRNA vaccines in vivo that results in significantly enhanced, and in many respects synergistic, immune responses including enhanced antigen generation and functional antibody production with neutralization capability. These results can be achieved even when significantly lower doses of the mRNA are administered in comparison with mRNA doses used in other classes of lipid based formulations. The formulations of the invention have demonstrated significant unexpected in vivo immune responses sufficient to establish the efficacy of functional mRNA vaccines as prophylactic and therapeutic agents. Additionally,

self-replicating RNA vaccines rely on viral replication pathways to deliver enough RNA to a cell to produce an immunogenic response. The formulations of the invention do not require viral replication to produce enough protein to result in a strong immune response. Thus, the mRNA of the invention are not self-replicating RNA and do not include components necessary for viral replication.

The invention involves, in some aspects, the surprising finding that lipid nanoparticle (LNP) formulations significantly enhance the effectiveness of mRNA vaccines, including chemically modified and unmodified mRNA vaccines. The efficacy of mRNA vaccines formulated in LNP was examined in vivo using several distinct antigens. The results presented herein demonstrate the unexpected superior efficacy of the mRNA vaccines formulated in LNP over other commercially available vaccines.

In addition to providing an enhanced immune response, the formulations of the invention generate a more rapid immune response with fewer doses of antigen than other vaccines tested. The mRNA-LNP formulations of the invention also produce quantitatively and qualitatively better immune responses than vaccines formulated in a different carriers.

The LNP used in the studies described herein has been used previously to deliver siRNA in various animal models as well as in humans. In view of the observations made in association with the siRNA delivery of LNP formulations, the fact that LNP is useful in vaccines is quite surprising. It has been observed that therapeutic delivery of siRNA formulated in LNP causes an undesirable inflammatory response associated with a transient IgM response, typically leading to a reduction in antigen production and a compromised immune response. In contrast to the findings observed with siRNA, the LNP-mRNA formulations of the invention are demonstrated herein to generate enhanced IgG levels, sufficient for prophylactic and therapeutic methods rather than transient IgM responses.

Nucleic Acids/Polynucleotides

HSV vaccines, as provided herein, comprise at least one (one or more) ribonucleic acid (RNA) polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide. The term "nucleic acid," in its broadest sense, includes any compound and/or substance that comprises a polymer of nucleotides. These polymers are referred to as polynucleotides.

In some embodiments, at least one RNA polynucleotide is encoded by at least one nucleic acid sequence selected from any of SEQ ID NO: 1-23, 54-64, or homologs having at least 80% identity with a nucleic acid sequence selected from any one of SEQ ID NO: 1-23

or 54-64. In some embodiments, at least one RNA polynucleotide is encoded by at least one nucleic acid sequence selected from any one of SEQ ID NO: 1-23, 54-64 or homologs having at least 90% (*e.g.* 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, 99.8%, or 99.9%) identity with a nucleic acid sequence selected from any one of SEQ ID NO: 1-23 or 54-64. In some embodiments, at least one RNA polynucleotide is encoded by at least one fragment of a nucleic acid sequence selected from any one of SEQ ID NO: 1-23 or 54-64. In some embodiments, the at least one RNA polynucleotide has at least one chemical modification.

Nucleic acids (also referred to as polynucleotides) may be or may include, for example, ribonucleic acids (RNAs), deoxyribonucleic acids (DNAs), threose nucleic acids (TNAs), glycol nucleic acids (GNAs), peptide nucleic acids (PNAs), locked nucleic acids (LNAs, including LNA having a β- D-ribo configuration, α-LNA having an α-L-ribo configuration (a diastereomer of LNA), 2′-amino-LNA having a 2′-amino functionalization, and 2′-amino- α-LNA having a 2′-amino functionalization), ethylene nucleic acids (ENA), cyclohexenyl nucleic acids (CeNA), or chimeras or combinations thereof.

In some embodiments, polynucleotides of the present disclosure function as messenger RNA (mRNA). "Messenger RNA" (mRNA) refers to any polynucleotide that encodes a (at least one) polypeptide (a naturally-occurring, non-naturally-occurring, or modified polymer of amino acids) and can be translated to produce the encoded polypeptide *in vitro*, *in vivo*, *in situ,* or *ex vivo*. The skilled artisan will appreciate that, except where otherwise noted, polynucleotide sequences set forth in the instant application will recite "T"s in a representative DNA sequence but where the sequence represents RNA (*e.g.*, mRNA), the "T"s would be substituted for "U"s. Thus, any of the RNA polynucleotides encoded by a DNA identified by a particular sequence identification number may also comprise the corresponding RNA (*e.g.*, mRNA) sequence encoded by the DNA, where each "T" of the DNA sequence is substituted with "U."

The basic components of an mRNA molecule typically include at least one coding region, a 5′ untranslated region (UTR), a 3′ UTR, a 5′ cap, and a poly-A tail. Polynucleotides of the present disclosure may function as mRNA but can be distinguished from wild-type mRNA in their functional and/or structural design features which serve to overcome existing problems of effective polypeptide expression using nucleic-acid based therapeutics.

In some embodiments, a RNA polynucleotide of a HSV vaccine encodes 2-10, 2-9, 2-8, 2-7, 2-6, 2-5, 2-4, 2-3, 3-10, 3-9, 3-8, 3-7, 3-6, 3-5, 3-4, 4-10, 4-9, 4-8, 4-7, 4-6, 4-5, 5-10, 5-9, 5-8, 5-7, 5-6, 6-10, 6-9, 6-8, 6-7, 7-10, 7-9, 7-8, 8-10, 8-9 or 9-10 antigenic polypeptides. In some embodiments, a RNA polynucleotide of a HSV vaccine encodes at least 10, 20, 30,

40, 50, 60, 70, 80, 90 or 100 antigenic polypeptides. In some embodiments, a RNA polynucleotide of a HSV vaccine encodes at least 100 or at least 200 antigenic polypeptides. In some embodiments, a RNA polynucleotide of a HSV vaccine encodes 1-10, 5-15, 10-20, 15-25, 20-30, 25-35, 30-40, 35-45, 40-50, 1-50, 1-100, 2-50, or 2-100 antigenic polypeptides.

Polynucleotides of the present disclosure, in some embodiments, are codon optimized. Codon optimization methods are known in the art and may be used as provided herein. Codon optimization, in some embodiments, may be used to match codon frequencies in target and host organisms to ensure proper folding; bias GC content to increase mRNA stability or reduce secondary structures; minimize tandem repeat codons or base runs that may impair gene construction or expression; customize transcriptional and translational control regions; insert or remove protein trafficking sequences; remove/add post translation modification sites in encoded protein (*e.g.* glycosylation sites); add, remove, or shuffle protein domains; insert or delete restriction sites; modify ribosome binding sites and mRNA degradation sites; adjust translational rates to allow the various domains of the protein to fold properly; or to reduce or eliminate problem secondary structures within the polynucleotide. Codon optimization tools, algorithms and services are known in the art – non-limiting examples include services from GeneArt (Life Technologies), DNA2.0 (Menlo Park CA), and/or proprietary methods. In some embodiments, the open reading frame (ORF) sequence is optimized using optimization algorithms.

In some embodiments, a codon optimized sequence shares less than 95% sequence identity to a naturally-occurring or wild-type sequence (*e.g.*, a naturally-occurring or wild-type mRNA sequence encoding a polypeptide or protein of interest (*e.g.*, an antigenic protein or polypeptide)). In some embodiments, a codon optimized sequence shares less than 90% sequence identity to a naturally-occurring or wild-type sequence (*e.g.*, a naturally-occurring or wild-type mRNA sequence encoding a polypeptide or protein of interest (*e.g.*, an antigenic protein or polypeptide)). In some embodiments, a codon optimized sequence shares less than 85% sequence identity to a naturally-occurring or wild-type sequence (*e.g.*, a naturally-occurring or wild-type mRNA sequence encoding a polypeptide or protein of interest (*e.g.*, an antigenic protein or polypeptide)). In some embodiments, a codon optimized sequence shares less than 80% sequence identity to a naturally-occurring or wild-type sequence (*e.g.*, a naturally-occurring or wild-type mRNA sequence encoding a polypeptide or protein of interest (*e.g.*, an antigenic protein or polypeptide)). In some embodiments, a codon optimized sequence shares less than 75% sequence identity to a naturally-occurring or wild-type

sequence (*e.g.*, a naturally-occurring or wild-type mRNA sequence encoding a polypeptide or protein of interest (*e.g.*, an antigenic protein or polypeptide)).

In some embodiments, a codon optimized sequence shares between 65% and 85% (*e.g.*, between about 67% and about 85% or between about 67% and about 80%) sequence identity to a naturally-occurring or wild-type sequence (*e.g.*, a naturally-occurring or wild-type mRNA sequence encoding a polypeptide or protein of interest (*e.g.*, an antigenic protein or polypeptide)). In some embodiments, a codon optimized sequence shares between 65% and 75% or about 80% sequence identity to a naturally-occurring or wild-type sequence (*e.g.*, a naturally-occurring or wild-type mRNA sequence encoding a polypeptide or protein of interest (*e.g.*, an antigenic protein or polypeptide)).

In some embodiments, the HSV vaccine includes at least one RNA polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide having at least one modification, at least one 5′ terminal cap, and is formulated within a lipid nanoparticle. 5′-capping of polynucleotides may be completed concomitantly during the *in vitro*-transcription reaction using the following chemical RNA cap analogs to generate the 5′-guanosine cap structure according to manufacturer protocols: 3´-O-Me-m7G(5')ppp(5') G [the ARCA cap];G(5')ppp(5')A; G(5')ppp(5')G; m7G(5')ppp(5')A; m7G(5')ppp(5')G (New England BioLabs, Ipswich, MA). 5′-capping of modified RNA may be completed post-transcriptionally using a Vaccinia Virus Capping Enzyme to generate the "Cap 0" structure: m7G(5')ppp(5')G (New England BioLabs, Ipswich, MA). Cap 1 structure may be generated using both Vaccinia Virus Capping Enzyme and a 2′-O methyl-transferase to generate m7G(5')ppp(5')G-2′-O-methyl. Cap 2 structure may be generated from the Cap 1 structure followed by the 2′-O-methylation of the 5′-antepenultimate nucleotide using a 2′-O methyl-transferase. Cap 3 structure may be generated from the Cap 2 structure followed by the 2′-O-methylation of the 5′-preantepenultimate nucleotide using a 2′-O methyl-transferase. Enzymes are preferably derived from a recombinant source.

When transfected into mammalian cells, the modified mRNAs have a stability of between 12-18 hours or more than 18 hours, *e.g.*, 24, 36, 48, 60, 72, or greater than 72 hours.

In some embodiments, a codon optimized RNA may, for instance, be one in which the levels of G/C are enhanced. The G/C-content of nucleic acid molecules may influence the stability of the RNA. RNA having an increased amount of guanine (G) and/or cytosine (C) residues may be functionally more stable than nucleic acids containing a large amount of adenine (A) and thymine (T) or uracil (U) nucleotides. WO02/098443 discloses a pharmaceutical composition containing an mRNA stabilized by sequence modifications in the

translated region. Due to the degeneracy of the genetic code, the modifications work by substituting existing codons for those that promote greater RNA stability without changing the resulting amino acid. The approach is limited to coding regions of the RNA.

Antigens/Antigenic Polypeptides

In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.*, mRNA) polynucleotide having an open reading frame encoding HSV-2 glycoprotein B or an immunogenic fragment capable of inducing an immune response to (*e.g.*, SEQ ID NO: 1, 6, 12, 18, 66, or 71).

In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.*, mRNA) polynucleotide having an open reading frame encoding HSV-2 glycoprotein C or an immunogenic fragment capable of inducing an immune response to (*e.g.*, SEQ ID NO: 2, 7, 13, 19, 67, or 72).

In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.*, mRNA) polynucleotide having an open reading frame encoding HSV-2 glycoprotein D or an immunogenic fragment capable of inducing an immune response to (*e.g.*, SEQ ID NO: 3, 11, 14, 20, 68, or 75).

In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.*, mRNA) polynucleotide having an open reading frame encoding HSV-2 glycoprotein E or an immunogenic fragment capable of inducing an immune response to (*e.g.*, SEQ ID NO: 4, 8, 15, 21, 69, or 73).

In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.*, mRNA) polynucleotide having an open reading frame encoding HSV-2 glycoprotein I or an immunogenic fragment capable of inducing an immune response to (*e.g.*, SEQ ID NO: 5, 10, 13, 16, 22, 70, or 74).

In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.*, mRNA) polynucleotide having an open reading frame encoding HSV-2 ICP4 protein or an immunogenic fragment capable of inducing an immune response to (*e.g.*, SEQ ID NO: 9, 23, or 77).

In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.*, mRNA) polynucleotide having an open reading frame encoding HSV-2 ICP0 protein or an immunogenic fragment capable of inducing an immune response to (*e.g.*, SEQ ID NO: 17 or 76).

In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.* mRNA) polynucleotide encoded by a nucleic acid selected from any one of SEQ ID NO: 1-23 or 54-64 (*e.g.*, from Tables 1 or 3). In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.* mRNA) polynucleotide that comprises a nucleic acid selected from any one of SEQ ID NO: 90-124 (*e.g.*, from Tables 1 or 3).

In some embodiments, a HSV vaccine comprises at least one RNA (*e.g.*, mRNA) having at least one modification, including at least one chemical modification.

In some embodiments, a HSV antigenic polypeptide is longer than 25 amino acids and shorter than 50 amino acids. Thus, polypeptides include gene products, naturally occurring polypeptides, synthetic polypeptides, homologs, orthologs, paralogs, fragments and other equivalents, variants, and analogs of the foregoing. A polypeptide may be a single molecule or may be a multi-molecular complex such as a dimer, trimer, or tetramer. Polypeptides may also comprise single chain or multichain polypeptides such as antibodies or insulin and may be associated or linked. Most commonly, disulfide linkages are found in multichain polypeptides. The term polypeptide may also apply to amino acid polymers in which at least one amino acid residue is an artificial chemical analogue of a corresponding naturally-occurring amino acid.

The term "polypeptide variant" refers to molecules which differ in their amino acid sequence from a native or reference sequence. The amino acid sequence variants may possess substitutions, deletions, and/or insertions at certain positions within the amino acid sequence, as compared to a native or reference sequence. Ordinarily, variants possess at least 50% identity to a native or reference sequence. In some embodiments, variants share at least 80%, or at least 90% identity with a native or reference sequence.

In some embodiments "variant mimics" are provided. As used herein, the term "variant mimic" is one which contains at least one amino acid that would mimic an activated sequence. For example, glutamate may serve as a mimic for phosphoro-threonine and/or phosphoro-serine. Alternatively, variant mimics may result in deactivation or in an inactivated product containing the mimic, for example, phenylalanine may act as an inactivating substitution for tyrosine; or alanine may act as an inactivating substitution for serine.

"Orthologs" refers to genes in different species that evolved from a common ancestral gene by speciation. Normally, orthologs retain the same function in the course of evolution. Identification of orthologs is critical for reliable prediction of gene function in newly sequenced genomes.

"Analogs" is meant to include polypeptide variants which differ by one or more amino acid alterations, for example, substitutions, additions or deletions of amino acid residues that still maintain one or more of the properties of the parent or starting polypeptide.

"Paralogs" are genes (or proteins) related by duplication within a genome. Orthologs retain the same function in the course of evolution, whereas paralogs evolve new functions, even if these are related to the original one.

The present disclosure provides several types of compositions that are polynucleotide or polypeptide based, including variants and derivatives. These include, for example, substitutional, insertional, deletion and covalent variants and derivatives. The term "derivative" is used synonymously with the term "variant" but generally refers to a molecule that has been modified and/or changed in any way relative to a reference molecule or starting molecule.

As such, polynucleotides encoding peptides or polypeptides containing substitutions, insertions and/or additions, deletions and covalent modifications with respect to reference sequences, in particular the polypeptide sequences disclosed herein, are included within the scope of this disclosure. For example, sequence tags or amino acids, such as one or more lysines, can be added to peptide sequences (*e.g.*, at the N-terminal or C-terminal ends). Sequence tags can be used for peptide detection, purification or localization. Lysines can be used to increase peptide solubility or to allow for biotinylation. Alternatively, amino acid residues located at the carboxy and amino terminal regions of the amino acid sequence of a peptide or protein may optionally be deleted providing for truncated sequences. Certain amino acids (*e.g.*, C-terminal or N-terminal residues) may alternatively be deleted depending on the use of the sequence, as for example, expression of the sequence as part of a larger sequence which is soluble, or linked to a solid support. In alternative embodiments, sequences for (or encoding) signal sequences, termination sequences, transmembrane domains, linkers, multimerization domains (such as, *e.g.*, foldon regions) and the like may be substituted with alternative sequences which achieve the same or a similar function. Such sequences are readily identifiable to one of skill in the art. It should also be understood that some of the sequences provided herein contain sequence tags or terminal peptide sequences (*e.g.*, at the N-terminal or C-terminal ends) that may be deleted, for example, prior to use in the preparation of an RNA (*e.g.*, mRNA) vaccine.

"Substitutional variants" when referring to polypeptides are those that have at least one amino acid residue in a native or starting sequence removed and a different amino acid inserted in its place at the same position. Substitutions may be single, where only one amino

acid in the molecule has been substituted, or they may be multiple, where two or more amino acids have been substituted in the same molecule.

As used herein the term "conservative amino acid substitution" refers to the substitution of an amino acid that is normally present in the sequence with a different amino acid of similar size, charge, or polarity. Examples of conservative substitutions include the substitution of a non-polar (hydrophobic) residue such as isoleucine, valine and leucine for another non-polar residue. Likewise, examples of conservative substitutions include the substitution of one polar (hydrophilic) residue for another such as between arginine and lysine, between glutamine and asparagine, and between glycine and serine. Additionally, the substitution of a basic residue such as lysine, arginine, or histidine for another, or the substitution of one acidic residue such as aspartic acid or glutamic acid for another acidic residue are additional examples of conservative substitutions. Examples of non-conservative substitutions include the substitution of a non-polar (hydrophobic) amino acid residue such as isoleucine, valine, leucine, alanine, or methionine for a polar (hydrophilic) residue such as cysteine, glutamine, glutamic acid or lysine and/or a polar residue for a non-polar residue.

"Features" when referring to polypeptide or polynucleotide are defined as distinct amino acid sequence-based or nucleotide-based components of a molecule respectively. Features of the polypeptides encoded by the polynucleotides include surface manifestations, local conformational shape, folds, loops, half-loops, domains, half-domains, sites, termini or any combination thereof.

As used herein when referring to polypeptides the term "domain" refers to a motif of a polypeptide having one or more identifiable structural or functional characteristics or properties (*e.g.*, binding capacity, serving as a site for protein-protein interactions).

As used herein when referring to polypeptides, the terms "site" as it pertains to amino acid based embodiments is used synonymously with "amino acid residue" and "amino acid side chain." As used herein when referring to polynucleotides the terms "site" as it pertains to nucleotide based embodiments is used synonymously with "nucleotide." A site represents a position within a peptide or polypeptide or polynucleotide that may be modified, manipulated, altered, derivatized or varied within the polypeptide or polynucleotide based molecules.

As used herein the terms "termini" or "terminus" when referring to polypeptides or polynucleotides refers to an extremity of a polypeptide or polynucleotide, respectively. Such extremity is not limited only to the first or final site of the polypeptide or polynucleotide but may include additional amino acids or nucleotides in the terminal regions. Polypeptide-based

molecules may be characterized as having both an N-terminus (terminated by an amino acid with a free amino group (NH_2)) and a C-terminus (terminated by an amino acid with a free carboxyl group (COOH)). Proteins are, in some cases, made up of multiple polypeptide chains brought together by disulfide bonds or by non-covalent forces (multimers, oligomers). These proteins have multiple N- and C-termini. Alternatively, the termini of the polypeptides may be modified such that they begin or end, as the case may be, with a non-polypeptide based moiety such as an organic conjugate.

As recognized by those skilled in the art, protein fragments, functional protein domains, and homologous proteins are also considered to be within the scope of polypeptides of interest. For example, provided herein is any protein fragment (meaning a polypeptide sequence at least one amino acid residue shorter than a reference polypeptide sequence but otherwise identical) of a reference protein 10, 20, 30, 40, 50, 60, 70, 80, 90, 100 or greater than 100 amino acids in length. In another example, any protein that includes a stretch of 20, 30, 40, 50, or 100 amino acids which are 40%, 50%, 60%, 70%, 80%, 90%, 95%, or 100% identical to any of the sequences described herein can be utilized in accordance with the disclosure. In some embodiments, a polypeptide includes 2, 3, 4, 5, 6, 7, 8, 9, 10, or more mutations as shown in any of the sequences provided or referenced herein.

Polypeptide or polynucleotide molecules of the present disclosure may share a certain degree of sequence similarity or identity with the reference molecules (*e.g.*, reference polypeptides or reference polynucleotides), for example, with art-described molecules (*e.g.*, engineered or designed molecules or wild-type molecules). The term "identity" as known in the art, refers to a relationship between the sequences of two or more polypeptides or polynucleotides, as determined by comparing the sequences. In the art, identity also means the degree of sequence relatedness between them as determined by the number of matches between strings of two or more amino acid residues or nucleic acid residues. Identity measures the percent of identical matches between the smaller of two or more sequences with gap alignments (if any) addressed by a particular mathematical model or computer program (*e.g.*, "algorithms"). Identity of related peptides can be readily calculated by known methods. "% identity" as it applies to polypeptide or polynucleotide sequences is defined as the percentage of residues (amino acid residues or nucleic acid residues) in the candidate amino acid or nucleic acid sequence that are identical with the residues in the amino acid sequence or nucleic acid sequence of a second sequence after aligning the sequences and introducing gaps, if necessary, to achieve the maximum percent identity. Methods and computer programs for the alignment are well known in the art. It is understood that identity depends

on a calculation of percent identity but may differ in value due to gaps and penalties introduced in the calculation. Generally, variants of a particular polynucleotide or polypeptide have at least 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% but less than 100% sequence identity to that particular reference polynucleotide or polypeptide as determined by sequence alignment programs and parameters described herein and known to those skilled in the art. Such tools for alignment include those of the BLAST suite (Stephen F. Altschul, *et al.,* (1997), "Gapped BLAST and PSI-BLAST: a new generation of protein database search programs", *Nucleic Acids Res.* 25:3389-3402). Another popular local alignment technique is based on the Smith-Waterman algorithm (Smith, T.F. & Waterman, M.S. (1981) "Identification of common molecular subsequences." *J. Mol. Biol.* 147:195-197). A general global alignment technique based on dynamic programming is the Needleman–Wunsch algorithm (Needleman, S.B. & Wunsch, C.D. (1970) "A general method applicable to the search for similarities in the amino acid sequences of two proteins." *J. Mol. Biol.* 48:443-453.). More recently a Fast Optimal Global Sequence Alignment Algorithm (FOGSAA) has been developed that purportedly produces global alignment of nucleotide and protein sequences faster than other optimal global alignment methods, including the Needleman–Wunsch algorithm. Other tools are described herein, specifically in the definition of "identity" below.

As used herein, the term "homology" refers to the overall relatedness between polymeric molecules, *e.g.* between nucleic acid molecules (*e.g.* DNA molecules and/or RNA molecules) and/or between polypeptide molecules. Polymeric molecules (*e.g.* nucleic acid molecules (*e.g.* DNA molecules and/or RNA molecules) and/or polypeptide molecules) that share a threshold level of similarity or identity determined by alignment of matching residues are termed homologous. Homology is a qualitative term that describes a relationship between molecules and can be based upon the quantitative similarity or identity. Similarity or identity is a quantitative term that defines the degree of sequence match between two compared sequences. In some embodiments, polymeric molecules are considered to be "homologous" to one another if their sequences are at least 25%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, or 99% identical or similar. The term "homologous" necessarily refers to a comparison between at least two sequences (polynucleotide or polypeptide sequences). Two polynucleotide sequences are considered homologous if the polypeptides they encode are at least 50%, 60%, 70%, 80%, 90%, 95%, or even 99% for at least one stretch of at least 20 amino acids. In some embodiments, homologous polynucleotide sequences are characterized by the ability to encode a stretch of at least 4–5

uniquely specified amino acids. For polynucleotide sequences less than 60 nucleotides in length, homology is determined by the ability to encode a stretch of at least 4–5 uniquely specified amino acids. Two protein sequences are considered homologous if the proteins are at least 50%, 60%, 70%, 80%, or 90% identical for at least one stretch of at least 20 amino acids.

Homology implies that the compared sequences diverged in evolution from a common origin. The term "homolog" refers to a first amino acid sequence or nucleic acid sequence (*e.g.*, gene (DNA or RNA) or protein sequence) that is related to a second amino acid sequence or nucleic acid sequence by descent from a common ancestral sequence. The term "homolog" may apply to the relationship between genes and/or proteins separated by the event of speciation or to the relationship between genes and/or proteins separated by the event of genetic duplication.

Multiprotein and Multicomponent Vaccines

The present disclosure encompasses HSV vaccines comprising multiple RNA (*e.g.*, mRNA) polynucleotides, each encoding a single antigenic polypeptide, as well as HSV vaccines comprising a single RNA polynucleotide encoding more than one antigenic polypeptide (*e.g.*, as a fusion polypeptide). Thus, it should be understood that a vaccine composition comprising a RNA polynucleotide having an open reading frame encoding a first HSV antigenic polypeptide and a RNA polynucleotide having an open reading frame encoding a second HSV antigenic polypeptide encompasses (a) vaccines that comprise a first RNA polynucleotide encoding a first HSV antigenic polypeptide and a second RNA polynucleotide encoding a second HSV antigenic polypeptide, and (b) vaccines that comprise a single RNA polynucleotide encoding a first and second HSV antigenic polypeptide (*e.g.*, as a fusion polypeptide). HSV RNA (*e.g.*, mRNA) vaccines of the present disclosure, in some embodiments, comprise 2-10 (*e.g.*, 2, 3, 4, 5, 6, 7, 8, 9 or 10), or more RNA polynucleotides having an open reading frame, each of which encodes a different HSV antigenic polypeptide (or a single RNA polynucleotide encoding 2-10, or more, different HSV antigenic polypeptides).

In some embodiments, a RNA (*e.g.*, mRNA) polynucleotide encodes a HSV antigenic polypeptide fused to a signal peptide (*e.g.*, SEQ ID NO: 281 or SEQ ID NO: 282). Thus, HSV vaccines comprising at least one ribonucleic acid (RNA) polynucleotide having an open reading frame encoding a signal peptide linked to a HSV antigenic peptide are provided.

Further provided herein are HSV vaccines comprising any HSV antigenic polypeptides disclosed herein fused to signal peptides. The signal peptide may be fused to the N- or C- terminus of the HSV antigenic polypeptides.

Signal peptides

In some embodiments, antigenic polypeptides encoded by HSV polynucleotides comprise a signal peptide. Signal peptides, comprising the N-terminal 15-60 amino acids of proteins, are typically needed for the translocation across the membrane on the secretory pathway and thus universally control the entry of most proteins both in eukaryotes and prokaryotes to the secretory pathway. Signal peptides generally include of three regions: an N-terminal region of differing length, which usually comprises positively charged amino acids; a hydrophobic region; and a short carboxy-terminal peptide region. In eukaryotes, the signal peptide of a nascent precursor protein (pre-protein) directs the ribosome to the rough endoplasmic reticulum (ER) membrane and initiates the transport of the growing peptide chain across it. The signal peptide is not responsible for the final destination of the mature protein, however. Secretory proteins devoid of further address tags in their sequence are by default secreted to the external environment. Signal peptides are cleaved from precursor proteins by an endoplasmic reticulum (ER)-resident signal peptidase or they remain uncleaved and function as a membrane anchor. During recent years, a more advanced view of signal peptides has evolved, showing that the functions and immunodominance of certain signal peptides are much more versatile than previously anticipated.

Signal peptides typically function to facilitate the targeting of newly synthesized protein to the endoplasmic reticulum (ER) for processing. ER processing produces a mature Envelope protein, wherein the signal peptide is cleaved, typically by a signal peptidase of the host cell. A signal peptide may also facilitate the targeting of the protein to the cell membrane. HSV vaccines of the present disclosure may comprise, for example, RNA polynucleotides encoding an artificial signal peptide, wherein the signal peptide coding sequence is operably linked to and is in frame with the coding sequence of the HSV antigenic polypeptide. Thus, HSV vaccines of the present disclosure, in some embodiments, produce an antigenic polypeptide comprising a HSV antigenic polypeptide fused to a signal peptide. In some embodiments, a signal peptide is fused to the N-terminus of the HSV antigenic polypeptide. In some embodiments, a signal peptide is fused to the C-terminus of the HSV antigenic polypeptide.

In some embodiments, the signal peptide fused to the HSV antigenic polypeptide is an artificial signal peptide. In some embodiments, an artificial signal peptide fused to the HSV antigenic polypeptide encoded by the HSV RNA (*e.g.*, mRNA) vaccine is obtained from an immunoglobulin protein, *e.g.*, an IgE signal peptide or an IgG signal peptide. In some embodiments, a signal peptide fused to the HSV antigenic polypeptide encoded by a HSV RNA (*e.g.*, mRNA) vaccine is an Ig heavy chain epsilon-1 signal peptide (IgE HC SP) having the sequence of: MDWTWILFLVAAATRVHS (SEQ ID NO: 79). In some embodiments, a signal peptide fused to a HSV antigenic polypeptide encoded by the HSV RNA (*e.g.*, mRNA) vaccine is an IgGk chain V-III region HAH signal peptide (IgGk SP) having the sequence of METPAQLLFLLLLWLPDTTG (SEQ ID NO: 78). In some embodiments, the HSV antigenic polypeptide encoded by a HSV RNA (*e.g.*, mRNA) vaccine has an amino acid sequence set forth in one of SEQ ID NO: 24-53 or 66-77 fused to a signal peptide of SEQ ID NO: 78-82. The examples disclosed herein are not meant to be limiting and any signal peptide that is known in the art to facilitate targeting of a protein to ER for processing and/or targeting of a protein to the cell membrane may be used in accordance with the present disclosure.

A signal peptide may have a length of 15-60 amino acids. For example, a signal peptide may have a length of 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55, 56, 57, 58, 59, or 60 amino acids. In some embodiments, a signal peptide may have a length of 20-60, 25-60, 30-60, 35-60, 40-60, 45-60, 50-60, 55-60, 15-55, 20-55, 25-55, 30-55, 35-55, 40-55, 45-55, 50-55, 15-50, 20-50, 25-50, 30-50, 35-50, 40-50, 45-50, 15-45, 20-45, 25-45, 30-45, 35-45, 40-45, 15-40, 20-40, 25-40, 30-40, 35-40, 15-35, 20-35, 25-35, 30-35, 15-30, 20-30, 25-30, 15-25, 20-25, or 15-20 amino acids.

A signal peptide is typically cleaved from the nascent polypeptide at the cleavage junction during ER processing. The mature HSV antigenic polypeptide produced by HSV RNA (*e.g.,* mRNA) vaccine of the present disclosure typically does not comprise a signal peptide.

Chemical Modifications

RNA (*e.g.*, mRNA) vaccines of the present disclosure comprise, in some embodiments, at least one ribonucleic acid (RNA) polynucleotide having an open reading frame encoding at least one herpes simplex virus (HSV) antigenic polypeptide, wherein said RNA comprises at least one chemical modification.

The terms "chemical modification" and "chemically modified" refer to modification with respect to adenosine (A), guanosine (G), uridine (U), thymidine (T), or cytidine (C) ribonucleosides or deoxyribnucleosides in at least one of their position, pattern, percent or population. Generally, these terms do not refer to the ribonucleotide modifications in naturally-occurring 5′-terminal mRNA cap moieties.

Modifications of polynucleotides include, without limitation, those described herein, and include, but are expressly not limited to, those modifications that comprise chemical modifications. Polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides) may comprise modifications that are naturally-occurring, non-naturally-occurring or the polynucleotide may comprise a combination of naturally-occurring and non-naturally-occurring modifications. Polynucleotides may include any useful modification, for example, of a sugar, a nucleobase, or an internucleoside linkage (*e.g.*, to a linking phosphate, to a phosphodiester linkage or to the phosphodiester backbone).

With respect to a polypeptide, the term "modification" refers to a modification relative to the canonical set of 20 amino acids. Polypeptides, as provided herein, are also considered "modified" if they contain amino acid substitutions, insertions, or a combination of substitutions and insertions.

Polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides), in some embodiments, comprise various (more than one) different modifications. In some embodiments, a particular region of a polynucleotide contains one, two, or more (optionally different) nucleoside or nucleotide modifications. In some embodiments, a modified RNA polynucleotide (*e.g.*, a modified mRNA polynucleotide), introduced to a cell or organism, exhibits reduced degradation in the cell or organism, respectively, relative to an unmodified polynucleotide. In some embodiments, a modified RNA polynucleotide (*e.g.*, a modified mRNA polynucleotide), introduced into a cell or organism, may exhibit reduced immunogenicity in the cell or organism, respectively (*e.g.*, a reduced innate response).

Polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides), in some embodiments, comprise non-natural modified nucleotides that are introduced during synthesis or post-synthesis of the polynucleotides to achieve desired functions or properties. The modifications may be present on internucleotide linkages, purine or pyrimidine bases, or sugars. The modification may be introduced with chemical synthesis or with a polymerase enzyme at the terminal of a chain or anywhere else in the chain. Any of the regions of a polynucleotide may be chemically modified.

The present disclosure provides for modified nucleosides and nucleotides of a polynucleotide (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides). A "nucleoside" refers to a compound containing a sugar molecule (*e.g.*, a pentose or ribose) or a derivative thereof in combination with an organic base (*e.g.*, a purine or pyrimidine) or a derivative thereof (also referred to herein as "nucleobase"). A "nucleotide" refers to a nucleoside including a phosphate group. Modified nucleotides may by synthesized by any useful method, such as, for example, chemically, enzymatically, or recombinantly, to include one or more modified or non-natural nucleosides. Polynucleotides may comprise a region or regions of linked nucleosides. Such regions may have variable backbone linkages. The linkages may be standard phosphdioester linkages, in which case the polynucleotides would comprise regions of nucleotides.

Modified nucleotide base pairing encompasses not only the standard adenosine-thymine, adenosine-uracil, or guanosine-cytosine base pairs, but also base pairs formed between nucleotides and/or modified nucleotides comprising non-standard or modified bases, wherein the arrangement of hydrogen bond donors and hydrogen bond acceptors permits hydrogen bonding between a non-standard base and a standard base or between two complementary non-standard base structures, such as, for example, in those polynucleotides having at least one chemical modification. One example of such non-standard base pairing is the base pairing between the modified nucleotide inosine and adenine, cytosine, or uracil. Any combination of base/sugar or linker may be incorporated into polynucleotides of the present disclosure.

Modifications of polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides), including but not limited to chemical modification, that are useful in the compositions, vaccines, methods and synthetic processes of the present disclosure include, but are not limited to the following: 2-methylthio-N6-(cis-hydroxyisopentenyl)adenosine; 2-methylthio-N6-methyladenosine; 2-methylthio-N6-threonyl carbamoyladenosine; N6-glycinylcarbamoyladenosine; N6-isopentenyladenosine; N6-methyladenosine; N6-threonylcarbamoyladenosine; 1,2′-O-dimethyladenosine; 1-methyladenosine; 2′-O-methyladenosine; 2′-O-ribosyladenosine (phosphate); 2-methyladenosine; 2-methylthio-N6 isopentenyladenosine; 2-methylthio-N6-hydroxynorvalyl carbamoyladenosine; 2'-O-methyladenosine; 2'-O-ribosyladenosine (phosphate); Isopentenyladenosine; N6-(cis-hydroxyisopentenyl)adenosine; N6,2′-O-dimethyladenosine; N6,2'-O-dimethyladenosine; N6,N6,2′-O-trimethyladenosine; N6,N6-dimethyladenosine; N6-acetyladenosine; N6-hydroxynorvalylcarbamoyladenosine; N6-methyl-N6-threonylcarbamoyladenosine; 2-

methyladenosine; 2-methylthio-N6-isopentenyladenosine; 7-deaza-adenosine; N1-methyl-adenosine; N6, N6 (dimethyl)adenine; N6-cis-hydroxy-isopentenyl-adenosine; α-thio-adenosine; 2 (amino)adenine; 2 (aminopropyl)adenine; 2 (methylthio) N6 (isopentenyl)adenine; 2-(alkyl)adenine; 2-(aminoalkyl)adenine; 2-(aminopropyl)adenine; 2-(halo)adenine; 2-(halo)adenine; 2-(propyl)adenine; 2'-Amino-2'-deoxy-ATP; 2'-Azido-2'-deoxy-ATP; 2'-Deoxy-2'-a-aminoadenosine TP; 2'-Deoxy-2'-a-azidoadenosine TP; 6 (alkyl)adenine; 6 (methyl)adenine; 6-(alkyl)adenine; 6-(methyl)adenine; 7 (deaza)adenine; 8 (alkenyl)adenine; 8 (alkynyl)adenine; 8 (amino)adenine; 8 (thioalkyl)adenine; 8-(alkenyl)adenine; 8-(alkyl)adenine; 8-(alkynyl)adenine; 8-(amino)adenine; 8-(halo)adenine; 8-(hydroxyl)adenine; 8-(thioalkyl)adenine; 8-(thiol)adenine; 8-azido-adenosine; aza adenine; deaza adenine; N6 (methyl)adenine; N6-(isopentyl)adenine; 7-deaza-8-aza-adenosine; 7-methyladenine; 1-Deazaadenosine TP; 2'Fluoro-N6-Bz-deoxyadenosine TP; 2'-OMe-2-Amino-ATP; 2'O-methyl-N6-Bz-deoxyadenosine TP; 2'-a-Ethynyladenosine TP; 2-aminoadenine; 2-Aminoadenosine TP; 2-Amino-ATP; 2'-a-Trifluoromethyladenosine TP; 2-Azidoadenosine TP; 2'-b-Ethynyladenosine TP; 2-Bromoadenosine TP; 2'-b-Trifluoromethyladenosine TP; 2-Chloroadenosine TP; 2'-Deoxy-2',2'-difluoroadenosine TP; 2'-Deoxy-2'-a-mercaptoadenosine TP; 2'-Deoxy-2'-a-thiomethoxyadenosine TP; 2'-Deoxy-2'-b-aminoadenosine TP; 2'-Deoxy-2'-b-azidoadenosine TP; 2'-Deoxy-2'-b-bromoadenosine TP; 2'-Deoxy-2'-b-chloroadenosine TP; 2'-Deoxy-2'-b-fluoroadenosine TP; 2'-Deoxy-2'-b-iodoadenosine TP; 2'-Deoxy-2'-b-mercaptoadenosine TP; 2'-Deoxy-2'-b-thiomethoxyadenosine TP; 2-Fluoroadenosine TP; 2-Iodoadenosine TP; 2-Mercaptoadenosine TP; 2-methoxy-adenine; 2-methylthio-adenine; 2-Trifluoromethyladenosine TP; 3-Deaza-3-bromoadenosine TP; 3-Deaza-3-chloroadenosine TP; 3-Deaza-3-fluoroadenosine TP; 3-Deaza-3-iodoadenosine TP; 3-Deazaadenosine TP; 4'-Azidoadenosine TP; 4'-Carbocyclic adenosine TP; 4'-Ethynyladenosine TP; 5'-Homo-adenosine TP; 8-Aza-ATP; 8-bromo-adenosine TP; 8-Trifluoromethyladenosine TP; 9-Deazaadenosine TP; 2-aminopurine; 7-deaza-2,6-diaminopurine; 7-deaza-8-aza-2,6-diaminopurine; 7-deaza-8-aza-2-aminopurine; 2,6-diaminopurine; 7-deaza-8-aza-adenine, 7-deaza-2-aminopurine; 2-thiocytidine; 3-methylcytidine; 5-formylcytidine; 5-hydroxymethylcytidine; 5-methylcytidine; N4-acetylcytidine; 2′-O-methylcytidine; 2'-O-methylcytidine; 5,2′-O-dimethylcytidine; 5-formyl-2′-O-methylcytidine; Lysidine; N4,2′-O-dimethylcytidine; N4-acetyl-2′-O-methylcytidine; N4-methylcytidine; N4,N4-Dimethyl-2'-OMe-Cytidine TP; 4-methylcytidine; 5-aza-cytidine; Pseudo-iso-cytidine; pyrrolo-cytidine; α-thio-cytidine; 2-(thio)cytosine; 2'-Amino-2'-deoxy-CTP; 2'-Azido-2'-deoxy-CTP; 2'-

Deoxy-2'-a-aminocytidine TP; 2'-Deoxy-2'-a-azidocytidine TP; 3 (deaza) 5 (aza)cytosine; 3 (methyl)cytosine; 3-(alkyl)cytosine; 3-(deaza) 5 (aza)cytosine; 3-(methyl)cytidine; 4,2'-O-dimethylcytidine; 5 (halo)cytosine; 5 (methyl)cytosine; 5 (propynyl)cytosine; 5 (trifluoromethyl)cytosine; 5-(alkyl)cytosine; 5-(alkynyl)cytosine; 5-(halo)cytosine; 5-(propynyl)cytosine; 5-(trifluoromethyl)cytosine; 5-bromo-cytidine; 5-iodo-cytidine; 5-propynyl cytosine; 6-(azo)cytosine; 6-aza-cytidine; aza cytosine; deaza cytosine; N4 (acetyl)cytosine; 1-methyl-1-deaza-pseudoisocytidine; 1-methyl-pseudoisocytidine; 2-methoxy-5-methyl-cytidine; 2-methoxy-cytidine; 2-thio-5-methyl-cytidine; 4-methoxy-1-methyl-pseudoisocytidine; 4-methoxy-pseudoisocytidine; 4-thio-1-methyl-1-deaza-pseudoisocytidine; 4-thio-1-methyl-pseudoisocytidine; 4-thio-pseudoisocytidine; 5-aza-zebularine; 5-methyl-zebularine; pyrrolo-pseudoisocytidine; Zebularine; (E)-5-(2-Bromo-vinyl)cytidine TP; 2,2'-anhydro-cytidine TP hydrochloride; 2'Fluor-N4-Bz-cytidine TP; 2'Fluoro-N4-Acetyl-cytidine TP; 2'-O-Methyl-N4-Acetyl-cytidine TP; 2'O-methyl-N4-Bz-cytidine TP; 2'-a-Ethynylcytidine TP; 2'-a-Trifluoromethylcytidine TP; 2'-b-Ethynylcytidine TP; 2'-b-Trifluoromethylcytidine TP; 2'-Deoxy-2',2'-difluorocytidine TP; 2'-Deoxy-2'-a-mercaptocytidine TP; 2'-Deoxy-2'-a-thiomethoxycytidine TP; 2'-Deoxy-2'-b-aminocytidine TP; 2'-Deoxy-2'-b-azidocytidine TP; 2'-Deoxy-2'-b-bromocytidine TP; 2'-Deoxy-2'-b-chlorocytidine TP; 2'-Deoxy-2'-b-fluorocytidine TP; 2'-Deoxy-2'-b-iodocytidine TP; 2'-Deoxy-2'-b-mercaptocytidine TP; 2'-Deoxy-2'-b-thiomethoxycytidine TP; 2'-O-Methyl-5-(1-propynyl)cytidine TP; 3'-Ethynylcytidine TP; 4'-Azidocytidine TP; 4'-Carbocyclic cytidine TP; 4'-Ethynylcytidine TP; 5-(1-Propynyl)ara-cytidine TP; 5-(2-Chloro-phenyl)-2-thiocytidine TP; 5-(4-Amino-phenyl)-2-thiocytidine TP; 5-Aminoallyl-CTP; 5-Cyanocytidine TP; 5-Ethynylara-cytidine TP; 5-Ethynylcytidine TP; 5'-Homo-cytidine TP; 5-Methoxycytidine TP; 5-Trifluoromethyl-Cytidine TP; N4-Amino-cytidine TP; N4-Benzoyl-cytidine TP; Pseudoisocytidine; 7-methylguanosine; N2,2′-O-dimethylguanosine; N2-methylguanosine; Wyosine; 1,2′-O-dimethylguanosine; 1-methylguanosine; 2′-O-methylguanosine; 2′-O-ribosylguanosine (phosphate); 2'-O-methylguanosine; 2'-O-ribosylguanosine (phosphate); 7-aminomethyl-7-deazaguanosine; 7-cyano-7-deazaguanosine; Archaeosine; Methylwyosine; N2,7-dimethylguanosine; N2,N2,2′-O-trimethylguanosine; N2,N2,7-trimethylguanosine; N2,N2-dimethylguanosine; N2,7,2'-O-trimethylguanosine; 6-thio-guanosine; 7-deaza-guanosine; 8-oxo-guanosine; N1-methyl-guanosine; α-thio-guanosine; 2 (propyl)guanine; 2-(alkyl)guanine; 2'-Amino-2'-deoxy-GTP; 2'-Azido-2'-deoxy-GTP; 2'-Deoxy-2'-a-aminoguanosine TP; 2'-Deoxy-2'-a-azidoguanosine TP; 6 (methyl)guanine; 6-(alkyl)guanine; 6-(methyl)guanine; 6-methyl-guanosine; 7

(alkyl)guanine; 7 (deaza)guanine; 7 (methyl)guanine; 7-(alkyl)guanine; 7-(deaza)guanine; 7-(methyl)guanine; 8 (alkyl)guanine; 8 (alkynyl)guanine; 8 (halo)guanine; 8 (thioalkyl)guanine; 8-(alkenyl)guanine; 8-(alkyl)guanine; 8-(alkynyl)guanine; 8-(amino)guanine; 8-(halo)guanine; 8-(hydroxyl)guanine; 8-(thioalkyl)guanine; 8-(thiol)guanine; aza guanine; deaza guanine; N (methyl)guanine; N-(methyl)guanine; 1-methyl-6-thio-guanosine; 6-methoxy-guanosine; 6-thio-7-deaza-8-aza-guanosine; 6-thio-7-deaza-guanosine; 6-thio-7-methyl-guanosine; 7-deaza-8-aza-guanosine; 7-methyl-8-oxo-guanosine; N2,N2-dimethyl-6-thio-guanosine; N2-methyl-6-thio-guanosine; 1-Me-GTP; 2'Fluoro-N2-isobutyl-guanosine TP; 2'O-methyl-N2-isobutyl-guanosine TP; 2'-a-Ethynylguanosine TP; 2'-a-Trifluoromethylguanosine TP; 2'-b-Ethynylguanosine TP; 2'-b-Trifluoromethylguanosine TP; 2'-Deoxy-2',2'-difluoroguanosine TP; 2'-Deoxy-2'-a-mercaptoguanosine TP; 2'-Deoxy-2'-a-thiomethoxyguanosine TP; 2'-Deoxy-2'-b-aminoguanosine TP; 2'-Deoxy-2'-b-azidoguanosine TP; 2'-Deoxy-2'-b-bromoguanosine TP; 2'-Deoxy-2'-b-chloroguanosine TP; 2'-Deoxy-2'-b-fluoroguanosine TP; 2'-Deoxy-2'-b-iodoguanosine TP; 2'-Deoxy-2'-b-mercaptoguanosine TP; 2'-Deoxy-2'-b-thiomethoxyguanosine TP; 4'-Azidoguanosine TP; 4'-Carbocyclic guanosine TP; 4'-Ethynylguanosine TP; 5'-Homo-guanosine TP; 8-bromo-guanosine TP; 9-Deazaguanosine TP; N2-isobutyl-guanosine TP; 1-methylinosine; Inosine; 1,2′-O-dimethylinosine; 2′-O-methylinosine; 7-methylinosine; 2'-O-methylinosine; Epoxyqueuosine; galactosyl-queuosine; Mannosylqueuosine; Queuosine; allyamino-thymidine; aza thymidine; deaza thymidine; deoxy-thymidine; 2'-O-methyluridine; 2-thiouridine; 3-methyluridine; 5-carboxymethyluridine; 5-hydroxyuridine; 5-methyluridine; 5-taurinomethyl-2-thiouridine; 5-taurinomethyluridine; Dihydrouridine; Pseudouridine; (3-(3-amino-3-carboxypropyl)uridine; 1-methyl-3-(3-amino-5-carboxypropyl)pseudouridine; 1-methylpseduouridine; 1-ethyl-pseudouridine; 2′-O-methyluridine; 2'-O-methylpseudouridine; 2'-O-methyluridine; 2-thio-2′-O-methyluridine; 3-(3-amino-3-carboxypropyl)uridine; 3,2′-O-dimethyluridine; 3-Methyl-pseudo-Uridine TP; 4-thiouridine; 5-(carboxyhydroxymethyl)uridine; 5-(carboxyhydroxymethyl)uridine methyl ester; 5,2′-O-dimethyluridine; 5,6-dihydro-uridine; 5-aminomethyl-2-thiouridine; 5-carbamoylmethyl-2′-O-methyluridine; 5-carbamoylmethyluridine; 5-carboxyhydroxymethyluridine; 5-carboxyhydroxymethyluridine methyl ester; 5-carboxymethylaminomethyl-2′-O-methyluridine; 5-carboxymethylaminomethyl-2-thiouridine; 5-carboxymethylaminomethyl-2-thiouridine; 5-carboxymethylaminomethyluridine; 5-carboxymethylaminomethyluridine; 5-Carbamoylmethyluridine TP; 5-methoxycarbonylmethyl-2′-O-methyluridine; 5-methoxycarbonylmethyl-2-thiouridine; 5-methoxycarbonylmethyluridine; 5-methyluridine,),

5-methoxyuridine; 5-methyl-2-thiouridine; 5-methylaminomethyl-2-selenouridine; 5-methylaminomethyl-2-thiouridine; 5-methylaminomethyluridine; 5-Methyldihydrouridine; 5-Oxyacetic acid- Uridine TP; 5-Oxyacetic acid-methyl ester-Uridine TP; N1-methyl-pseudo-uracil; N1-ethyl-pseudo-uracil; uridine 5-oxyacetic acid; uridine 5-oxyacetic acid methyl ester; 3-(3-Amino-3-carboxypropyl)-Uridine TP; 5-(iso-Pentenylaminomethyl)- 2-thiouridine TP; 5-(iso-Pentenylaminomethyl)-2'-O-methyluridine TP; 5-(iso-Pentenylaminomethyl)uridine TP; 5-propynyl uracil; α-thio-uridine; 1 (aminoalkylamino-carbonylethylenyl)-2(thio)-pseudouracil; 1 (aminoalkylaminocarbonylethylenyl)-2,4-(dithio)pseudouracil; 1 (aminoalkylaminocarbonylethylenyl)-4 (thio)pseudouracil; 1 (aminoalkylaminocarbonylethylenyl)-pseudouracil; 1 (aminocarbonylethylenyl)-2(thio)-pseudouracil; 1 (aminocarbonylethylenyl)-2,4-(dithio)pseudouracil; 1 (aminocarbonylethylenyl)-4 (thio)pseudouracil; 1 (aminocarbonylethylenyl)-pseudouracil; 1 substituted 2(thio)-pseudouracil; 1 substituted 2,4-(dithio)pseudouracil; 1 substituted 4 (thio)pseudouracil; 1 substituted pseudouracil; 1-(aminoalkylamino-carbonylethylenyl)-2-(thio)-pseudouracil; 1-Methyl-3-(3-amino-3-carboxypropyl) pseudouridine TP; 1-Methyl-3-(3-amino-3-carboxypropyl)pseudo-UTP; 1-Methyl-pseudo-UTP; 1-Ethyl-pseudo-UTP; 2 (thio)pseudouracil; 2' deoxy uridine; 2' fluorouridine; 2-(thio)uracil; 2,4-(dithio)psuedouracil; 2' methyl, 2'amino, 2'azido, 2'fluro-guanosine; 2'-Amino-2'-deoxy-UTP; 2'-Azido-2'-deoxy-UTP; 2'-Azido-deoxyuridine TP; 2'-O-methylpseudouridine; 2′ deoxy uridine; 2′ fluorouridine; 2'-Deoxy-2'-a-aminouridine TP; 2'-Deoxy-2'-a-azidouridine TP; 2-methylpseudouridine; 3 (3 amino-3 carboxypropyl)uracil; 4 (thio)pseudouracil; 4-(thio)pseudouracil; 4-(thio)uracil; 4-thiouracil; 5 (1,3-diazole-1-alkyl)uracil; 5 (2-aminopropyl)uracil; 5 (aminoalkyl)uracil; 5 (dimethylaminoalkyl)uracil; 5 (guanidiniumalkyl)uracil; 5 (methoxycarbonylmethyl)-2-(thio)uracil; 5 (methoxycarbonyl-methyl)uracil; 5 (methyl) 2 (thio)uracil; 5 (methyl) 2,4 (dithio)uracil; 5 (methyl) 4 (thio)uracil; 5 (methylaminomethyl)-2 (thio)uracil; 5 (methylaminomethyl)-2,4 (dithio)uracil; 5 (methylaminomethyl)-4 (thio)uracil; 5 (propynyl)uracil; 5 (trifluoromethyl)uracil; 5-(2-aminopropyl)uracil; 5-(alkyl)-2-(thio)pseudouracil; 5-(alkyl)-2,4 (dithio)pseudouracil; 5-(alkyl)-4 (thio)pseudouracil; 5-(alkyl)pseudouracil; 5-(alkyl)uracil; 5-(alkynyl)uracil; 5-(allylamino)uracil; 5-(cyanoalkyl)uracil; 5-(dialkylaminoalkyl)uracil; 5-(dimethylaminoalkyl)uracil; 5-(guanidiniumalkyl)uracil; 5-(halo)uracil; 5-(l,3-diazole-l-alkyl)uracil; 5-(methoxy)uracil; 5-(methoxycarbonylmethyl)-2-(thio)uracil; 5-(methoxycarbonyl-methyl)uracil; 5-(methyl) 2(thio)uracil; 5-(methyl) 2,4 (dithio)uracil; 5-(methyl) 4 (thio)uracil; 5-(methyl)-2-(thio)pseudouracil; 5-(methyl)-2,4 (dithio)pseudouracil;

5-(methyl)-4 (thio)pseudouracil; 5-(methyl)pseudouracil; 5-(methylaminomethyl)-2 (thio)uracil; 5-(methylaminomethyl)-2,4(dithio)uracil; 5-(methylaminomethyl)-4-(thio)uracil; 5-(propynyl)uracil; 5-(trifluoromethyl)uracil; 5-aminoallyl-uridine; 5-bromo-uridine; 5-iodo-uridine; 5-uracil; 6 (azo)uracil; 6-(azo)uracil; 6-aza-uridine; allyamino-uracil; aza uracil; deaza uracil; N3 (methyl)uracil; P seudo-UTP-1-2-ethanoic acid; Pseudouracil; 4-Thio-pseudo-UTP; 1-carboxymethyl-pseudouridine; 1-methyl-1-deaza-pseudouridine; 1-propynyl-uridine; 1-taurinomethyl-1-methyl-uridine; 1-taurinomethyl-4-thio-uridine; 1-taurinomethyl-pseudouridine; 2-methoxy-4-thio-pseudouridine; 2-thio-1-methyl-1-deaza-pseudouridine; 2-thio-1-methyl-pseudouridine; 2-thio-5-aza-uridine; 2-thio-dihydropseudouridine; 2-thio-dihydrouridine; 2-thio-pseudouridine; 4-methoxy-2-thio-pseudouridine; 4-methoxy-pseudouridine; 4-thio-1-methyl-pseudouridine; 4-thio-pseudouridine; 5-aza-uridine; Dihydropseudouridine; (±)1-(2-Hydroxypropyl)pseudouridine TP; (2R)-1-(2-Hydroxypropyl)pseudouridine TP; (2S)-1-(2-Hydroxypropyl)pseudouridine TP; (E)-5-(2-Bromo-vinyl)ara-uridine TP; (E)-5-(2-Bromo-vinyl)uridine TP; (Z)-5-(2-Bromo-vinyl)ara-uridine TP; (Z)-5-(2-Bromo-vinyl)uridine TP; 1-(2,2,2-Trifluoroethyl)-pseudo-UTP; 1-(2,2,3,3,3-Pentafluoropropyl)pseudouridine TP; 1-(2,2-Diethoxyethyl)pseudouridine TP; 1-(2,4,6-Trimethylbenzyl)pseudouridine TP; 1-(2,4,6-Trimethyl-benzyl)pseudo-UTP; 1-(2,4,6-Trimethyl-phenyl)pseudo-UTP; 1-(2-Amino-2-carboxyethyl)pseudo-UTP; 1-(2-Amino-ethyl)pseudo-UTP; 1-(2-Hydroxyethyl)pseudouridine TP; 1-(2-Methoxyethyl)pseudouridine TP; 1-(3,4-Bis-trifluoromethoxybenzyl)pseudouridine TP; 1-(3,4-Dimethoxybenzyl)pseudouridine TP; 1-(3-Amino-3-carboxypropyl)pseudo-UTP; 1-(3-Amino-propyl)pseudo-UTP; 1-(3-Cyclopropyl-prop-2-ynyl)pseudouridine TP; 1-(4-Amino-4-carboxybutyl)pseudo-UTP; 1-(4-Amino-benzyl)pseudo-UTP; 1-(4-Amino-butyl)pseudo-UTP; 1-(4-Amino-phenyl)pseudo-UTP; 1-(4-Azidobenzyl)pseudouridine TP; 1-(4-Bromobenzyl)pseudouridine TP; 1-(4-Chlorobenzyl)pseudouridine TP; 1-(4-Fluorobenzyl)pseudouridine TP; 1-(4-Iodobenzyl)pseudouridine TP; 1-(4-Methanesulfonylbenzyl)pseudouridine TP; 1-(4-Methoxybenzyl)pseudouridine TP; 1-(4-Methoxy-benzyl)pseudo-UTP; 1-(4-Methoxy-phenyl)pseudo-UTP; 1-(4-Methylbenzyl)pseudouridine TP; 1-(4-Methyl-benzyl)pseudo-UTP; 1-(4-Nitrobenzyl)pseudouridine TP; 1-(4-Nitro-benzyl)pseudo-UTP; 1(4-Nitro-phenyl)pseudo-UTP; 1-(4-Thiomethoxybenzyl)pseudouridine TP; 1-(4-Trifluoromethoxybenzyl)pseudouridine TP; 1-(4-Trifluoromethylbenzyl)pseudouridine TP; 1-(5-Amino-pentyl)pseudo-UTP; 1-(6-Amino-hexyl)pseudo-UTP; 1,6-Dimethyl-pseudo-UTP; 1-[3-(2-{2-[2-(2-Aminoethoxy)-ethoxy]-ethoxy}-ethoxy)-propionyl]pseudouridine TP;

1-{3-[2-(2-Aminoethoxy)-ethoxy]-propionyl } pseudouridine TP; 1-Acetylpseudouridine TP; 1-Alkyl-6-(1-propynyl)-pseudo-UTP; 1-Alkyl-6-(2-propynyl)-pseudo-UTP; 1-Alkyl-6-allyl-pseudo-UTP; 1-Alkyl-6-ethynyl-pseudo-UTP; 1-Alkyl-6-homoallyl-pseudo-UTP; 1-Alkyl-6-vinyl-pseudo-UTP; 1-Allylpseudouridine TP; 1-Aminomethyl-pseudo-UTP; 1-Benzoylpseudouridine TP; 1-Benzyloxymethylpseudouridine TP; 1-Benzyl-pseudo-UTP; 1-Biotinyl-PEG2-pseudouridine TP; 1-Biotinylpseudouridine TP; 1-Butyl-pseudo-UTP; 1-Cyanomethylpseudouridine TP; 1-Cyclobutylmethyl-pseudo-UTP; 1-Cyclobutyl-pseudo-UTP; 1-Cycloheptylmethyl-pseudo-UTP; 1-Cycloheptyl-pseudo-UTP; 1-Cyclohexylmethyl-pseudo-UTP; 1-Cyclohexyl-pseudo-UTP; 1-Cyclooctylmethyl-pseudo-UTP; 1-Cyclooctyl-pseudo-UTP; 1-Cyclopentylmethyl-pseudo-UTP; 1-Cyclopentyl-pseudo-UTP; 1-Cyclopropylmethyl-pseudo-UTP; 1-Cyclopropyl-pseudo-UTP; 1-Ethyl-pseudo-UTP; 1-Hexyl-pseudo-UTP; 1-Homoallylpseudouridine TP; 1-Hydroxymethylpseudouridine TP; 1-iso-propyl-pseudo-UTP; 1-Me-2-thio-pseudo-UTP; 1-Me-4-thio-pseudo-UTP; 1-Me-alpha-thio-pseudo-UTP; 1-Methanesulfonylmethylpseudouridine TP; 1-Methoxymethylpseudouridine TP; 1-Methyl-6-(2,2,2-Trifluoroethyl)pseudo-UTP; 1-Methyl-6-(4-morpholino)-pseudo-UTP; 1-Methyl-6-(4-thiomorpholino)-pseudo-UTP; 1-Methyl-6-(substituted phenyl)pseudo-UTP; 1-Methyl-6-amino-pseudo-UTP; 1-Methyl-6-azido-pseudo-UTP; 1-Methyl-6-bromo-pseudo-UTP; 1-Methyl-6-butyl-pseudo-UTP; 1-Methyl-6-chloro-pseudo-UTP; 1-Methyl-6-cyano-pseudo-UTP; 1-Methyl-6-dimethylamino-pseudo-UTP; 1-Methyl-6-ethoxy-pseudo-UTP; 1-Methyl-6-ethylcarboxylate-pseudo-UTP; 1-Methyl-6-ethyl-pseudo-UTP; 1-Methyl-6-fluoro-pseudo-UTP; 1-Methyl-6-formyl-pseudo-UTP; 1-Methyl-6-hydroxyamino-pseudo-UTP; 1-Methyl-6-hydroxy-pseudo-UTP; 1-Methyl-6-iodo-pseudo-UTP; 1-Methyl-6-iso-propyl-pseudo-UTP; 1-Methyl-6-methoxy-pseudo-UTP; 1-Methyl-6-methylamino-pseudo-UTP; 1-Methyl-6-phenyl-pseudo-UTP; 1-Methyl-6-propyl-pseudo-UTP; 1-Methyl-6-tert-butyl-pseudo-UTP; 1-Methyl-6-trifluoromethoxy-pseudo-UTP; 1-Methyl-6-trifluoromethyl-pseudo-UTP; 1-Morpholinomethylpseudouridine TP; 1-Pentyl-pseudo-UTP; 1-Phenyl-pseudo-UTP; 1-Pivaloylpseudouridine TP; 1-Propargylpseudouridine TP; 1-Propyl-pseudo-UTP; 1-propynyl-pseudouridine; 1-p-tolyl-pseudo-UTP; 1-tert-Butyl-pseudo-UTP; 1-Thiomethoxymethylpseudouridine TP; 1-Thiomorpholinomethylpseudouridine TP; 1-Trifluoroacetylpseudouridine TP; 1-Trifluoromethyl-pseudo-UTP; 1-Vinylpseudouridine TP; 2,2'-anhydro-uridine TP; 2'-bromo-deoxyuridine TP; 2'-F-5-Methyl-2'-deoxy-UTP; 2'-OMe-5-Me-UTP; 2'-OMe-pseudo-UTP; 2'-a-Ethynyluridine TP; 2'-a-Trifluoromethyluridine TP; 2'-b-Ethynyluridine TP; 2'-b-Trifluoromethyluridine TP; 2'-Deoxy-2',2'-difluorouridine TP; 2'-Deoxy-2'-a-mercaptouridine

TP; 2'-Deoxy-2'-a-thiomethoxyuridine TP; 2'-Deoxy-2'-b-aminouridine TP; 2'-Deoxy-2'-b-azidouridine TP; 2'-Deoxy-2'-b-bromouridine TP; 2'-Deoxy-2'-b-chlorouridine TP; 2'-Deoxy-2'-b-fluorouridine TP; 2'-Deoxy-2'-b-iodouridine TP; 2'-Deoxy-2'-b-mercaptouridine TP; 2'-Deoxy-2'-b-thiomethoxyuridine TP; 2-methoxy-4-thio-uridine; 2-methoxyuridine; 2'-O-Methyl-5-(1-propynyl)uridine TP; 3-Alkyl-pseudo-UTP; 4'-Azidouridine TP; 4'-Carbocyclic uridine TP; 4'-Ethynyluridine TP; 5-(1-Propynyl)ara-uridine TP; 5-(2-Furanyl)uridine TP; 5-Cyanouridine TP; 5-Dimethylaminouridine TP; 5'-Homo-uridine TP; 5-iodo-2'-fluoro-deoxyuridine TP; 5-Phenylethynyluridine TP; 5-Trideuteromethyl-6-deuterouridine TP; 5-Trifluoromethyl-Uridine TP; 5-Vinylarauridine TP; 6-(2,2,2-Trifluoroethyl)-pseudo-UTP; 6-(4-Morpholino)-pseudo-UTP; 6-(4-Thiomorpholino)-pseudo-UTP; 6-(Substituted-Phenyl)-pseudo-UTP; 6-Amino-pseudo-UTP; 6-Azido-pseudo-UTP; 6-Bromo-pseudo-UTP; 6-Butyl-pseudo-UTP; 6-Chloro-pseudo-UTP; 6-Cyano-pseudo-UTP; 6-Dimethylamino-pseudo-UTP; 6-Ethoxy-pseudo-UTP; 6-Ethylcarboxylate-pseudo-UTP; 6-Ethyl-pseudo-UTP; 6-Fluoro-pseudo-UTP; 6-Formyl-pseudo-UTP; 6-Hydroxyamino-pseudo-UTP; 6-Hydroxy-pseudo-UTP; 6-Iodo-pseudo-UTP; 6-iso-Propyl-pseudo-UTP; 6-Methoxy-pseudo-UTP; 6-Methylamino-pseudo-UTP; 6-Methyl-pseudo-UTP; 6-Phenyl-pseudo-UTP; 6-Phenyl-pseudo-UTP; 6-Propyl-pseudo-UTP; 6-tert-Butyl-pseudo-UTP; 6-Trifluoromethoxy-pseudo-UTP; 6-Trifluoromethyl-pseudo-UTP; Alpha-thio-pseudo-UTP; Pseudouridine 1-(4-methylbenzenesulfonic acid) TP; Pseudouridine 1-(4-methylbenzoic acid) TP; Pseudouridine TP 1-[3-(2-ethoxy)]propionic acid; Pseudouridine TP 1-[3-{2-(2-[2-(2-ethoxy)-ethoxy]-ethoxy)-ethoxy}]propionic acid; Pseudouridine TP 1-[3-{2-(2-[2-{2(2-ethoxy)-ethoxy}-ethoxy]-ethoxy)-ethoxy}]propionic acid; Pseudouridine TP 1-[3-{2-(2-[2-ethoxy]-ethoxy)-ethoxy}]propionic acid; Pseudouridine TP 1-[3-{2-(2-ethoxy)-ethoxy}] propionic acid; Pseudouridine TP 1-methylphosphonic acid; Pseudouridine TP 1-methylphosphonic acid diethyl ester; Pseudo-UTP-N1-3-propionic acid; Pseudo-UTP-N1-4-butanoic acid; Pseudo-UTP-N1-5-pentanoic acid; Pseudo-UTP-N1-6-hexanoic acid; Pseudo-UTP-N1-7-heptanoic acid; Pseudo-UTP-N1-methyl-p-benzoic acid; Pseudo-UTP-N1-p-benzoic acid; Wybutosine; Hydroxywybutosine; Isowyosine; Peroxywybutosine; undermodified hydroxywybutosine; 4-demethylwyosine; 2,6-(diamino)purine;1-(aza)-2-(thio)-3-(aza)-phenoxazin-1-yl: 1,3-(diaza)-2-(oxo)-phenthiazin-l-yl;1,3-(diaza)-2-(oxo)-phenoxazin-1-yl;1,3,5-(triaza)-2,6-(dioxa)-naphthalene;2 (amino)purine;2,4,5-(trimethyl)phenyl;2' methyl, 2'amino, 2'azido, 2'fluro-cytidine;2' methyl, 2'amino, 2'azido, 2'fluro-adenine;2'methyl, 2'amino, 2'azido, 2'fluro-uridine;2'-amino-2'-deoxyribose; 2-amino-6-Chloro-purine; 2-aza-inosinyl; 2'-azido-2'-deoxyribose; 2'fluoro-2'-deoxyribose; 2'-fluoro-modified bases; 2'-O-methyl-ribose; 2-oxo-7-

aminopyridopyrimidin-3-yl; 2-oxo-pyridopyrimidine-3-yl; 2-pyridinone; 3 nitropyrrole; 3-(methyl)-7-(propynyl)isocarbostyrilyl; 3-(methyl)isocarbostyrilyl; 4-(fluoro)-6-(methyl)benzimidazole; 4-(methyl)benzimidazole; 4-(methyl)indolyl; 4,6-(dimethyl)indolyl; 5 nitroindole; 5 substituted pyrimidines; 5-(methyl)isocarbostyrilyl; 5-nitroindole; 6-(aza)pyrimidine; 6-(azo)thymine; 6-(methyl)-7-(aza)indolyl; 6-chloro-purine; 6-phenyl-pyrrolo-pyrimidin-2-on-3-yl; 7-(aminoalkylhydroxy)-1-(aza)-2-(thio)-3-(aza)-phenthiazin-l-yl; 7-(aminoalkylhydroxy)-1-(aza)-2-(thio)-3-(aza)-phenoxazin-1-yl; 7-(aminoalkylhydroxy)-1,3-(diaza)-2-(oxo)-phenoxazin-1-yl; 7-(aminoalkylhydroxy)-l,3-(diaza)-2-(oxo)-phenthiazin-l-yl; 7-(aminoalkylhydroxy)-l,3-(diaza)-2-(oxo)-phenoxazin-l-yl; 7-(aza)indolyl; 7-(guanidiniumalkylhydroxy)-1-(aza)-2-(thio)-3-(aza)-phenoxazinl-yl; 7-(guanidiniumalkylhydroxy)-1-(aza)-2-(thio)-3-(aza)-phenthiazin-l-yl; 7-(guanidiniumalkylhydroxy)-1-(aza)-2-(thio)-3-(aza)-phenoxazin-1-yl; 7-(guanidiniumalkylhydroxy)-1,3-(diaza)-2-(oxo)-phenoxazin-1-yl; 7-(guanidiniumalkyl-hydroxy)-l,3-(diaza)-2-(oxo)-phenthiazin-l-yl; 7-(guanidiniumalkylhydroxy)-l,3-(diaza)-2-(oxo)-phenoxazin-l-yl; 7-(propynyl)isocarbostyrilyl; 7-(propynyl)isocarbostyrilyl, propynyl-7-(aza)indolyl; 7-deaza-inosinyl; 7-substituted 1-(aza)-2-(thio)-3-(aza)-phenoxazin-1-yl; 7-substituted 1,3-(diaza)-2-(oxo)-phenoxazin-1-yl; 9-(methyl)-imidizopyridinyl; Aminoindolyl; Anthracenyl; bis-ortho-(aminoalkylhydroxy)-6-phenyl-pyrrolo-pyrimidin-2-on-3-yl; bis-ortho-substituted-6-phenyl-pyrrolo-pyrimidin-2-on-3-yl; Difluorotolyl; Hypoxanthine; Imidizopyridinyl; Inosinyl; Isocarbostyrilyl; Isoguanisine; N2-substituted purines; N6-methyl-2-amino-purine; N6-substituted purines; N-alkylated derivative; Napthalenyl; Nitrobenzimidazolyl; Nitroimidazolyl; Nitroindazolyl; Nitropyrazolyl; Nubularine; O6-substituted purines; O-alkylated derivative; ortho-(aminoalkylhydroxy)-6-phenyl-pyrrolo-pyrimidin-2-on-3-yl; ortho-substituted-6-phenyl-pyrrolo-pyrimidin-2-on-3-yl; Oxoformycin TP; para-(aminoalkylhydroxy)-6-phenyl-pyrrolo-pyrimidin-2-on-3-yl; para-substituted-6-phenyl-pyrrolo-pyrimidin-2-on-3-yl; Pentacenyl; Phenanthracenyl; Phenyl; propynyl-7-(aza)indolyl; Pyrenyl; pyridopyrimidin-3-yl; pyridopyrimidin-3-yl, 2-oxo-7-amino-pyridopyrimidin-3-yl; pyrrolo-pyrimidin-2-on-3-yl; Pyrrolopyrimidinyl; Pyrrolopyrizinyl; Stilbenzyl; substituted 1,2,4-triazoles; Tetracenyl; Tubercidine; Xanthine; Xanthosine-5'-TP; 2-thio-zebularine; 5-aza-2-thio-zebularine; 7-deaza-2-amino-purine; pyridin-4-one ribonucleoside; 2-Amino-riboside-TP; Formycin A TP; Formycin B TP; Pyrrolosine TP; 2'-OH-ara-adenosine TP; 2'-OH-ara-cytidine TP; 2'-OH-ara-uridine TP; 2'-OH-ara-guanosine TP; 5-(2-carbomethoxyvinyl)uridine TP; and N6-(19-Amino-pentaoxanonadecyl)adenosine TP.

In some embodiments, polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides) include a combination of at least two (*e.g.*, 2, 3, 4 or more) of the aforementioned modified nucleobases.

In some embodiments, modified nucleobases in polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides) are selected from the group consisting of pseudouridine (ψ), 2-thiouridine (s2U), 4'-thiouridine, 5-methylcytosine, 2-thio-1-methyl-1-deaza-pseudouridine, 2-thio-1-methyl-pseudouridine, 2-thio-5-aza-uridine, 2-thio-dihydropseudouridine, 2-thio-dihydrouridine, 2-thio-pseudouridine, 4-methoxy-2-thio-pseudouridine, 4-methoxy-pseudouridine, 4-thio-1-methyl-pseudouridine, 4-thio-pseudouridine, 5-aza-uridine, dihydropseudouridine, 5-methyluridine, 5-methoxyuridine, 2'-O-methyl uridine, 1-methyl-pseudouridine (m1ψ), 1-ethyl-pseudouridine (e1ψ), 5-methoxy-uridine (mo5U), 5-methyl-cytidine (m5C), α-thio-guanosine, α-thio-adenosine, 5-cyano uridine, 4'-thio uridine 7-deaza-adenine, 1-methyl-adenosine (m1A), 2-methyl-adenine (m2A), N6-methyl-adenosine (m6A), and 2,6-Diaminopurine, (I), 1-methyl-inosine (m1I), wyosine (imG), methylwyosine (mimG), 7-deaza-guanosine, 7-cyano-7-deaza-guanosine (preQ0), 7-aminomethyl-7-deaza-guanosine (preQ1), 7-methyl-guanosine (m7G), 1-methyl-guanosine (m1G), 8-oxo-guanosine, 7-methyl-8-oxo-guanosine, 2,8-dimethyladenosine, 2-geranylthiouridine, 2-lysidine, 2-selenouridine, 3-(3-amino-3-carboxypropyl)-5,6-dihydrouridine, 3-(3-amino-3-carboxypropyl)pseudouridine, 3-methylpseudouridine, 5-(carboxyhydroxymethyl)-2′-O-methyluridine methyl ester, 5-aminomethyl-2-geranylthiouridine, 5-aminomethyl-2-selenouridine, 5-aminomethyluridine, 5-carbamoylhydroxymethyluridine, 5-carbamoylmethyl-2-thiouridine, 5-carboxymethyl-2-thiouridine, 5-carboxymethylaminomethyl-2-geranylthiouridine, 5-carboxymethylaminomethyl-2-selenouridine, 5-cyanomethyluridine, 5-hydroxycytidine, 5-methylaminomethyl-2-geranylthiouridine, 7-aminocarboxypropyl-demethylwyosine, 7-aminocarboxypropylwyosine, 7-aminocarboxypropylwyosine methyl ester, 8-methyladenosine, N4,N4-dimethylcytidine, N6-formyladenosine, N6-hydroxymethyladenosine, agmatidine, cyclic N6-threonylcarbamoyladenosine, glutamyl-queuosine, methylated undermodified hydroxywybutosine, N4,N4,2′-O-trimethylcytidine, geranylated 5-methylaminomethyl-2-thiouridine, geranylated 5-carboxymethylaminomethyl-2-thiouridine, Qbase, preQ0base, preQ1base, and combinations of two or more thereof. In some embodiments, the at least one chemically modified nucleoside is selected from the group consisting of pseudouridine, 1-methyl-pseudouridine, 1-ethyl-pseudouridine, 5-methylcytosine, 5-methoxyuridine, and a combination thereof. In some embodiments, the

polyribonucleotide (*e.g.*, RNA polyribonucleotide, such as mRNA polyribonucleotide) includes a combination of at least two (*e.g.*, 2, 3, 4 or more) of the aforementioned modified nucleobases. In some embodiments, polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides) include a combination of at least two (*e.g.*, 2, 3, 4 or more) of the aforementioned modified nucleobases.

In some embodiments, modified nucleobases in polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides) are selected from the group consisting of 1-methyl-pseudouridine (m1ψ), 1-ethyl-pseudouridine (e1ψ), 5-methoxy-uridine (mo5U), 5-methyl-cytidine (m5C), pseudouridine (ψ), α-thio-guanosine and α-thio-adenosine. In some embodiments, the polyribonucleotide includes a combination of at least two (*e.g.*, 2, 3, 4 or more) of the aforementioned modified nucleobases, including but not limited to chemical modifications.

In some embodiments, polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides) comprise pseudouridine (ψ) and 5-methyl-cytidine (m5C). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise 1-methyl-pseudouridine (m1ψ). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise 1-ethyl-pseudouridine (e1ψ). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise 1-methyl-pseudouridine (m1ψ) and 5-methyl-cytidine (m5C). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise 1-ethyl-pseudouridine (e1ψ) and 5-methyl-cytidine (m5C). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise 2-thiouridine (s2U). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise 2-thiouridine and 5-methyl-cytidine (m5C). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise methoxy-uridine (mo5U). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise 5-methoxy-uridine (mo5U) and 5-methyl-cytidine (m5C). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise 2'-O-methyl uridine. In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise 2'-O-methyl uridine and 5-methyl-cytidine (m5C). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise N6-methyl-adenosine (m6A). In some embodiments, the polyribonucleotides (*e.g.*, RNA, such as mRNA) comprise N6-methyl-adenosine (m6A) and 5-methyl-cytidine (m5C).

In some embodiments, polynucleotides (*e.g.*, RNA polynucleotides, such as mRNA polynucleotides) are uniformly modified (*e.g.*, fully modified, modified throughout the entire sequence) with a particular modification. For example, a polynucleotide can be uniformly

modified with 1-methyl-pseudouridine, meaning that all uridine residues in the mRNA sequence are replaced with 1-methyl-pseudouridine. Similarly, a polynucleotide can be uniformly modified for any type of nucleoside residue present in the sequence by replacement with a modified residue such as those set forth above.

Exemplary nucleobases and nucleosides having a modified cytosine include N4-acetyl-cytidine (ac4C), 5-methyl-cytidine (m5C), 5-halo-cytidine (*e.g.*, 5-iodo-cytidine), 5-hydroxymethyl-cytidine (hm5C), 1-methyl-pseudoisocytidine, 2-thio-cytidine (s2C), and 2-thio-5-methyl-cytidine.

In some embodiments, a modified nucleobase is a modified uridine. Exemplary nucleobases and nucleosides having a modified uridine include 1-methyl-pseudouridine (m1ψ), 1-ethyl-pseudouridine (e1ψ), 5-methoxy uridine, 2-thio uridine, 5-cyano uridine, 2'-O-methyl uridine, and 4'-thio uridine.

In some embodiments, a modified nucleobase is a modified adenine. Exemplary nucleobases and nucleosides having a modified adenine include 7-deaza-adenine, 1-methyl-adenosine (m1A), 2-methyl-adenine (m2A), and N6-methyl-adenosine (m6A).

In some embodiments, a modified nucleobase is a modified guanine. Exemplary nucleobases and nucleosides having a modified guanine include inosine (I), 1-methyl-inosine (m1I), wyosine (imG), methylwyosine (mimG), 7-deaza-guanosine, 7-cyano-7-deaza-guanosine (preQ0), 7-aminomethyl-7-deaza-guanosine (preQ1), 7-methyl-guanosine (m7G), 1-methyl-guanosine (m1G), 8-oxo-guanosine, and 7-methyl-8-oxo-guanosine.

The polynucleotides of the present disclosure may be partially or fully modified along the entire length of the molecule. For example, one or more or all or a given type of nucleotide (*e.g.*, purine or pyrimidine, or any one or more or all of A, G, U, C) may be uniformly modified in a polynucleotide of the invention, or in a given predetermined sequence region thereof (*e.g.*, in the mRNA including or excluding the polyA tail). In some embodiments, all nucleotides X in a polynucleotide of the present disclosure (or in a given sequence region thereof) are modified nucleotides, wherein X may be any one of nucleotides A, G, U, C, or any one of the combinations A+G, A+U, A+C, G+U, G+C, U+C, A+G+U, A+G+C, G+U+C, or A+G+C.

The polynucleotide may contain from about 1% to about 100% modified nucleotides (either in relation to overall nucleotide content, or in relation to one or more types of nucleotide, *i.e.*, any one or more of A, G, U or C) or any intervening percentage (*e.g.*, from 1% to 20%, from 1% to 25%, from 1% to 50%, from 1% to 60%, from 1% to 70%, from 1% to 80%, from 1% to 90%, from 1% to 95%, from 10% to 20%, from 10% to 25%, from 10%

to 50%, from 10% to 60%, from 10% to 70%, from 10% to 80%, from 10% to 90%, from 10% to 95%, from 10% to 100%, from 20% to 25%, from 20% to 50%, from 20% to 60%, from 20% to 70%, from 20% to 80%, from 20% to 90%, from 20% to 95%, from 20% to 100%, from 50% to 60%, from 50% to 70%, from 50% to 80%, from 50% to 90%, from 50% to 95%, from 50% to 100%, from 70% to 80%, from 70% to 90%, from 70% to 95%, from 70% to 100%, from 80% to 90%, from 80% to 95%, from 80% to 100%, from 90% to 95%, from 90% to 100%, and from 95% to 100%). It will be understood that any remaining percentage is accounted for by the presence of unmodified A, G, U, or C.

The polynucleotides may contain at a minimum 1% and at maximum 100% modified nucleotides, or any intervening percentage, such as at least 5% modified nucleotides, at least 10% modified nucleotides, at least 25% modified nucleotides, at least 50% modified nucleotides, at least 80% modified nucleotides, or at least 90% modified nucleotides. For example, the polynucleotides may contain a modified pyrimidine such as a modified uracil or cytosine. In some embodiments, at least 5%, at least 10%, at least 25%, at least 50%, at least 80%, at least 90% or 100% of the uracil in the polynucleotide is replaced with a modified uracil (*e.g.*, a 5-substituted uracil). The modified uracil can be replaced by a compound having a single unique structure, or can be replaced by a plurality of compounds having different structures (*e.g.*, 2, 3, 4, or more unique structures). In some embodiments, at least 5%, at least 10%, at least 25%, at least 50%, at least 80%, at least 90%, or 100% of the cytosine in the polynucleotide is replaced with a modified cytosine (*e.g.*, a 5-substituted cytosine). The modified cytosine can be replaced by a compound having a single unique structure, or can be replaced by a plurality of compounds having different structures (*e.g.*, 2, 3, 4, or more unique structures).

Thus, in some embodiments, the RNA vaccines comprise a 5′UTR element, an optionally codon optimized open reading frame, and a 3′UTR element, a poly(A) sequence and/or a polyadenylation signal wherein the RNA is not chemically modified.

In some embodiments, the modified nucleobase is a modified uracil. Exemplary nucleobases and nucleosides having a modified uracil include pseudouridine (ψ), pyridin-4-one ribonucleoside, 5-aza-uridine, 6-aza-uridine, 2-thio-5-aza-uridine, 2-thio-uridine (s^2U), 4-thio-uridine (s^4U), 4-thio-pseudouridine, 2-thio-pseudouridine, 5-hydroxy-uridine (ho^5U), 5-aminoallyl-uridine, 5-halo-uridine (*e.g.*, 5-iodo-uridineor 5-bromo-uridine), 3-methyl-uridine (m^3U), 5-methoxy-uridine (mo^5U), uridine 5-oxyacetic acid (cmo^5U), uridine 5-oxyacetic acid methyl ester ($mcmo^5U$), 5-carboxymethyl-uridine (cm^5U), 1-carboxymethyl-pseudouridine, 5-carboxyhydroxymethyl-uridine (chm^5U), 5-carboxyhydroxymethyl-uridine

methyl ester (mchm^{5}U), 5-methoxycarbonylmethyl-uridine (mcm^{5}U), 5-methoxycarbonylmethyl-2-thio-uridine (mcm^{5}s^{2}U), 5-aminomethyl-2-thio-uridine (nm^{5}s^{2}U), 5-methylaminomethyl-uridine (mnm^{5}U), 5-methylaminomethyl-2-thio-uridine (mnm^{5}s^{2}U), 5-methylaminomethyl-2-seleno-uridine (mnm^{5}se^{2}U), 5-carbamoylmethyl-uridine (ncm^{5}U), 5-carboxymethylaminomethyl-uridine (cmnm^{5}U), 5-carboxymethylaminomethyl-2-thio-uridine (cmnm^{5}s^{2}U), 5-propynyl-uridine, 1-propynyl-pseudouridine, 5-taurinomethyl-uridine (τm^{5}U), 1-taurinomethyl-pseudouridine, 5-taurinomethyl-2-thio-uridine(τm^{5}s^{2}U), 1-taurinomethyl-4-thio-pseudouridine, 5-methyl-uridine (m^{5}U, i.e., having the nucleobase deoxythymine), 1-methyl-pseudouridine (m^{1}ψ), 1-ethyl-pseudouridine (e1ψ), 5-methyl-2-thio-uridine (m^{5}s^{2}U), 1-methyl-4-thio-pseudouridine (m^{1}s^{4}ψ), 4-thio-1-methyl-pseudouridine, 3-methyl-pseudouridine (m^{3}ψ), 2-thio-1-methyl-pseudouridine, 1-methyl-1-deaza-pseudouridine, 2-thio-1-methyl-1-deaza-pseudouridine, dihydrouridine (D), dihydropseudouridine, 5,6-dihydrouridine, 5-methyl-dihydrouridine (m^{5}D), 2-thio-dihydrouridine, 2-thio-dihydropseudouridine, 2-methoxy-uridine, 2-methoxy-4-thio-uridine, 4-methoxy-pseudouridine, 4-methoxy-2-thio-pseudouridine, N1-methyl-pseudouridine, 3-(3-amino-3-carboxypropyl)uridine (acp^{3}U), 1-methyl-3-(3-amino-3-carboxypropyl)pseudouridine (acp^{3}ψ), 5-(isopentenylaminomethyl)uridine (inm^{5}U), 5-(isopentenylaminomethyl)-2-thio-uridine (inm^{5}s^{2}U), α-thio-uridine, 2′-O-methyl-uridine (Um), 5,2′-O-dimethyl-uridine (m^{5}Um), 2′-O-methyl-pseudouridine (ψm), 2-thio-2′-O-methyl-uridine (s^{2}Um), 5-methoxycarbonylmethyl-2′-O-methyl-uridine (mcm^{5}Um), 5-carbamoylmethyl-2′-O-methyl-uridine (ncm^{5}Um), 5-carboxymethylaminomethyl-2′-O-methyl-uridine (cmnm^{5}Um), 3,2′-O-dimethyl-uridine (m^{3}Um), and 5-(isopentenylaminomethyl)-2′-O-methyl-uridine (inm^{5}Um), 1-thio-uridine, deoxythymidine, 2’-F-ara-uridine, 2’-F-uridine, 2’-OH-ara-uridine, 5-(2-carbomethoxyvinyl) uridine, and 5-[3-(1-E-propenylamino)]uridine.

In some embodiments, the modified nucleobase is a modified cytosine. Exemplary nucleobases and nucleosides having a modified cytosine include 5-aza-cytidine, 6-aza-cytidine, pseudoisocytidine, 3-methyl-cytidine (m^{3}C), N4-acetyl-cytidine (ac^{4}C), 5-formyl-cytidine (f^{5}C), N4-methyl-cytidine (m^{4}C), 5-methyl-cytidine (m^{5}C), 5-halo-cytidine (*e.g.*, 5-iodo-cytidine), 5-hydroxymethyl-cytidine (hm^{5}C), 1-methyl-pseudoisocytidine, pyrrolo-cytidine, pyrrolo-pseudoisocytidine, 2-thio-cytidine (s^{2}C), 2-thio-5-methyl-cytidine, 4-thio-pseudoisocytidine, 4-thio-1-methyl-pseudoisocytidine, 4-thio-1-methyl-1-deaza-pseudoisocytidine, 1-methyl-1-deaza-pseudoisocytidine, zebularine, 5-aza-zebularine, 5-methyl-zebularine, 5-aza-2-thio-zebularine, 2-thio-zebularine, 2-methoxy-cytidine, 2-methoxy-5-methyl-cytidine, 4-methoxy-pseudoisocytidine, 4-methoxy-1-methyl-

pseudoisocytidine, lysidine (k_2C), α-thio-cytidine, 2′-O-methyl-cytidine (Cm), 5,2′-O-dimethyl-cytidine (m^5Cm), N4-acetyl-2′-O-methyl-cytidine (ac^4Cm), N4,2′-O-dimethyl-cytidine (m^4Cm), 5-formyl-2′-O-methyl-cytidine (f^5Cm), N4,N4,2′-O-trimethyl-cytidine ($m^4{}_2Cm$), 1-thio-cytidine, 2'-F-ara-cytidine, 2'-F-cytidine, and 2'-OH-ara-cytidine.

In some embodiments, the modified nucleobase is a modified adenine. Exemplary nucleobases and nucleosides having a modified adenine include 2-amino-purine, 2, 6-diaminopurine, 2-amino-6-halo-purine (*e.g.*, 2-amino-6-chloro-purine), 6-halo-purine (*e.g.*, 6-chloro-purine), 2-amino-6-methyl-purine, 8-azido-adenosine, 7-deaza-adenine, 7-deaza-8-aza-adenine, 7-deaza-2-amino-purine, 7-deaza-8-aza-2-amino-purine, 7-deaza-2,6-diaminopurine, 7-deaza-8-aza-2,6-diaminopurine, 1-methyl-adenosine (m^1A), 2-methyl-adenine (m^2A), N6-methyl-adenosine (m^6A), 2-methylthio-N6-methyl-adenosine (ms^2m^6A), N6-isopentenyl-adenosine (i^6A), 2-methylthio-N6-isopentenyl-adenosine (ms^2i^6A), N6-(cis-hydroxyisopentenyl)adenosine (io^6A), 2-methylthio-N6-(cis-hydroxyisopentenyl)adenosine (ms^2io^6A), N6-glycinylcarbamoyl-adenosine (g^6A), N6-threonylcarbamoyl-adenosine (t^6A), N6-methyl-N6-threonylcarbamoyl-adenosine (m^6t^6A), 2-methylthio-N6-threonylcarbamoyl-adenosine (ms^2g^6A), N6,N6-dimethyl-adenosine ($m^6{}_2A$), N6-hydroxynorvalylcarbamoyl-adenosine (hn^6A), 2-methylthio-N6-hydroxynorvalylcarbamoyl-adenosine (ms^2hn^6A), N6-acetyl-adenosine (ac^6A), 7-methyl-adenine, 2-methylthio-adenine, 2-methoxy-adenine, α-thio-adenosine, 2′-O-methyl-adenosine (Am), N6,2′-O-dimethyl-adenosine (m^6Am), N6,N6,2′-O-trimethyl-adenosine ($m^6{}_2Am$), 1,2′-O-dimethyl-adenosine (m^1Am), 2′-O-ribosyladenosine (phosphate) (Ar(p)), 2-amino-N6-methyl-purine, 1-thio-adenosine, 8-azido-adenosine, 2'-F-ara-adenosine, 2'-F-adenosine, 2'-OH-ara-adenosine, and N6-(19-amino-pentaoxanonadecyl)-adenosine.

In some embodiments, the modified nucleobase is a modified guanine. Exemplary nucleobases and nucleosides having a modified guanine include inosine (I), 1-methyl-inosine (m^1I), wyosine (imG), methylwyosine (mimG), 4-demethyl-wyosine (imG-14), isowyosine (imG2), wybutosine (yW), peroxywybutosine (o_2yW), hydroxywybutosine (OhyW), undermodified hydroxywybutosine (OhyW*), 7-deaza-guanosine, queuosine (Q), epoxyqueuosine (oQ), galactosyl-queuosine (galQ), mannosyl-queuosine (manQ), 7-cyano-7-deaza-guanosine ($preQ_0$), 7-aminomethyl-7-deaza-guanosine ($preQ_1$), archaeosine (G^+), 7-deaza-8-aza-guanosine, 6-thio-guanosine, 6-thio-7-deaza-guanosine, 6-thio-7-deaza-8-aza-guanosine, 7-methyl-guanosine (m^7G), 6-thio-7-methyl-guanosine, 7-methyl-inosine, 6-methoxy-guanosine, 1-methyl-guanosine (m^1G), N2-methyl-guanosine (m^2G), N2,N2-dimethyl-guanosine ($m^2{}_2G$), N2,7-dimethyl-guanosine ($m^{2,7}G$), N2, N2,7-dimethyl-guanosine

($m^{2,2,7}G$), 8-oxo-guanosine, 7-methyl-8-oxo-guanosine, 1-methyl-6-thio-guanosine, N2-methyl-6-thio-guanosine, N2,N2-dimethyl-6-thio-guanosine, α-thio-guanosine, 2′-O-methyl-guanosine (Gm), N2-methyl-2′-O-methyl-guanosine (m^2Gm), N2,N2-dimethyl-2′-O-methyl-guanosine ($m^2{}_2Gm$), 1-methyl-2′-O-methyl-guanosine (m^1Gm), N2,7-dimethyl-2′-O-methyl-guanosine ($m^{2,7}Gm$), 2′-O-methyl-inosine (Im), 1,2′-O-dimethyl-inosine (m^1Im), 2′-O-ribosylguanosine (phosphate) (Gr(p)) , 1-thio-guanosine, O6-methyl-guanosine, 2'-F-ara-guanosine, and 2'-F-guanosine.

HSV Vaccines

In Vitro Transcription of RNA (e.g., mRNA)

HSV vaccines of the present disclosure comprise at least one RNA polynucleotide, such as a mRNA (*e.g.*, modified mRNA). mRNA, for example, is transcribed *in vitro* from template DNA, referred to as an "*in vitro* transcription template." In some embodiments, the at least one RNA polynucleotide has at least one chemical modification. The at least one chemical modification may include, but is expressly not limited to, any modification described herein.

In vitro transcription of RNA is known in the art and is described in WO/2014/152027, which is incorporated by reference herein in its entirety. For example, in some embodiments, the RNA transcript is generated using a non-amplified, linearized DNA template in an *in vitro* transcription reaction to generate the RNA transcript. In some embodiments, the RNA transcript is capped *via* enzymatic capping. In some embodiments, the RNA transcript is purified via chromatographic methods, *e.g.*, use of an oligo dT substrate. Some embodiments exclude the use of DNase. In some embodiments, the RNA transcript is synthesized from a non-amplified, linear DNA template coding for the gene of interest via an enzymatic *in vitro* transcription reaction utilizing a T7 phage RNA polymerase and nucleotide triphosphates of the desired chemistry. Any number of RNA polymerases or variants may be used in the method of the present invention. The polymerase may be selected from, but is not limited to, a phage RNA polymerase, *e.g.*, a T7 RNA polymerase, a T3 RNA polymerase, a SP6 RNa polymerase, and/or mutant polymerases such as, but not limited to, polymerases able to incorporate modified nucleic acids and/or modified nucleotides, including chemically modified nucleic acids and/or nucleotides.

In some embodiments, a non-amplified, linearized plasmid DNA is utilized as the template DNA for *in vitro* transcription. In some embodiments, the template DNA is isolated DNA. In some embodiments, the template DNA is cDNA. In some embodiments, the cDNA

is formed by reverse transcription of a RNA polynucleotide, for example, but not limited to HSV RNA, *e.g.* HSV mRNA. In some embodiments, cells, *e.g.*, bacterial cells, *e.g.*, *E. coli*, *e.g.*, DH-1 cells are transfected with the plasmid DNA template. In some embodiments, the transfected cells are cultured to replicate the plasmid DNA which is then isolated and purified. In some embodiments, the DNA template includes a RNA polymerase promoter, *e.g.*, a T7 promoter located 5 ' to and operably linked to the gene of interest.

In some embodiments, an *in vitro* transcription template encodes a 5′ untranslated (UTR) region, contains an open reading frame, and encodes a 3′ UTR and a polyA tail. The particular nucleic acid sequence composition and length of an *in vitro* transcription template will depend on the mRNA encoded by the template.

A "5′ untranslated region" (UTR) refers to a region of an mRNA that is directly upstream (*i.e.*, 5′) from the start codon (*i.e.*, the first codon of an mRNA transcript translated by a ribosome) that does not encode a polypeptide.

A "3′ untranslated region" (UTR) refers to a region of an mRNA that is directly downstream (*i.e.*, 3′) from the stop codon (*i.e.*, the codon of an mRNA transcript that signals a termination of translation) that does not encode a polypeptide.

An "open reading frame" is a continuous stretch of DNA beginning with a start codon (*e.g.*, methionine (ATG)), and ending with a stop codon (*e.g.*, TAA, TAG or TGA) and encodes a polypeptide.

A "polyA tail" is a region of mRNA that is downstream, *e.g.*, directly downstream (*i.e.*, 3′), from the 3′ UTR that contains multiple consecutive adenosine monophosphates. A polyA tail may contain 10 to 300 adenosine monophosphates. For example, a polyA tail may contain 10, 20, 30, 40, 50, 60, 70, 80, 90, 100, 110, 120, 130, 140, 150, 160, 170, 180, 190, 200, 210, 220, 230, 240, 250, 260, 270, 280, 290, or 300 adenosine monophosphates. In some embodiments, a polyA tail contains 50 to 250 adenosine monophosphates. In a relevant biological setting (*e.g.*, in cells, *in vivo*), the poly(A) tail functions to protect mRNA from enzymatic degradation, *e.g.*, in the cytoplasm, and aids in transcription termination, export of the mRNA from the nucleus, and translation.

In some embodiments, a polynucleotide includes 200 to 3,000 nucleotides. For example, a polynucleotide may include 200 to 500, 200 to 1000, 200 to 1500, 200 to 3000, 500 to 1000, 500 to 1500, 500 to 2000, 500 to 3000, 1000 to 1500, 1000 to 2000, 1000 to 3000, 1500 to 3000, or 2000 to 3000 nucleotides.

Methods of Treatment

Provided herein are compositions (*e.g.*, pharmaceutical compositions), methods, kits and reagents for prevention and/or treatment of HSV in humans and other mammals. HSV RNA (*e.g.* mRNA) vaccines can be used as therapeutic or prophylactic agents. They may be used in medicine to prevent and/or treat infectious disease. In exemplary aspects, the HSV RNA (*e.g.* mRNA) vaccines of the present disclosure are used to provide prophylactic protection from HSV. Prophylactic protection from HSV can be achieved following administration of a HSV RNA (*e.g.* mRNA) vaccine of the present disclosure. Vaccines can be administered once, twice, three times, four times or more, but it is likely sufficient to administer the vaccine once (optionally followed by a single booster). It is possible, although less desirable, to administer the vaccine to an infected individual to achieve a therapeutic response. Dosing may need to be adjusted accordingly.

In some embodiments, the HSV vaccines of the present disclosure can be used as a method of preventing a HSV infection in a subject, the method comprising administering to said subject at least one HSV vaccine of this invention. In other embodiments, the HSV vaccines of this invention can be used as a method of inhibiting a primary HSV infection in a subject, the method comprising administering to said subject at least one HSV vaccine of this invention. In other embodiments, the HSV vaccines of this invention can be used as a method of treating a HSV infection in a subject, the method comprising administering to said subject at least one HSV vaccine of this invention. In other embodiments, the HSV vaccines of this invention can be used as a method of reducing an incidence of HSV infection in a subject, the method comprising administering to said subject at least one HSV vaccine of this invention. In other embodiments, the HSV vaccines of this invention can be used as a method of inhibiting spread of HSV from a first subject infected with HSV to a second subject not infected with HSV, the method comprising administering to at least one of said first subject sand said second subject at least one HSV vaccine of this invention.

A method of eliciting an immune response in a subject against a HSV is provided in aspects of the present disclosure. The method involves administering to the subject a HSV RNA vaccine comprising at least one RNA (*e.g.* mRNA) polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide or an immunogenic fragment thereof, thereby inducing in the subject an immune response specific to HSV antigenic polypeptide or an immunogenic fragment thereof, wherein anti-antigenic polypeptide antibody titer in the subject is increased following vaccination relative to anti-antigenic polypeptide antibody titer in a subject vaccinated with a prophylactically effective dose of a

traditional vaccine against the HSV. An "anti-antigenic polypeptide antibody" is a serum antibody the binds specifically to the antigenic polypeptide.

A prophylactically effective dose is a therapeutically effective dose that prevents infection with the virus at a clinically acceptable level. In some embodiments, the therapeutically effective dose is a dose listed in a package insert for the vaccine. A traditional vaccine, as used herein, refers to a vaccine other than the RNA vaccines of the invention. For instance, a traditional vaccine includes but is not limited to live microorganism vaccines, killed microorganism vaccines, subunit vaccines, protein antigen vaccines, DNA vaccines, etc. In exemplary embodiments, a traditional vaccine is a vaccine that has achieved regulatory approval and/or is registered by a national drug regulatory body, for example the Food and Drug Administration (FDA) in the United States or the European Medicines Agency (EMA).

In some embodiments, the anti-antigenic polypeptide antibody titer in the subject is increased 1 log to 10 log following vaccination relative to anti-antigenic polypeptide antibody titer in a subject vaccinated with a prophylactically effective dose of a traditional vaccine against the HSV.

In some embodiments, the anti-antigenic polypeptide antibody titer in the subject is increased 1 log following vaccination relative to anti-antigenic polypeptide antibody titer in a subject vaccinated with a prophylactically effective dose of a traditional vaccine against the HSV.

In some embodiments, the anti-antigenic polypeptide antibody titer in the subject is increased 2 log following vaccination relative to anti-antigenic polypeptide antibody titer in a subject vaccinated with a prophylactically effective dose of a traditional vaccine against the HSV.

In some embodiments, the anti-antigenic polypeptide antibody titer in the subject is increased 3 log following vaccination relative to anti-antigenic polypeptide antibody titer in a subject vaccinated with a prophylactically effective dose of a traditional vaccine against the HSV.

In some embodiments, the anti-antigenic polypeptide antibody titer in the subject is increased 5 log following vaccination relative to anti-antigenic polypeptide antibody titer in a subject vaccinated with a prophylactically effective dose of a traditional vaccine against the HSV.

In some embodiments, the anti-antigenic polypeptide antibody titer in the subject is increased 10 log following vaccination relative to anti-antigenic polypeptide antibody titer in

a subject vaccinated with a prophylactically effective dose of a traditional vaccine against the HSV.

A method of eliciting an immune response in a subject against a HSV is provided in other aspects of the invention. The method involves administering to the subject a HSV RNA (*e.g.* mRNA) vaccine comprising at least one RNA polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide or an immunogenic fragment thereof, thereby inducing in the subject an immune response specific to HSV antigenic polypeptide or an immunogenic fragment thereof, wherein the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine against the HSV at 2 times to 100 times the dosage level relative to the RNA vaccine.

In some embodiments, the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine at twice the dosage level relative to the HSV RNA (*e.g.* mRNA) vaccine.

In some embodiments, the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine at three times the dosage level relative to the HSV RNA (*e.g.* mRNA) vaccine.

In some embodiments, the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine at 4 times the dosage level relative to the HSV RNA (*e.g.* mRNA) vaccine.

In some embodiments, the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine at 5 times the dosage level relative to the HSV RNA (*e.g.* mRNA) vaccine.

In some embodiments, the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine at 10 times the dosage level relative to the HSV RNA (*e.g.* mRNA) vaccine.

In some embodiments, the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine at 50 times the dosage level relative to the HSV RNA (*e.g.* mRNA) vaccine.

In some embodiments, the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine at 100 times the dosage level relative to the HSV RNA (*e.g.* mRNA) vaccine.

In some embodiments, the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine at 10 times to 1000 times the dosage level relative to the HSV RNA (*e.g.* mRNA) vaccine.

In some embodiments, the immune response in the subject is equivalent to an immune response in a subject vaccinated with a traditional vaccine at 100 times to 1000 times the dosage level relative to the HSV RNA (*e.g.* mRNA) vaccine.

In other embodiments, the immune response is assessed by determining anti-antigenic polypeptide antibody titer in the subject.

In other aspects, the invention is a method of eliciting an immune response in a subject against a HSV by administering to the subject a HSV RNA (*e.g.* mRNA) vaccine comprising at least one RNA (*e.g.* mRNA) polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide or an immunogenic fragment thereof, thereby inducing in the subject an immune response specific to HSV antigenic polypeptide or an immunogenic fragment thereof, wherein the immune response in the subject is induced 2 days to 10 weeks earlier relative to an immune response induced in a subject vaccinated with a prophylactically effective dose of a traditional vaccine against the HSV. In some embodiments, the immune response in the subject is induced in a subject vaccinated with a prophylactically effective dose of a traditional vaccine at 2 times to 100 times the dosage level relative to the RNA (*e.g.* mRNA) vaccine.

In some embodiments, the immune response in the subject is induced 2 days earlier relative to an immune response induced in a subject vaccinated with a prophylactically effective dose of a traditional vaccine.

In some embodiments, the immune response in the subject is induced 3 days earlier relative to an immune response induced in a subject vaccinated a prophylactically effective dose of a traditional vaccine.

In some embodiments, the immune response in the subject is induced 1 week earlier relative to an immune response induced in a subject vaccinated with a prophylactically effective dose of a traditional vaccine.

In some embodiments, the immune response in the subject is induced 2 weeks earlier relative to an immune response induced in a subject vaccinated with a prophylactically effective dose of a traditional vaccine.

In some embodiments, the immune response in the subject is induced 3 weeks earlier relative to an immune response induced in a subject vaccinated with a prophylactically effective dose of a traditional vaccine.

In some embodiments, the immune response in the subject is induced 5 weeks earlier relative to an immune response induced in a subject vaccinated with a prophylactically effective dose of a traditional vaccine.

In some embodiments, the immune response in the subject is induced 10 weeks earlier relative to an immune response induced in a subject vaccinated with a prophylactically effective dose of a traditional vaccine.

Aspects of the present disclosure further include a method of eliciting an immune response in a subject against a HSV by administering to the subject a HSV RNA (*e.g.* mRNA) vaccine having an open reading frame encoding a first antigenic polypeptide, wherein the RNA polynucleotide does not include a stabilization element, and wherein an adjuvant is not coformulated or co-administered with the vaccine.

Broad spectrum HSV vaccines

It is envisioned that there may be situations where persons are at risk for infection with more than one strain of HSV. RNA (mRNA) therapeutic vaccines are particularly amenable to combination vaccination approaches due to a number of factors including, but not limited to, speed of manufacture, ability to rapidly tailor vaccines to accommodate perceived geographical threat, and the like. Moreover, because the vaccines utilize the human body to produce the antigenic protein, the vaccines are amenable to the production of larger, more complex antigenic proteins, allowing for proper folding, surface expression, antigen presentation, *etc.* in the human subject. To protect against more than one strain of HSV, a combination vaccine can be administered that includes RNA (*e.g.* mRNA) encoding at least one antigenic polypeptide protein (or antigenic portion thereof) of a first HSV and further includes RNA (*e.g.* mRNA) encoding at least one antigenic polypeptide protein (or antigenic portion thereof) of a second HSV. RNAs (mRNAs) can be co-formulated, for example, in a single lipid nanoparticle (LNP) or can be formulated in separate LNPs destined for co-administration.

Flagellin Adjuvants

Flagellin is an approximately 500 amino acid monomeric protein that polymerizes to form the flagella associated with bacterial motion. Flagellin is expressed by a variety of flagellated bacteria (*Salmonella typhimurium* for example) as well as non-flagellated bacteria (such as *Escherichia coli*). Sensing of flagellin by cells of the innate immune system (dendritic cells, macrophages, *etc.*) is mediated by the Toll-like receptor 5 (TLR5) as well as by Nod-like receptors (NLRs) Ipaf and Naip5. TLRs and NLRs have been identified as

playing a role in the activation of innate immune response and adaptive immune response. As such, flagellin provides an adjuvant effect in a vaccine.

The nucleotide and amino acid sequences encoding known flagellin polypeptides are publicly available in the NCBI GenBank database. The flagellin sequences from *S. Typhimurium, H. Pylori, V. Cholera, S. marcesens, S. flexneri, T. Pallidum, L. pneumophila, B. burgdorferei, C. difficile, R. meliloti, A. tumefaciens, R. lupini, B. clarridgeiae, P. mirabilis, B. subtilus, L. monocytogenes, P. aeruginosa*, and *E. coli*, among others are known.

A flagellin polypeptide, as used herein, refers to a full length flagellin protein, immunogenic fragments thereof, and peptides having at least 50% sequence identity to a flagellin protein or immunogenic fragments thereof. Exemplary flagellin proteins include flagellin from *Salmonella typhi* (UniPro Entry number: Q56086), *Salmonella typhimurium* (A0A0C9DG09), *Salmonella enteritidis* (A0A0C9BAB7), and *Salmonella choleraesuis* (Q6V2X8), and SEQ ID NO: 89, 125 or 126. In some embodiments, the flagellin polypeptide has at least 60%, 70%, 75%, 80%, 90%, 95%, 97%, 98%, or 99% sequence identity to a flagellin protein or immunogenic fragments thereof (*e.g.*, SEQ ID NO: 89, 125 or 126).

In some embodiments, the flagellin polypeptide is an immunogenic fragment. An immunogenic fragment is a portion of a flagellin protein that provokes an immune response. In some embodiments, the immune response is a TLR5 immune response. An example of an immunogenic fragment is a flagellin protein in which all or a portion of a hinge region has been deleted or replaced with other amino acids. For example, an antigenic polypeptide may be inserted in the hinge region. Hinge regions are the hypervariable regions of a flagellin. Hinge regions of a flagellin are also referred to as "D3 domain or region, "propeller domain or region," "hypervariable domain or region," and "variable domain or region." "At least a portion of a hinge region," as used herein, refers to any part of the hinge region of the flagellin, or the entirety of the hinge region. In other embodiments, an immunogenic fragment of flagellin is a 20, 25, 30, 35, or 40 amino acid C-terminal fragment of flagellin.

The flagellin monomer is formed by domains D0 through D3. D0 and D1, which form the stem, are composed of tandem long alpha helices and are highly conserved among different bacteria. The D1 domain includes several stretches of amino acids that are useful for TLR5 activation. The entire D1 domain or one or more of the active regions within the domain are immunogenic fragments of flagellin. Examples of immunogenic regions within the D1 domain include residues 88-114 and residues 411-431 in *Salmonella typhimurium* FliC flagellin. Within the 13 amino acids in the 88-100 region, at least 6 substitutions are

permitted between *Salmonella* flagellin and other flagellins that still preserve TLR5 activation. Thus, immunogenic fragments of flagellin include flagellin-like sequences that activate TLR5 and contain a 13 amino acid motif that is 53% or more identical to the *Salmonella* sequence in 88-100 of FliC (LQRVRELAVQSAN; SEQ ID NO: 127).

In some embodiments, the RNA (*e.g.*, mRNA) vaccine includes an RNA that encodes a fusion protein of flagellin and one or more antigenic polypeptides. A "fusion protein" as used herein, refers to a linking of two components of the construct. In some embodiments, a carboxy-terminus of the antigenic polypeptide is fused or linked to an amino terminus of the flagellin polypeptide. In other embodiments, an amino-terminus of the antigenic polypeptide is fused or linked to a carboxy-terminus of the flagellin polypeptide. The fusion protein may include, for example, one, two, three, four, five, six or more flagellin polypeptides linked to one, two, three, four, five, six or more antigenic polypeptides. When two or more flagellin polypeptides and/or two or more antigenic polypeptides are linked such a construct may be referred to as a "multimer."

Each of the components of a fusion protein may be directly linked to one another or they may be connected through a linker. For instance, the linker may be an amino acid linker. The amino acid linker encoded for by the RNA (*e.g.*, mRNA) vaccine to link the components of the fusion protein may include, for instance, at least one member selected from the group consisting of a lysine residue, a glutamic acid residue, a serine residue, and an arginine residue. In some embodiments, the linker is 1-30, 1-25, 1-25, 5-10, 5, 15, or 5-20 amino acids in length.

In other embodiments, the RNA (*e.g.*, mRNA) vaccine includes at least two separate RNA polynucleotides, one encoding one or more antigenic polypeptides and the other encoding the flagellin polypeptide. The at least two RNA (*e.g.* mRNA) polynucleotides may be co-formulated in a carrier such as a lipid nanoparticle.

Therapeutic and Prophylactic Compositions

Provided herein are compositions (*e.g.*, pharmaceutical compositions), methods, kits and reagents for prevention, treatment or diagnosis of HSV in humans and other mammals, for example. HSV RNA (*e.g.*, mRNA) vaccines can be used as therapeutic or prophylactic agents. They may be used in medicine to prevent and/or treat infectious disease. In some embodiments, the HSV vaccines of the invention can be envisioned for use in the priming of immune effector cells, for example, to activate peripheral blood mononuclear cells (PBMCs) *ex vivo*, which are then infused (re-infused) into a subject.

In exemplary embodiments, a HSV vaccine containing RNA polynucleotides as described herein can be administered to a subject (*e.g.*, a mammalian subject, such as a human subject), and the RNA polynucleotides are translated *in vivo* to produce an antigenic polypeptide.

The HSV RNA (*e.g.*, mRNA) vaccines may be induced for translation of a polypeptide (*e.g.*, antigen or immunogen) in a cell, tissue or organism. In exemplary embodiments, such translation occurs *in vivo*, although there can be envisioned embodiments where such translation occurs *ex vivo*, in culture or *in vitro*. In exemplary embodiments, the cell, tissue, or organism is contacted with an effective amount of a composition containing a HSV RNA (*e.g.* mRNA) vaccine that contains a polynucleotide that has at least one a translatable region encoding an antigenic polypeptide.

An "effective amount" of the HSV RNA (*e.g.* mRNA) vaccine is provided based, at least in part, on the target tissue, target cell type, means of administration, physical characteristics of the polynucleotide (*e.g.*, size, and extent of modified nucleosides), and other components of the HSV RNA (*e.g.* mRNA) vaccine, and other determinants. In general, an effective amount of the HSV RNA (*e.g.* mRNA) vaccine composition provides an induced or boosted immune response as a function of antigen production in the cell. In general, an effective amount of the HSV RNA (*e.g.* mRNA) vaccine containing RNA polynucleotides having at least one chemical modifications are preferably more efficient than a composition containing a corresponding unmodified RNA polynucleotides encoding the same antigen or a peptide antigen. Increased antigen production may be demonstrated by increased cell transfection (the percentage of cells transfected with the RNA vaccine), increased protein translation from the polynucleotide, decreased nucleic acid degradation (as demonstrated, for example, by increased duration of protein translation from a modified polynucleotide), or altered antigen specific immune response of the host cell.

The term "pharmaceutical composition" refers to the combination of an active agent with a carrier, inert or active, making the composition especially suitable for diagnostic or therapeutic use *in vivo* or *ex vivo*. A "pharmaceutically acceptable carrier," after administeration to or upon a subject, does not cause undesirable physiological effects. The carrier in the pharmaceutical composition must be "acceptable" also in the sense that it is compatible with the active ingredient and can be capable of stabilizing it. One or more solubilizing agents can be utilized as pharmaceutical carriers for delivery of an active agent. Examples of a pharmaceutically acceptable carrier include, but are not limited to, biocompatible vehicles, adjuvants, additives, and diluents to achieve a composition usable as

a dosage form. Examples of other carriers include colloidal silicon oxide, magnesium stearate, cellulose, and sodium lauryl sulfate. Additional suitable pharmaceutical carriers and diluents, as well as pharmaceutical necessities for their use, are described in Remington's Pharmaceutical Sciences.

In some embodiments, RNA (*e.g.*, mRNA) vaccines (including polynucleotides their encoded polypeptides) in accordance with the present disclosure may be used for treatment of HSV.

HSV RNA (*e.g.*, mRNA) vaccines may be administered prophylactically or therapeutically as part of an active immunization scheme to healthy individuals or early in infection during the incubation phase or during active infection after onset of symptoms. In some embodiments, the amount of RNA vaccines of the present disclosure provided to a cell, a tissue or a subject may be an amount effective for immune prophylaxis.

HSV RNA (*e.g.*, mRNA) vaccines may be administrated with other prophylactic or therapeutic compounds. As a non-limiting example, a prophylactic or therapeutic compound may be an adjuvant or a booster. As used herein, when referring to a prophylactic composition, such as a vaccine, the term "booster" refers to an extra administration of the prophylactic (vaccine) composition. A booster (or booster vaccine) may be given after an earlier administration of the prophylactic composition. The time of administration between the initial administration of the prophylactic composition and the booster may be, but is not limited to, 1 minute, 2 minutes, 3 minutes, 4 minutes, 5 minutes, 6 minutes, 7 minutes, 8 minutes, 9 minutes, 10 minutes, 15 minutes, 20 minutes 35 minutes, 40 minutes, 45 minutes, 50 minutes, 55 minutes, 1 hour, 2 hours, 3 hours, 4 hours, 5 hours, 6 hours, 7 hours, 8 hours, 9 hours, 10 hours, 11 hours, 12 hours, 13 hours, 14 hours, 15 hours, 16 hours, 17 hours, 18 hours, 19 hours, 20 hours, 21 hours, 22 hours, 23 hours, 1 day, 36 hours, 2 days, 3 days, 4 days, 5 days, 6 days, 1 week, 10 days, 2 weeks, 3 weeks, 1 month, 2 months, 3 months, 4 months, 5 months, 6 months, 7 months, 8 months, 9 months, 10 months, 11 months, 1 year, 18 months, 2 years, 3 years, 4 years, 5 years, 6 years, 7 years, 8 years, 9 years, 10 years, 11 years, 12 years, 13 years, 14 years, 15 years, 16 years, 17 years, 18 years, 19 years, 20 years, 25 years, 30 years, 35 years, 40 years, 45 years, 50 years, 55 years, 60 years, 65 years, 70 years, 75 years, 80 years, 85 years, 90 years, 95 years or more than 99 years. In exemplary embodiments, the time of administration between the initial administration of the prophylactic composition and the booster may be, but is not limited to, 1 week, 2 weeks, 3 weeks, 1 month, 2 months, 3 months, 6 months, or 1 year.

In some embodiments, HSV RNA (*e.g.*, mRNA) vaccines may be administered intramuscularly or intradermally, similarly to the administration of inactivated vaccines known in the art.

The HSV RNA (*e.g.*, mRNA) vaccines may be utilized in various settings depending on the prevalence of the infection or the degree or level of unmet medical need. As a non-limiting example, the RNA vaccines may be utilized to treat and/or prevent a variety of infectious disease. RNA vaccines have superior properties in that they produce much larger antibody titers and produce responses early than commercially available anti-virals.

Provided herein are pharmaceutical compositions including HSV RNA (*e.g.*, mRNA) vaccines and RNA vaccine compositions and/or complexes optionally in combination with one or more pharmaceutically acceptable excipients.

HSV RNA (*e.g.*, mRNA) vaccines may be formulated or administered alone or in conjunction with one or more other components. For instance, HSV RNA (*e.g.* mRNA) vaccines (vaccine compositions) may comprise other components including, but not limited to, adjuvants.

In some embodiments, RNA (*e.g.*, mRNA) RNA vaccines do not include an adjuvant (they are adjuvant free).

HSV RNA (*e.g.*, mRNA) vaccines may be formulated or administered in combination with one or more pharmaceutically-acceptable excipients. In some embodiments, vaccine compositions comprise at least one additional active substances, such as, for example, a therapeutically-active substance, a prophylactically-active substance, or a combination of both. Vaccine compositions may be sterile, pyrogen-free, or both sterile and pyrogen-free. General considerations in the formulation and/or manufacture of pharmaceutical agents, such as vaccine compositions, may be found, for example, in Remington: The Science and Practice of Pharmacy 21st ed., Lippincott Williams & Wilkins, 2005 (incorporated herein by reference in its entirety).

In some embodiments, HSV RNA (*e.g.*, mRNA) vaccines are administered to humans, human patients, or subjects. For the purposes of the present disclosure, the phrase "active ingredient" generally refers to the RNA (*e.g.* mRNA) vaccines or the polynucleotides contained therein, for example, RNA polynucleotides (*e.g.*, mRNA polynucleotides) encoding antigenic polypeptides.

Formulations of the vaccine compositions described herein may be prepared by any method known or hereafter developed in the art of pharmacology. In general, such preparatory methods include the step of bringing the active ingredient (*e.g.*, mRNA

polynucleotide) into association with an excipient and/or one or more other accessory ingredients, and then, if necessary and/or desirable, dividing, shaping and/or packaging the product into a desired single- or multi-dose unit.

Relative amounts of the active ingredient, the pharmaceutically acceptable excipient, and/or any additional ingredients in a pharmaceutical composition in accordance with the disclosure will vary, depending upon the identity, size, and/or condition of the subject treated and further depending upon the route by which the composition is to be administered. By way of example, the composition may comprise between 0.1% and 100%, *e.g.*, between 0.5 and 50%, between 1-30%, between 5-80%, at least 80% (w/w) active ingredient.

HSV RNA (*e.g.*, mRNA) vaccines can be formulated using one or more excipients to: (1) increase stability; (2) increase cell transfection; (3) permit the sustained or delayed release (*e.g.*, from a depot formulation); (4) alter the biodistribution (*e.g.*, target to specific tissues or cell types); (5) increase the translation of encoded protein *in vivo*; and/or (6) alter the release profile of encoded protein (antigen) *in vivo*. In addition to traditional excipients, such as any and all solvents, dispersion media, diluents, or other liquid vehicles, dispersion or suspension aids, surface active agents, isotonic agents, thickening or emulsifying agents, preservatives, excipients can include, without limitation, lipidoids, liposomes, lipid nanoparticles, polymers, lipoplexes, core-shell nanoparticles, peptides, proteins, cells transfected with HSV RNA (*e.g.* mRNA) vaccines (*e.g.*, for transplantation into a subject), hyaluronidase, nanoparticle mimics and combinations thereof.

Stabilizing Elements

Naturally-occurring eukaryotic mRNA molecules have been found to contain stabilizing elements, including, but not limited to untranslated regions (UTR) at their 5′-end (5′UTR) and/or at their 3′-end (3′UTR), in addition to other structural features, such as a 5′-cap structure or a 3′-poly(A) tail. Both the 5′UTR and the 3′UTR are typically transcribed from the genomic DNA and are elements of the premature mRNA. Characteristic structural features of mature mRNA, such as the 5′-cap and the 3′-poly(A) tail, are usually added to the transcribed (premature) mRNA during mRNA processing. The 3′-poly(A) tail is typically a stretch of adenine nucleotides added to the 3′-end of the transcribed mRNA. It can comprise up to about 400 adenine nucleotides. In some embodiments, the length of the 3′-poly(A) tail may be an essential element with respect to the stability of the individual mRNA.

In some embodiments, the RNA vaccine may include one or more stabilizing elements. Stabilizing elements may include, for instance, a histone stem-loop. A stem-loop

binding protein (SLBP), a 32 kDa protein, has been identified. It is associated with the histone stem-loop at the 3'-end of the histone messages in both the nucleus and the cytoplasm. Its expression level is regulated by the cell cycle; it is peaks during the S-phase, when histone mRNA levels are also elevated. The protein has been shown to be essential for efficient 3'-end processing of histone pre-mRNA by the U7 snRNP. SLBP continues to be associated with the stem-loop after processing, and then stimulates the translation of mature histone mRNAs into histone proteins in the cytoplasm. The RNA binding domain of SLBP is conserved through metazoa and protozoa; its binding to the histone stem-loop depends on the structure of the loop. The minimum binding site includes at least three nucleotides 5' and two nucleotides 3' relative to the stem-loop.

In some embodiments, the RNA vaccines include a coding region, at least one histone stem-loop, and optionally, a poly(A) sequence or polyadenylation signal. The poly(A) sequence or polyadenylation signal generally should enhance the expression level of the encoded protein. The encoded protein, in some embodiments, is not a histone protein, a reporter protein (*e.g.* Luciferase, GFP, EGFP, β-Galactosidase, EGFP), or a marker or selection protein (*e.g.* alpha-Globin, Galactokinase and Xanthine:guanine phosphoribosyl transferase (GPT)).

In some embodiments, the combination of a poly(A) sequence or polyadenylation signal and at least one histone stem-loop, even though both represent alternative mechanisms in nature, acts synergistically to increase the protein expression beyond the level observed with either of the individual elements. It has been found that the synergistic effect of the combination of poly(A) and at least one histone stem-loop does not depend on the order of the elements or the length of the poly(A) sequence.

In some embodiments, the RNA vaccine does not comprise a histone downstream element (HDE). "Histone downstream element" (HDE) includes a purine-rich polynucleotide stretch of approximately 15 to 20 nucleotides 3′ of naturally occurring stem-loops, representing the binding site for the U7 snRNA, which is involved in processing of histone pre-mRNA into mature histone mRNA. Ideally, the inventive nucleic acid does not include an intron.

In some embodiments, the RNA vaccine may or may not contain an enhancer and/or promoter sequence, which may be modified or unmodified or which may be activated or inactivated. In some embodiments, the histone stem-loop is generally derived from histone genes, and includes an intramolecular base pairing of two neighbored partially or entirely reverse complementary sequences separated by a spacer, consisting of a short sequence,

which forms the loop of the structure. The unpaired loop region is typically unable to base pair with either of the stem loop elements. It occurs more often in RNA, as is a key component of many RNA secondary structures, but may be present in single-stranded DNA as well. Stability of the stem-loop structure generally depends on the length, number of mismatches or bulges, and base composition of the paired region. In some embodiments, wobble base pairing (non-Watson-Crick base pairing) may result. In some embodiments, the at least one histone stem-loop sequence comprises a length of 15 to 45 nucleotides.

In other embodiments, the RNA vaccine may have one or more AU-rich sequences removed. These sequences, sometimes referred to as AURES, are destabilizing sequences found in the 3'UTR. The AURES may be removed from the RNA vaccines. Alternatively, the AURES may remain in the RNA vaccine.

Nanoparticle Formulations

In some embodiments, HSV RNA (*e.g.*, mRNA) vaccines are formulated in a nanoparticle. In some embodiments, HSV RNA (*e.g.* mRNA) vaccines are formulated in a lipid nanoparticle. In some embodiments, HSV RNA (*e.g.* mRNA) vaccines are formulated in a lipid-polycation complex, referred to as a cationic lipid nanoparticle. The formation of the lipid nanoparticle may be accomplished by methods known in the art and/or as described in U.S. Publication No. 20120178702, herein incorporated by reference in its entirety. As a non-limiting example, the polycation may include a cationic peptide or a polypeptide such as, but not limited to, polylysine, polyornithine and/or polyarginine and the cationic peptides described in International Publication No. WO2012013326 or U.S. Publication No. US20130142818; each of which is herein incorporated by reference in its entirety. In some embodiments, HSV RNA (*e.g.* mRNA) vaccines are formulated in a lipid nanoparticle that includes a non-cationic lipid such as, but not limited to, cholesterol or dioleoyl phosphatidylethanolamine (DOPE).

A lipid nanoparticle formulation may be influenced by, but not limited to, the selection of the cationic lipid component, the degree of cationic lipid saturation, the nature of the PEGylation, ratio of all components, and biophysical parameters such as size. In one example by Semple *et al.* (*Nature Biotech.* 2010 28:172-176; herein incorporated by reference in its entirety), the lipid nanoparticle formulation is composed of 57.1 % cationic lipid, 7.1% dipalmitoylphosphatidylcholine, 34.3 % cholesterol, and 1.4% PEG-c-DMA. As another example, changing the composition of the cationic lipid was shown to more

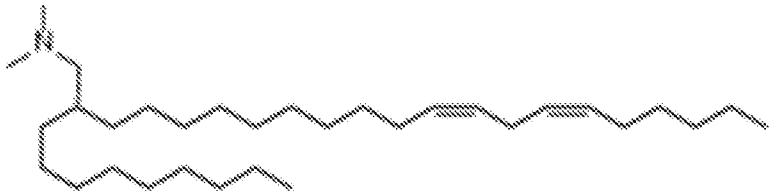
effectively deliver siRNA to various antigen presenting cells (Basha *et al. Mol Ther.* 2011 19:2186-2200; herein incorporated by reference in its entirety).

In some embodiments, lipid nanoparticle formulations may comprise 35% to 45% cationic lipid, 40% to 50% cationic lipid, 50% to 60% cationic lipid and/or 55% to 65% cationic lipid. In some embodiments, the ratio of lipid to RNA (*e.g.*, mRNA) in lipid nanoparticles may be 5:1 to 20:1, 10:1 to 25:1, 15:1 to 30:1, and/or at least 30:1.

In some embodiments, the ratio of PEG in the lipid nanoparticle formulations may be increased or decreased and/or the carbon chain length of the PEG lipid may be modified from C14 to C18 to alter the pharmacokinetics and/or biodistribution of the lipid nanoparticle formulations. As a non-limiting example, lipid nanoparticle formulations may contain 0.5% to 3.0%, 1.0% to 3.5%, 1.5% to 4.0%, 2.0% to 4.5%, 2.5% to 5.0%, and/or 3.0% to 6.0% of the lipid molar ratio of PEG-c-DOMG (R-3-[(ω-methoxy-poly(ethyleneglycol)2000)carbamoyl)]-1,2-dimyristyloxypropyl-3-amine) (also referred to herein as PEG-DOMG) as compared to the cationic lipid, DSPC, and cholesterol. In some embodiments, the PEG-c-DOMG may be replaced with a PEG lipid such as, but not limited to, PEG- DSG (1,2-Distearoyl-sn-glycerol, methoxypolyethylene glycol), PEG-DMG (1,2-Dimyristoyl-sn-glycerol) and/or PEG-DPG (1,2-Dipalmitoyl-sn-glycerol, methoxypolyethylene glycol). The cationic lipid may be selected from any lipid known in the art such as, but not limited to, DLin-MC3-DMA, DLin-DMA, C12-200, and DLin-KC2-DMA.

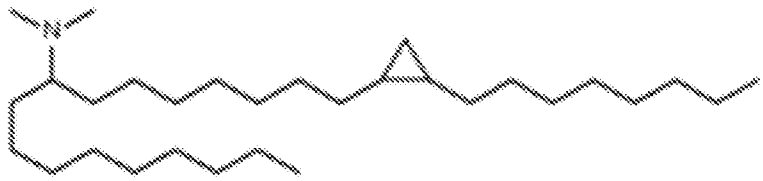
In some embodiments, a HSV RNA (*e.g.*, mRNA) vaccine formulation is a nanoparticle that comprises at least one lipid. The lipid may be selected from, but is not limited to, DLin-DMA, DLin-K-DMA, 98N12-5, C12-200, DLin-MC3-DMA, DLin-KC2-DMA, DODMA, PLGA, PEG, PEG-DMG, (12Z,15Z)-N,N-dimethyl-2-nonylhenicosa-12,15-dien-1-amine (L608), N,N-dimethyl-1-[(1S,2R)-2-octylcyclopropyl]heptadecan-8-amine (L530), PEGylated lipids, and amino alcohol lipids.

In some embodiments, the lipid is



(L608).

In some embodiments, the lipid is



(L530).

In some embodiments, the lipid may be a cationic lipid such as, but not limited to, DLin-DMA, DLin-D-DMA, DLin-MC3-DMA, DLin-KC2-DMA, DODMA, and amino alcohol lipids. The amino alcohol cationic lipid may be the lipids described in and/or made by the methods described in U.S. Publication No. US20130150625, herein incorporated by reference in its entirety. As a non-limiting example, the cationic lipid may be 2-amino-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]-2-{[(9Z,2Z)-octadeca-9,12-dien-1-yloxy]methyl}propan-1-ol (Compound 1 in US20130150625); 2-amino-3-[(9Z)-octadec-9-en-1-yloxy]-2-{[(9Z)-octadec-9-en-1-yloxy]methyl}propan-1-ol (Compound 2 in US20130150625); 2-amino-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]-2-[(octyloxy)methyl]propan-1-ol (Compound 3 in US20130150625); and 2-(dimethylamino)-3-[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]-2-{[(9Z,12Z)-octadeca-9,12-dien-1-yloxy]methyl}propan-1-ol (Compound 4 in US20130150625); or any pharmaceutically acceptable salt or stereoisomer thereof.

Lipid nanoparticle formulations typically comprise a lipid, in particular, an ionizable cationic lipid, for example, 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), or di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), and further comprise a neutral lipid, a sterol and a molecule capable of reducing particle aggregation, for example a PEG or PEG-modified lipid.

In some embodiments, a lipid nanoparticle formulation consists essentially of (i) at least one lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319); (ii) a neutral lipid selected from DSPC, DPPC, POPC, DOPE and SM; (iii) a sterol, *e.g.*, cholesterol; and (iv) a PEG-lipid, *e.g.*, PEG-DMG or PEG-cDMA, in a molar ratio of 20-60% cationic lipid: 5-25% neutral lipid: 25-55% sterol: 0.5-15% PEG-lipid.

In some embodiments, a lipid nanoparticle formulation includes 25% to 75% on a molar basis of a cationic lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-

dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), *e.g.*, 35% to 65%, 45% to 65%, 60%, 57.5%, 50% or 40% on a molar basis.

In some embodiments, a lipid nanoparticle formulation includes 0.5% to 15% on a molar basis of the neutral lipid, *e.g.*, 3% to 12%, 5% to 10% or 15%, 10%, or 7.5% on a molar basis. Examples of neutral lipids include, without limitation, DSPC, POPC, DPPC, DOPE, and SM. In some embodiments, the formulation includes 5% to 50% on a molar basis of the sterol (*e.g.*, 15% to 45%, 20% to 40%, 40%, 38.5%, 35%, or 31% on a molar basis. A non-limiting example of a sterol is cholesterol. In some embodiments, a lipid nanoparticle formulation includes 0.5% to 20% on a molar basis of the PEG or PEG-modified lipid (*e.g.*, 0.5% to 10%, 0.5% to 5%, 1.5%, 0.5%, 1.5%, 3.5%, or 5% on a molar basis. In some embodiments, a PEG or PEG modified lipid comprises a PEG molecule of an average molecular weight of 2,000 Da. In some embodiments, a PEG or PEG modified lipid comprises a PEG molecule of an average molecular weight of less than 2,000, for example around 1,500 Da, around 1,000 Da, or around 500 Da. Non-limiting examples of PEG-modified lipids include PEG-distearoyl glycerol (PEG-DMG) (also referred herein as PEG-C14 or C14-PEG), and PEG-cDMA (further discussed in Reyes *et al. J. Controlled Release*, 107, 276-287 (2005) the content of which is herein incorporated by reference in its entirety).

In some embodiments, lipid nanoparticle formulations include 25-75% of a cationic lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), 0.5-15% of the neutral lipid, 5-50% of the sterol, and 0.5-20% of the PEG or PEG-modified lipid on a molar basis.

In some embodiments, lipid nanoparticle formulations include 35-65% of a cationic lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), 3-12% of the neutral lipid, 15-45% of the sterol, and 0.5-10% of the PEG or PEG-modified lipid on a molar basis.

In some embodiments, lipid nanoparticle formulations include 45-65% of a cationic lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate

(L319), 5-10% of the neutral lipid, 25-40% of the sterol, and 0.5-10% of the PEG or PEG-modified lipid on a molar basis.

In some embodiments, lipid nanoparticle formulations include 60% of a cationic lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), 7.5% of the neutral lipid, 31% of the sterol, and 1.5% of the PEG or PEG-modified lipid on a molar basis.

In some embodiments, lipid nanoparticle formulations include 50% of a cationic lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), 10% of the neutral lipid, 38.5% of the sterol, and 1.5% of the PEG or PEG-modified lipid on a molar basis.

In some embodiments, lipid nanoparticle formulations include 50% of a cationic lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), 10% of the neutral lipid, 35% of the sterol, 4.5% or 5% of the PEG or PEG-modified lipid, and 0.5% of the targeting lipid on a molar basis.

In some embodiments, lipid nanoparticle formulations include 40% of a cationic lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), 15% of the neutral lipid, 40% of the sterol, and 5% of the PEG or PEG-modified lipid on a molar basis.

In some embodiments, lipid nanoparticle formulations include 57.2% of a cationic lipid selected from the group consisting of 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319), 7.1% of the neutral lipid, 34.3% of the sterol, and 1.4% of the PEG or PEG-modified lipid on a molar basis.

In some embodiments, lipid nanoparticle formulations include 57.5% of a cationic lipid selected from the PEG lipid is PEG-cDMA (PEG-cDMA is further discussed in Reyes *et*

al. (*J. Controlled Release*, 107, 276-287 (2005), the content of which is herein incorporated by reference in its entirety), 7.5% of the neutral lipid, 31.5% of the sterol, and 3.5% of the PEG or PEG-modified lipid on a molar basis.

In some embodiments, lipid nanoparticle formulations consist essentially of a lipid mixture in molar ratios of 20-70% cationic lipid: 5-45% neutral lipid: 20-55% cholesterol: 0.5-15% PEG-modified lipid. In some embodiments, lipid nanoparticle formulations consist essentially of a lipid mixture in a molar ratio of 20-60% cationic lipid: 5-25% neutral lipid: 25-55% cholesterol: 0.5-15% PEG-modified lipid.

In some embodiments, the molar lipid ratio is 50/10/38.5/1.5 (mol% cationic lipid/neutral lipid, *e.g.*, DSPC/Chol/PEG-modified lipid, *e.g.*, PEG-DMG, PEG-DSG or PEG-DPG), 57.2/7.1134.3/1.4 (mol% cationic lipid/ neutral lipid, *e.g.*, DPPC/Chol/ PEG-modified lipid, *e.g.*, PEG-cDMA), 40/15/40/5 (mol% cationic lipid/ neutral lipid, *e.g.*, DSPC/Chol/ PEG-modified lipid, *e.g.*, PEG-DMG), 50/10/35/4.5/0.5 (mol% cationic lipid/ neutral lipid, *e.g.*, DSPC/Chol/ PEG-modified lipid, *e.g.*, PEG-DSG), 50/10/35/5 (cationic lipid/ neutral lipid, *e.g.*, DSPC/Chol/ PEG-modified lipid, *e.g.*, PEG-DMG), 40/10/40/10 (mol% cationic lipid/ neutral lipid, *e.g.*, DSPC/Chol/ PEG-modified lipid, *e.g.*, PEG-DMG or PEG-cDMA), 35/15/40/10 (mol% cationic lipid/ neutral lipid, *e.g.*, DSPC/Chol/ PEG-modified lipid, *e.g.*, PEG-DMG or PEG-cDMA), or 52/13/30/5 (mol% cationic lipid/ neutral lipid, *e.g.*, DSPC/Chol/ PEG-modified lipid, *e.g.*, PEG-DMG or PEG-cDMA).

Non-limiting examples of lipid nanoparticle compositions and methods of making them are described, for example, in Semple *et al.* (2010) *Nat. Biotechnol.* 28:172-176; Jayarama *et al.* (2012), *Angew. Chem. Int. Ed.,* 51: 8529–8533; and Maier *et al.* (2013) *Molecular Therapy* 21, 1570-1578 (the contents of each of which are incorporated herein by reference in their entirety).

In some embodiments, lipid nanoparticle formulations may comprise a cationic lipid, a PEG lipid, and a structural lipid, and optionally comprise a non-cationic lipid. As a non-limiting example, a lipid nanoparticle may comprise 40-60% of a cationic lipid, 5-15% of a non-cationic lipid, 1-2% of a PEG lipid and 30-50% of a structural lipid. As another non-limiting example, the lipid nanoparticle may comprise 50% cationic lipid, 10% non-cationic lipid, 1.5% PEG lipid and 38.5% structural lipid. As yet another non-limiting example, a lipid nanoparticle may comprise 55% cationic lipid, 10% non-cationic lipid, 2.5% PEG lipid and 32.5% structural lipid. In some embodiments, the cationic lipid may be any cationic lipid described herein such as, but not limited to, DLin-KC2-DMA, DLin-MC3-DMA, and L319.

In some embodiments, the lipid nanoparticle formulations described herein may be 4 component lipid nanoparticles. The lipid nanoparticle may comprise a cationic lipid, a non-cationic lipid, a PEG lipid and a structural lipid. As a non-limiting example, the lipid nanoparticle may comprise 40-60% of a cationic lipid, 5-15% of a non-cationic lipid, 1-2% of a PEG lipid, and 30-50% of a structural lipid. As another non-limiting example, the lipid nanoparticle may comprise 50% cationic lipid, 10% non-cationic lipid, 1.5% PEG lipid, and 38.5% structural lipid. As yet another non-limiting example, the lipid nanoparticle may comprise 55% cationic lipid, 10% non-cationic lipid, 2.5% PEG lipid, and 32.5% structural lipid. In some embodiments, the cationic lipid may be any cationic lipid described herein such as, but not limited to, DLin-KC2-DMA, DLin-MC3-DMA, and L319.

In some embodiments, the lipid nanoparticle formulations described herein may comprise a cationic lipid, a non-cationic lipid, a PEG lipid and a structural lipid. As a non-limiting example, the lipid nanoparticle may comprise 50% of the cationic lipid DLin-KC2-DMA, 10% of the non-cationic lipid DSPC, 1.5% of the PEG lipid PEG-DOMG and 38.5% of the structural lipid cholesterol. As a non-limiting example, the lipid nanoparticle may comprise 50% of the cationic lipid DLin-MC3-DMA, 10% of the non-cationic lipid DSPC, 1.5% of the PEG lipid PEG-DOMG and 38.5% of the structural lipid cholesterol. As a non-limiting example, the lipid nanoparticle may comprise 50% of the cationic lipid DLin-MC3-DMA, 10% of the non-cationic lipid DSPC, 1.5% of the PEG lipid PEG-DMG and 38.5% of the structural lipid cholesterol. As yet another non-limiting example, the lipid nanoparticle may comprise 55% of the cationic lipid L319, 10% of the non-cationic lipid DSPC, 2.5% of the PEG lipid PEG-DMG and 32.5% of the structural lipid cholesterol.

Relative amounts of the active ingredient, the pharmaceutically acceptable excipient, and/or any additional ingredients in a vaccine composition may vary, depending upon the identity, size, and/or condition of the subject being treated and further depending upon the route by which the composition is to be administered. For example, the composition may comprise between 0.1% and 99% (w/w) of the active ingredient. By way of example, the composition may comprise between 0.1% and 100%, *e.g.*, between 0.5 and 50%, between 1-30%, between 5-80%, at least 80% (w/w) active ingredient.

In some embodiments, the RNA vaccine composition may comprise the polynucleotide described herein, formulated in a lipid nanoparticle comprising MC3, Cholesterol, DSPC and PEG2000-DMG, the buffer trisodium citrate, sucrose and water for injection. As a non-limiting example, the composition comprises: 2.0 mg/mL of drug substance (*e.g.*, polynucleotides encoding HSV), 21.8 mg/mL of MC3, 10.1 mg/mL of

cholesterol, 5.4 mg/mL of DSPC, 2.7 mg/mL of PEG2000-DMG, 5.16 mg/mL of trisodium citrate, 71 mg/mL of sucrose and 1.0 mL of water for injection.

In some embodiments, a nanoparticle (*e.g.*, a lipid nanoparticle) has a mean diameter of 10-500 nm, 20-400 nm, 30-300 nm, or 40-200 nm. In some embodiments, a nanoparticle (*e.g.*, a lipid nanoparticle) has a mean diameter of 50-150 nm, 50-200 nm, 80-100 nm, or 80-200 nm.

Liposomes, Lipoplexes, and Lipid Nanoparticles

In some embodiments, the RNA vaccine pharmaceutical compositions may be formulated in liposomes such as, but not limited to, DiLa2 liposomes (Marina Biotech, Bothell, WA), SMARTICLES® (Marina Biotech, Bothell, WA), neutral DOPC (1,2-dioleoyl-sn-glycero-3-phosphocholine) based liposomes (*e.g.*, siRNA delivery for ovarian cancer (Landen *et al. Cancer Biology & Therapy* 2006 5(12)1708-1713); herein incorporated by reference in its entirety) and hyaluronan-coated liposomes (Quiet Therapeutics, Israel).

In some embodiments, the RNA vaccines may be formulated in a lyophilized gel-phase liposomal composition as described in U.S. Publication No. US2012060293, herein incorporated by reference in its entirety.

The nanoparticle formulations may comprise a phosphate conjugate. The phosphate conjugate may increase *in vivo* circulation times and/or increase the targeted delivery of the nanoparticle. Phosphate conjugates for use with the present invention may be made by the methods described in International Publication No. WO2013033438 or U.S. Publication No. US20130196948, the content of each of which is herein incorporated by reference in its entirety. As a non-limiting example, the phosphate conjugates may include a compound of any one of the formulas described in International Publication No. WO2013033438, herein incorporated by reference in its entirety.

The nanoparticle formulation may comprise a polymer conjugate. The polymer conjugate may be a water-soluble conjugate. The polymer conjugate may have a structure as described in U.S. Publication No. 20130059360, the content of which is herein incorporated by reference in its entirety. In some aspects, polymer conjugates with the polynucleotides of the present invention may be made using the methods and/or segmented polymeric reagents described in U.S. Publication No. 20130072709, herein incorporated by reference in its entirety. In other aspects, the polymer conjugate may have pendant side groups comprising ring moieties such as, but not limited to, the polymer conjugates described in U.S. Publication No. US20130196948, the contents of which is herein incorporated by reference in its entirety.

The nanoparticle formulations may comprise a conjugate to enhance the delivery of nanoparticles of the present invention in a subject. Further, the conjugate may inhibit phagocytic clearance of the nanoparticles in a subject. In some aspects, the conjugate may be a "self" peptide designed from the human membrane protein CD47 (*e.g.*, the "self" particles described by Rodriguez *et al.* (*Science* 2013, 339, 971-975), herein incorporated by reference in its entirety). As shown by Rodriguez *et al.*, the self peptides delayed macrophage-mediated clearance of nanoparticles which enhanced delivery of the nanoparticles. In other aspects, the conjugate may be the membrane protein CD47 (*e.g.*, *see* Rodriguez *et al. Science* 2013, 339, 971-975, herein incorporated by reference in its entirety). Rodriguez *et al.* showed that, similarly to "self" peptides, CD47 can increase the circulating particle ratio in a subject as compared to scrambled peptides and PEG coated nanoparticles.

In some embodiments, the RNA (*e.g.* mRNA) vaccines of the present invention are formulated in nanoparticles which comprise a conjugate to enhance the delivery of the nanoparticles of the present invention in a subject. The conjugate may be the CD47 membrane or the conjugate may be derived from the CD47 membrane protein, such as the "self" peptide described previously. In other embodiments, the nanoparticle may comprise PEG and a conjugate of CD47 or a derivative thereof. In yet other embodiments, the nanoparticle may comprise both the "self" peptide described above and the membrane protein CD47.

In some embodiments, a "self" peptide and/or CD47 protein may be conjugated to a virus-like particle or pseudovirion, as described herein for delivery of the RNA (*e.g.* mRNA) vaccines of the present invention.

In other embodiments, RNA (*e.g.* mRNA) vaccine pharmaceutical compositions comprise the polynucleotides of the present invention and a conjugate, which may have a degradable linkage. Non-limiting examples of conjugates include an aromatic moiety comprising an ionizable hydrogen atom, a spacer moiety, and a water-soluble polymer. As a non-limiting example, pharmaceutical compositions comprising a conjugate with a degradable linkage and methods for delivering such pharmaceutical compositions are described in U.S. Publication No. US20130184443, the content of which is herein incorporated by reference in its entirety.

The nanoparticle formulations may be a carbohydrate nanoparticle comprising a carbohydrate carrier and a RNA (*e.g.* mRNA) vaccine. As a non-limiting example, the carbohydrate carrier may include, but is not limited to, an anhydride-modified phytoglycogen or glycogen-type material, phtoglycogen octenyl succinate, phytoglycogen beta-dextrin, or

anhydride-modified phytoglycogen beta-dextrin. (*See e.g.*, International Publication No. WO2012109121, the content of which is herein incorporated by reference in its entirety).

Nanoparticle formulations of the present invention may be coated with a surfactant or polymer in order to improve the delivery of the particle. In some embodiments, the nanoparticle may be coated with a hydrophilic coating such as, but not limited to, PEG coatings and/or coatings that have a neutral surface charge. The hydrophilic coatings may help to deliver nanoparticles with larger payloads such as, but not limited to, RNA (*e.g.* mRNA) vaccines, within the central nervous system. As a non-limiting example nanoparticles comprising a hydrophilic coating and methods of making such nanoparticles are described in U.S. Publication No. US20130183244, the content of which is herein incorporated by reference in its entirety.

In some embodiments, the lipid nanoparticles of the present invention may be hydrophilic polymer particles. Non-limiting examples of hydrophilic polymer particles and methods of making hydrophilic polymer particles are described in U.S. Publication No. US20130210991, the content of which is herein incorporated by reference in its entirety.

In other embodiments, the lipid nanoparticles of the present invention may be hydrophobic polymer particles.

Lipid nanoparticle formulations may be improved by replacing the cationic lipid with a biodegradable cationic lipid which is known as a rapidly eliminated lipid nanoparticle (reLNP). Ionizable cationic lipids, such as, but not limited to, DLinDMA, DLin-KC2-DMA, and DLin-MC3-DMA, have been shown to accumulate in plasma and tissues over time and may be a potential source of toxicity. The rapid metabolism of the rapidly eliminated lipids can improve the tolerability and therapeutic index of the lipid nanoparticles by an order of magnitude from a 1 mg/kg dose to a 10 mg/kg dose in rat. Inclusion of an enzymatically degraded ester linkage can improve the degradation and metabolism profile of the cationic component, while still maintaining the activity of the reLNP formulation. The ester linkage can be internally located within the lipid chain or it may be terminally located at the terminal end of the lipid chain. The internal ester linkage may replace any carbon in the lipid chain.

In some embodiments, the internal ester linkage may be located on either side of the saturated carbon.

In some embodiments, an immune response may be elicited by delivering a lipid nanoparticle which may include a nanospecies, a polymer and an immunogen. (U.S. Publication No. 20120189700 and International Publication No. WO2012099805, each of which is herein incorporated by reference in its entirety).

The polymer may encapsulate the nanospecies or partially encapsulate the nanospecies. The immunogen may be a recombinant protein, a modified RNA and/or a polynucleotide described herein. In some embodiments, the lipid nanoparticle may be formulated for use in a vaccine such as, but not limited to, against a pathogen.

Lipid nanoparticles may be engineered to alter the surface properties of particles so the lipid nanoparticles may penetrate the mucosal barrier. Mucus is located on mucosal tissue such as, but not limited to, oral (*e.g.*, the buccal and esophageal membranes and tonsil tissue), ophthalmic, gastrointestinal (*e.g.*, stomach, small intestine, large intestine, colon, rectum), nasal, respiratory (*e.g.*, nasal, pharyngeal, tracheal and bronchial membranes), and genital (*e.g.*, vaginal, cervical and urethral membranes). Nanoparticles larger than 10-200 nm, which are preferred for higher drug encapsulation efficiency and the ability to provide the sustained delivery of a wide array of drugs, have been thought to be too large to rapidly diffuse through mucosal barriers. Mucus is continuously secreted, shed, discarded or digested, and recycled so most of the trapped particles may be removed from the mucosal tissue within seconds or within a few hours. Large polymeric nanoparticles (200 nm to 500 nm in diameter) which have been coated densely with a low molecular weight polyethylene glycol (PEG) diffused through mucus only 4- to 6-fold lower than the same particles diffusing in water (Lai *et al. PNAS* 2007 104(5):1482-487; Lai *et al. Adv Drug Deliv Rev.* 2009 61(2): 158-171; each of which is herein incorporated by reference in its entirety). The transport of nanoparticles may be determined using rates of permeation and/or fluorescent microscopy techniques including, but not limited to, fluorescence recovery after photobleaching (FRAP) and high resolution multiple particle tracking (MPT). As a non-limiting example, compositions which can penetrate a mucosal barrier may be made as described in U.S. Patent No. 8,241,670 or International Publication No. WO2013110028, the content of each of which is herein incorporated by reference in its entirety.

The lipid nanoparticle engineered to penetrate mucus may comprise a polymeric material (*e.g.*, a polymeric core) and/or a polymer-vitamin conjugate and/or a tri-block co-polymer. The polymeric material may include, but is not limited to, polyamines, polyethers, polyamides, polyesters, polycarbamates, polyureas, polycarbonates, poly(styrenes), polyimides, polysulfones, polyurethanes, polyacetylenes, polyethylenes, polyethyeneimines, polyisocyanates, polyacrylates, polymethacrylates, polyacrylonitriles, and polyarylates. The polymeric material may be biodegradable and/or biocompatible. Non-limiting examples of biocompatible polymers are described in International Publication No. WO2013116804, the content of which is herein incorporated by reference in its entirety. The polymeric material may additionally be irradiated.

As a non-limiting example, the polymeric material may be gamma irradiated (*see e.g.*, International Publication No. WO201282165, herein incorporated by reference in its entirety). Non-limiting examples of specific polymers include poly(caprolactone) (PCL), ethylene vinyl acetate polymer (EVA), poly(lactic acid) (PLA), poly(L-lactic acid) (PLLA), poly(glycolic acid) (PGA), poly(lactic acid-co-glycolic acid) (PLGA), poly(L-lactic acid-co-glycolic acid) (PLLGA), poly(D,L-lactide) (PDLA), poly(L-lactide) (PLLA), poly(D,L-lactide-co-caprolactone), poly(D,L-lactide-co-caprolactone-co-glycolide), poly(D,L-lactide-co-PEO-co-D,L-lactide), poly(D,L-lactide-co-PPO-co-D,L-lactide), polyalkyl cyanoacralate, polyurethane, poly-L-lysine (PLL), hydroxypropyl methacrylate (HPMA), polyethyleneglycol, poly-L-glutamic acid, poly(hydroxy acids), polyanhydrides, polyorthoesters, poly(ester amides), polyamides, poly(ester ethers), polycarbonates, polyalkylenes such as polyethylene and polypropylene, polyalkylene
glycols such as poly(ethylene glycol) (PEG), polyalkylene oxides (PEO), polyalkylene terephthalates such as poly(ethylene terephthalate), polyvinyl alcohols (PVA), polyvinyl ethers, polyvinyl esters such as poly(vinyl acetate), polyvinyl halides such as poly(vinyl chloride) (PVC), polyvinylpyrrolidone, polysiloxanes, polystyrene (PS), polyurethanes, derivatized celluloses such as alkyl celluloses, hydroxyalkyl celluloses, cellulose ethers, cellulose esters, nitro celluloses, hydroxypropylcellulose, carboxymethylcellulose, polymers of acrylic acids, such as poly(methyl(meth)acrylate) (PMMA), poly(ethyl(meth)acrylate), poly(butyl(meth)acrylate), poly(isobutyl(meth)acrylate), poly(hexyl(meth)acrylate), poly(isodecyl(meth)acrylate), poly(lauryl(meth)acrylate), poly(phenyl(meth)acrylate), poly(methyl acrylate), poly(isopropyl acrylate), poly(isobutyl acrylate), poly(octadecyl acrylate) and copolymers and mixtures thereof, polydioxanone and its copolymers, polyhydroxyalkanoates, polypropylene fumarate, polyoxymethylene, poloxamers, poly(ortho)esters, poly(butyric acid), poly(valeric acid), poly(lactide-co- caprolactone), PEG-PLGA-PEG, trimethylene carbonate, and polyvinylpyrrolidone. The lipid nanoparticle may be coated or associated with a copolymer such as, but not limited to, a block co-polymer (such as a branched polyether-polyamide block copolymer described in International Publication No. WO2013012476, herein incorporated by reference in its entirety), and (poly(ethylene glycol))-(poly(propylene oxide))-(poly(ethylene glycol)) triblock copolymer (*see e.g.*, U.S. Publication 20120121718, U.S. Publication 20100003337, and U.S. Patent No. 8,263,665, each of which is herein incorporated by reference in its entirety). The co-polymer may be a polymer that is generally regarded as safe (GRAS) and the formation of the lipid nanoparticle may be in such a way that no new chemical entities

are created. For example, the lipid nanoparticle may comprise poloxamers coating PLGA nanoparticles without forming new chemical entities which are still able to rapidly penetrate human mucus (Yang *et al. Angew. Chem. Int. Ed.* 2011 50:2597-2600, the content of which is herein incorporated by reference in its entirety). A non-limiting scalable method to produce nanoparticles which can penetrate human mucus is described by Xu *et al.* (*see e.g., J Control Release* 2013, 170(2):279-86, the content of which is herein incorporated by reference in its entirety).

The vitamin of the polymer-vitamin conjugate may be vitamin E. The vitamin portion of the conjugate may be substituted with other suitable components such as, but not limited to, vitamin A, vitamin E, other vitamins, cholesterol, a hydrophobic moiety, or a hydrophobic component of other surfactants (*e.g.*, sterol chains, fatty acids, hydrocarbon chains and alkylene oxide chains).

In some embodiments, the RNA (*e.g.*, mRNA) vaccine pharmaceutical compositions may be formulated in liposomes such as, but not limited to, DiLa2 liposomes (Marina Biotech, Bothell, WA), SMARTICLES® (Marina Biotech, Bothell, WA), neutral DOPC (1,2-dioleoyl-sn-glycero-3-phosphocholine) based liposomes (*e.g.*, siRNA delivery for ovarian cancer (Landen *et al. Cancer Biology & Therapy* 2006 5(12)1708-1713, herein incorporated by reference in its entirety)), and hyaluronan-coated liposomes (Quiet Therapeutics, Israel).

In some embodiments, the RNA (*e.g.* mRNA) vaccines may be formulated in a lyophilized gel-phase liposomal composition as described in U.S. Publication No. US2012060293, herein incorporated by reference in its entirety.

The nanoparticle formulations may comprise a phosphate conjugate. The phosphate conjugate may increase *in vivo* circulation times and/or increase the targeted delivery of the nanoparticle. Phosphate conjugates for use with the present invention may be made by the methods described in International Publication No. WO2013033438 or U.S. Publication No. 20130196948, the content of each of which is herein incorporated by reference in its entirety. As a non-limiting example, the phosphate conjugates may include a compound of any one of the formulas described in International Publication No. WO2013033438, herein incorporated by reference in its entirety.

The nanoparticle formulation may comprise a polymer conjugate. The polymer conjugate may be a water-soluble conjugate. The polymer conjugate may have a structure as described in U.S. Application No. 20130059360, the content of which is herein incorporated by reference in its entirety. In some aspects, polymer conjugates with the polynucleotides of the present invention may be made using the methods and/or segmented polymeric reagents

described in U.S. Patent Application No. 20130072709, herein incorporated by reference in its entirety. In other aspects, the polymer conjugate may have pendant side groups comprising ring moieties such as, but not limited to, the polymer conjugates described in U.S. Publication No. US20130196948, the content of which is herein incorporated by reference in its entirety.

The lipid nanoparticle engineered to penetrate mucus may include surface altering agents such as, but not limited to, polynucleotides, anionic proteins (*e.g.*, bovine serum albumin), surfactants (*e.g.*, cationic surfactants such as for example dimethyldioctadecyl-ammonium bromide), sugars or sugar derivatives (*e.g.*, cyclodextrin), nucleic acids, polymers (*e.g.*, heparin, polyethylene glycol and poloxamer), mucolytic agents (*e.g.*, N-acetylcysteine, mugwort, bromelain, papain, clerodendrum, acetylcysteine, bromhexine, carbocisteine, eprazinone, mesna, ambroxol, sobrerol, domiodol, letosteine, stepronin, tiopronin, gelsolin, thymosin β4 dornase alfa, neltenexine, erdosteine) and various DNases including rhDNase. The surface altering agent may be embedded or enmeshed in the particle's surface or disposed (*e.g.*, by coating, adsorption, covalent linkage, or other process) on the surface of the lipid nanoparticle (see *e.g.*, U.S. Publication 20100215580 and U.S. Publication 20080166414 and US20130164343 the content of each of which is herein incorporated by reference in its entirety).

In some embodiments, the mucus penetrating lipid nanoparticles may comprise at least one polynucleotide described herein. The polynucleotide may be encapsulated in the lipid nanoparticle and/or disposed on the surface of the paricle. The polynucleotide may be covalently coupled to the lipid nanoparticle. Formulations of mucus penetrating lipid nanoparticles may comprise a plurality of nanoparticles. Further, the formulations may contain particles which may interact with the mucus and alter the structural and/or adhesive properties of the surrounding mucus to decrease mucoadhesion which may increase the delivery of the mucus penetrating lipid nanoparticles to the mucosal tissue.

In other embodiments, the mucus penetrating lipid nanoparticles may be a hypotonic formulation comprising a mucosal penetration enhancing coating. The formulation may be hypotonice for the epithelium to which it is being delivered.

Non-limiting examples of hypotonic formulations may be found in International Publication No. WO2013110028, the content of which is herein incorporated by reference in its entirety.

In some embodiments, in order to enhance the delivery through the mucosal barrier the RNA vaccine formulation may comprise or be a hypotonic solution. Hypotonic solutions were found to increase the rate at which mucoinert particles such as, but not limited to, mucus-

penetrating particles, were able to reach the vaginal epithelial surface (*see e.g.*, Ensign *et al. Biomaterials* 2013, 34(28):6922-9, the content of which is herein incorporated by reference in its entirety).

In some embodiments, the RNA vaccine is formulated as a lipoplex, such as, without limitation, the ATUPLEX™ system, the DACC system, the DBTC system and other siRNA-lipoplex technology from Silence Therapeutics (London, United Kingdom), STEMFECT™ from STEMGENT® (Cambridge, MA), and polyethylenimine (PEI) or protamine-based targeted and non-targeted delivery of nucleic acids (Aleku *et al. Cancer Res.* 2008 68:9788-9798; Strumberg *et al. Int J Clin Pharmacol Ther* 2012 50:76-78; Santel *et al., Gene Ther* 2006 13:1222-1234; Santel *et al., Gene Ther* 2006 13:1360-1370; Gutbier *et al., Pulm Pharmacol. Ther.* 2010 23:334-344; Kaufmann *et al. Microvasc Res* 2010 80:286-293; Weide *et al. J Immunother.* 2009 32:498-507; Weide *et al. J Immunoth*er. 2008 31:180-188; Pascolo, *Expert Opin. Biol. Ther.* 4:1285-1294; Fotin-Mleczek *et al.*, 2011 *J. Immunother.* 34:1-15; Song *et al., Nature Biotechnol.* 2005, 23:709-717; Peer *et al., Proc Natl Acad Sci U S A.* 2007 6;104:4095-4100; deFougerolles *Hum Gene Ther.* 2008 19:125-132; each of which is incorporated herein by reference in its entirety).

In some embodiments, such formulations may also be constructed or compositions altered such that they passively or actively are directed to different cell types *in vivo*, including but not limited to hepatocytes, immune cells, tumor cells, endothelial cells, antigen presenting cells, and leukocytes (Akinc *et al. Mol Ther.* 2010 18:1357-1364; Song *et al., Nat Biotechnol.* 2005 23:709-717; Judge *et al., J Clin Invest.* 2009 119:661-673; Kaufmann *et al., Microvasc Res* 2010 80:286-293; Santel *et al., Gene Ther* 2006 13:1222-1234; Santel *et al., Gene Ther* 2006 13:1360-1370; Gutbier *et al., Pulm Pharmacol. Ther.* 2010 23:334-344; Basha *et al., Mol. Ther.* 2011 19:2186-2200; Fenske and Cullis, *Expert Opin Drug Deliv.* 2008 5:25-44; Peer *et al., Science.* 2008 319:627-630; Peer and Lieberman, *Gene Ther.* 2011 18:1127-1133; each of which is incorporated herein by reference in its entirety). One example of passive targeting of formulations to liver cells includes the DLin-DMA, DLin-KC2-DMA, and DLin-MC3-DMA-based lipid nanoparticle formulations which have been shown to bind to apolipoprotein E and promote binding and uptake of these formulations into hepatocytes *in vivo* (Akinc *et al. Mol Ther.* 2010 18:1357-1364; herein incorporated by reference in its entirety). Formulations can also be selectively targeted through expression of different ligands on their surface as exemplified by, but not limited by, folate, transferrin, N-acetylgalactosamine (GalNAc), and antibody targeted approaches (Kolhatkar *et al.*, *Curr Drug Discov Technol.* 2011 8:197-206; Musacchio and Torchilin, *Front Biosci.* 2011 16:1388-1412; Yu *et al., Mol Membr Biol.* 2010

27:286-298; Patil *et al., Crit Rev Ther Drug Carrier Syst.* 2008 25:1-61; Benoit *et al., Biomacromolecules.* 2011 12:2708-2714; Zhao *et al., Expert Opin Drug Deliv.* 2008 5:309-319; Akinc *et al., Mol Ther.* 2010 18:1357-1364; Srinivasan *et al., Methods Mol Biol.* 2012 820:105-116; Ben-Arie *et al., Methods Mol Biol.* 2012 757:497-507; Peer *2010 J Control Release.* 20:63-68; Peer *et al., Proc Natl Acad Sci U S A.* 2007 104:4095-4100; Kim *et al., Methods Mol Biol.* 2011 721:339-353; Subramanya *et al., Mol Ther.* 2010 18:2028-2037; Song *et al., Nat Biotechnol.* 2005 23:709-717; Peer *et al., Science.* 2008 319:627-630; Peer and Lieberman, *Gene Ther.* 2011 18:1127-1133; each of which is incorporated herein by reference in its entirety).

In some embodiments, the RNA (*e.g.*, mRNA) vaccine is formulated as a solid lipid nanoparticle. A solid lipid nanoparticle (SLN) may be spherical with an average diameter between to 1000 nm. SLNs possess a solid lipid core matrix that can solubilize lipophilic molecules and may be stabilized with surfactants and/or emulsifiers. In other embodiments, the lipid nanoparticle may be a self-assembly lipid-polymer nanoparticle (*see* Zhang *et al., ACS Nano*, 2008, 2 (8), pp 1696–1702; the content of which is herein incorporated by reference in its entirety). As a non-limiting example, the SLN may be the SLN described in International Publication No. WO2013105101, the content of which is herein incorporated by reference in its entirety. As another non-limiting example, the SLN may be made by the methods or processes described in International Publication No. WO2013105101, the content of which is herein incorporated by reference in its entirety.

Liposomes, lipoplexes, or lipid nanoparticles may be used to improve the efficacy of polynucleotides directed protein production as these formulations may be able to increase cell transfection by the RNA (*e.g.* mRNA) vaccine; and/or increase the translation of encoded protein. One such example involves the use of lipid encapsulation to enable the effective systemic delivery of polyplex plasmid DNA (Heyes *et al., Mol Ther.* 2007 15:713-720; herein incorporated by reference in its entirety). The liposomes, lipoplexes, or lipid nanoparticles may also be used to increase the stability of the polynucleotide.

In some embodiments, the RNA (*e.g.*, mRNA) vaccines of the present invention can be formulated for controlled release and/or targeted delivery. As used herein, "controlled release" refers to a pharmaceutical composition or compound release profile that conforms to a particular pattern of release to effect a therapeutic outcome. In some embodiments, the RNA vaccines may be encapsulated into a delivery agent described herein and/or known in the art for controlled release and/or targeted delivery. As used herein, the term "encapsulate" means to enclose, surround, or encase. As it relates to the formulation of the compounds of the

invention, encapsulation may be substantial, complete, or partial. The term "substantially encapsulated" means that at least greater than 50, 60, 70, 80, 85, 90, 95, 96, 97, 98, 99, 99.9, 99.99 or greater than 99.999% of the pharmaceutical composition or compound of the invention may be enclosed, surrounded, or encased within the delivery agent. "Partially encapsulation" means that less than 10, 10, 20, 30, 40, 50% or less of the pharmaceutical composition or compound of the invention may be enclosed, surrounded, or encased within the delivery agent. Advantageously, encapsulation may be determined by measuring the escape or the activity of the pharmaceutical composition or compound of the invention using fluorescence and/or electron micrograph. For example, at least 1, 5, 10, 20, 30, 40, 50, 60, 70, 80, 85, 90, 95, 96, 97, 98, 99, 99.9, 99.99 or greater than 99.99% of the pharmaceutical composition or compound of the present disclosure are encapsulated in the delivery agent.

In some embodiments, the controlled release formulation may include, but is not limited to, tri-block co-polymers. As a non-limiting example, the formulation may include two different types of tri-block co-polymers (International Pub. No. WO2012131104 and WO2012131106; the contents of each of which is herein incorporated by reference in its entirety).

In other embodiments, the RNA vaccines may be encapsulated into a lipid nanoparticle or a rapidly eliminated lipid nanoparticle and the lipid nanoparticles or a rapidly eliminated lipid nanoparticle may then be encapsulated into a polymer, hydrogel, and/or surgical sealant described herein and/or known in the art. As a non-limiting example, the polymer, hydrogel or surgical sealant may be PLGA, ethylene vinyl acetate (EVAc), poloxamer, GELSITE® (Nanotherapeutics, Inc. Alachua, FL), HYLENEX® (Halozyme Therapeutics, San Diego CA), surgical sealants such as fibrinogen polymers (Ethicon Inc. Cornelia, GA), TISSELL® (Baxter International, Inc Deerfield, IL), PEG-based sealants, and COSEAL® (Baxter International, Inc Deerfield, IL).

In other embodiments, the lipid nanoparticle may be encapsulated into any polymer known in the art which may form a gel when injected into a subject. As another non-limiting example, the lipid nanoparticle may be encapsulated into a polymer matrix which may be biodegradable.

In some embodiments, the RNA (*e.g.* mRNA) vaccine formulation for controlled release and/or targeted delivery may also include at least one controlled release coating. Controlled release coatings include, but are not limited to, OPADRY®, polyvinylpyrrolidone/vinyl acetate copolymer, polyvinylpyrrolidone, hydroxypropyl methylcellulose, hydroxypropyl cellulose, hydroxyethyl cellulose, EUDRAGIT RL®,

EUDRAGIT RS® and cellulose derivatives such as ethylcellulose aqueous dispersions (AQUACOAT® and SURELEASE®).

In some embodiments, the RNA (*e.g.*, mRNA) vaccine controlled release and/or targeted delivery formulation may comprise at least one degradable polyester which may contain polycationic side chains. Degradeable polyesters include, but are not limited to, poly(serine ester), poly(L-lactide-co-L-lysine), poly(4-hydroxy-L-proline ester), and combinations thereof. In other embodiments, the degradable polyesters may include a PEG conjugation to form a PEGylated polymer.

In some embodiments, the RNA vaccine controlled release and/or targeted delivery formulation comprising at least one polynucleotide may comprise at least one PEG and/or PEG related polymer derivatives as described in U.S. Patent No. 8,404,222, herein incorporated by reference in its entirety.

In other embodiments, the RNA vaccine controlled release delivery formulation comprising at least one polynucleotide may be the controlled release polymer system described in U.S. Publication No. 20130130348, herein incorporated by reference in its entirety.

In some embodiments, the RNA (*e.g.*, mRNA) vaccines of the present invention may be encapsulated in a therapeutic nanoparticle, referred to herein as "therapeutic nanoparticle RNA vaccines." Therapeutic nanoparticles may be formulated by methods described herein and known in the art such as, but not limited to, International Publication Nos. WO2010005740, WO2010030763, WO2010005721, WO2010005723, and WO2012054923, U.S. Pubication Nos. US20110262491, US20100104645, US20100087337, US20100068285, US20110274759, US20100068286, US20120288541, US20130123351 and US20130230567, and US Patent Nos. 8,206,747, 8,293,276, 8,318,208 and 8,318,211, the content of each of which is herein incorporated by reference in its entirety. In other embodiments, therapeutic polymer nanoparticles may be identified by the methods described in U.S. Publication No. US20120140790, the content of which is herein incorporated by reference in its entirety.

In some embodiments, the therapeutic nanoparticle RNA vaccine may be formulated for sustained release. As used herein, "sustained release" refers to a pharmaceutical composition or compound that conforms to a release rate over a specific period of time. The period of time may include, but is not limited to, hours, days, weeks, months, and years. As a non-limiting example, the sustained release nanoparticle may comprise a polymer and a therapeutic agent such as, but not limited to, the polynucleotides of the present invention (*see* International Publication No. 2010075072 and U.S. Publication Nos. US20100216804, US20110217377 and US20120201859, each of which is herein incorporated by reference in its entirety). In

another non-limiting example, the sustained release formulation may comprise agents which permit persistent bioavailability such as, but not limited to, crystals, macromolecular gels and/or particulate suspensions (*see* U.S. Publication No. US20130150295, the content of which is herein incorporated by reference in its entirety).

In some embodiments, the therapeutic nanoparticle RNA (*e.g.* mRNA) vaccines may be formulated to be target specific. As a non-limiting example, the therapeutic nanoparticles may include a corticosteroid (*see* International Publication No. WO2011084518, herein incorporated by reference in its entirety). As a non-limiting example, the therapeutic nanoparticles may be formulated in nanoparticles described in International Publication Nos. WO2008121949, WO2010005726, WO2010005725, WO2011084521 and U.S. Publication Nos. US20100069426, US20120004293 and US20100104655, each of which is herein incorporated by reference in its entirety.

In some embodiments, the nanoparticles of the present invention may comprise a polymeric matrix. As a non-limiting example, the nanoparticle may comprise two or more polymers such as, but not limited to, polyethylenes, polycarbonates, polyanhydrides, polyhydroxyacids, polypropylfumerates, polycaprolactones, polyamides, polyacetals, polyethers, polyesters, poly(orthoesters), polycyanoacrylates, polyvinyl alcohols, polyurethanes, polyphosphazenes, polyacrylates, polymethacrylates, polycyanoacrylates, polyureas, polystyrenes, polyamines, polylysine, poly(ethylene imine), poly(serine ester), poly(L-lactide-co-L-lysine), poly(4-hydroxy-L-proline ester), or combinations thereof.

In some embodiments, the therapeutic nanoparticle comprises a diblock copolymer. In some embodiments, the diblock copolymer may include PEG in combination with a polymer such as, but not limited to, polyethylenes, polycarbonates, polyanhydrides, polyhydroxyacids, polypropylfumerates, polycaprolactones, polyamides, polyacetals, polyethers, polyesters, poly(orthoesters), polycyanoacrylates, polyvinyl alcohols, polyurethanes, polyphosphazenes, polyacrylates, polymethacrylates, polycyanoacrylates, polyureas, polystyrenes, polyamines, polylysine, poly(ethylene imine), poly(serine ester), poly(L-lactide-co-L-lysine), poly(4-hydroxy-L-proline ester), or combinations thereof. In yet other embodiments, the diblock copolymer may be a high-X diblock copolymer such as those described in International Publication No. WO2013120052, the content of which is herein incorporated by reference in its entirety.

As a non-limiting example, the therapeutic nanoparticle comprises a PLGA-PEG block copolymer (*see* U.S. Publication No. US20120004293 and U.S. Patent No. 8,236,330, each of which is herein incorporated by reference in its entirety). In another non-limiting

example, the therapeutic nanoparticle is a stealth nanoparticle comprising a diblock copolymer of PEG and PLA or PEG and PLGA (*see* U.S. Patent No. 8,246,968 and International Publication No. WO2012166923, the content of each of which is herein incorporated by reference in its entirety). In yet another non-limiting example, the therapeutic nanoparticle is a stealth nanoparticle or a target-specific stealth nanoparticle as described in U.S. Publication No. 20130172406, the content of which is herein incorporated by reference in its entirety.

In some embodiments, the therapeutic nanoparticle may comprise a multiblock copolymer (*see e.g.*, U.S. Patent Nos. 8,263,665 and 8,287,910 and U.S. Publication No. 20130195987, the content of each of which is herein incorporated by reference in its entirety).

In yet another non-limiting example, the lipid nanoparticle comprises the block copolymer PEG-PLGA-PEG (*see e.g.*, the thermosensitive hydrogel (PEG-PLGA-PEG) used as a TGF-beta1 gene delivery vehicle in Lee *et al.* "Thermosensitive Hydrogel as a Tgf-β1 Gene Delivery Vehicle Enhances Diabetic Wound Healing." *Pharmaceutical Research*, 2003 20(12): 1995-2000; and used as a controlled gene delivery system in Li *et al.* "Controlled Gene Delivery System Based on Thermosensitive Biodegradable Hydrogel" *Pharmaceutical Research* 2003 20(6):884- 888; and Chang *et al.*, "Non-ionic amphiphilic biodegradable PEG-PLGA-PEG copolymer enhances gene delivery efficiency in rat skeletal muscle." *J Controlled Release*. 2007 118:245-253; each of which is herein incorporated by reference in its entirety). The RNA (*e.g.*, mRNA) vaccines of the present disclosure may be formulated in lipid nanoparticles comprising the PEG-PLGA-PEG block copolymer.

In some embodiments, the therapeutic nanoparticle may comprise a multiblock copolymer (*see e.g.*, U.S. Patent Nos. 8,263,665 and 8,287,910 and U.S. Publication No. 20130195987, the content of each of which is herein incorporated by reference in its entirety).

In some embodiments, the block copolymers described herein may be included in a polyion complex comprising a non-polymeric micelle and the block copolymer. (*see e.g.*, U.S. Publication No. 20120076836, herein incorporated by reference in its entirety).

In some embodiments, the therapeutic nanoparticle may comprise at least one acrylic polymer. Acrylic polymers include but are not limited to, acrylic acid, methacrylic acid, acrylic acid and methacrylic acid copolymers, methyl methacrylate copolymers, ethoxyethyl methacrylates, cyanoethyl methacrylate, amino alkyl methacrylate copolymer, poly(acrylic acid), poly(methacrylic acid), polycyanoacrylates, and combinations thereof.

In some embodiments, the therapeutic nanoparticles may comprise at least one poly(vinyl ester) polymer. The poly(vinyl ester) polymer may be a copolymer such as a random copolymer. As a non-limiting example, the random copolymer may have a structure such as those described in International Publication No. WO2013032829 or U.S. Publication No. 20130121954, the content of which is herein incorporated by reference in its entirety. In some aspects, the poly(vinyl ester) polymers may be conjugated to the polynucleotides described herein.

In some embodiments, the therapeutic nanoparticle may comprise at least one diblock copolymer. The diblock copolymer may be, but it not limited to, a poly(lactic) acid-poly(ethylene)glycol copolymer (*see e.g.*, International Publication No. WO2013044219; herein incorporated by reference in its entirety). As a non-limiting example, the therapeutic nanoparticle may be used to treat cancer (*see* International Publication No. WO2013044219, herein incorporated by reference in its entirety).

In some embodiments, the therapeutic nanoparticles may comprise at least one cationic polymer described herein and/or known in the art.

In some embodiments, the therapeutic nanoparticles may comprise at least one amine-containing polymer such as, but not limited to polylysine, polyethyleneimine, poly(amidoamine) dendrimers, poly(beta-amino esters) (*see e.g.*, U.S. Patent No. 8,287,849, herein incorporated by reference in its entirety), and combinations thereof. In other embodiments, the nanoparticles described herein may comprise an amine cationic lipid such as those described in International Publication No. WO2013059496, the content of which is herein incorporated by reference in its entirety. In some aspects, the cationic lipids may have an amino-amine or an amino-amide moiety.

In some embodiments, the therapeutic nanoparticles may comprise at least one degradable polyester, which may contain polycationic side chains. Degradeable polyesters include, but are not limited to, poly(serine ester), poly(L-lactide-co-L-lysine), poly(4-hydroxy-L-proline ester), and combinations thereof. In other embodiments, the degradable polyesters may include a PEG conjugation to form a PEGylated polymer.

In other embodiments, the therapeutic nanoparticle may include a conjugation of at least one targeting ligand. The targeting ligand may be any ligand known in the art such as, but not limited to, a monoclonal antibody (Kirpotin *et al, Cancer Res.* 2006 66:6732-6740, herein incorporated by reference in its entirety).

In some embodiments, the therapeutic nanoparticle may be formulated in an aqueous solution, which may be used to target cancer (*see* International Publication No.

WO2011084513 and U.S. Publication No. 20110294717, each of which is herein incorporated by reference in its entirety).

In some embodiments, the therapeutic nanoparticle RNA (*e.g.* mRNA) vaccines, *e.g.*, therapeutic nanoparticles comprising at least one RNA vaccine may be formulated using the methods described by Podobinski *et al* in U.S. Patent No. 8,404,799, the content of which is herein incorporated by reference in its entirety.

In some embodiments, the RNA (*e.g.*, mRNA) vaccines may be encapsulated in, linked to and/or associated with synthetic nanocarriers. Synthetic nanocarriers include, but are not limited to, those described in International Publication Nos. WO2010005740, WO2012149454, and WO2013019669, and U.S. Publicaiton Nos. US20110262491, US20100104645, US20100087337, and US20120244222, each of which is herein incorporated by reference in its entirety. The synthetic nanocarriers may be formulated using methods known in the art and/or described herein. As a non-limiting example, the synthetic nanocarriers may be formulated by the methods described in International Publication Nos. WO2010005740, WO2010030763, and WO201213501, and U.S. Publication Nos. US20110262491, US20100104645, US20100087337, and US2012024422, each of which is herein incorporated by reference in its entirety. In other embodiments, the synthetic nanocarrier formulations may be lyophilized by methods described in International Publication No. WO2011072218 and U.S. Patent No. 8,211,473, the content of each of which is herein incorporated by reference in its entirety. In yet other embodiments, formulations of the present invention, including, but not limited to, synthetic nanocarriers, may be lyophilized or reconstituted by the methods described in U.S. Publication No. 20130230568, the content of which is herein incorporated by reference in its entirety.

In some embodiments, the synthetic nanocarriers may contain reactive groups to release the polynucleotides described herein (*see* International Publication No. WO20120952552 and U.S. Publication No. US20120171229, each of which is herein incorporated by reference in its entirety).

In some embodiments, the synthetic nanocarriers may contain an immunostimulatory agent to enhance the immune response from delivery of the synthetic nanocarrier. As a non-limiting example, the synthetic nanocarrier may comprise a Th1 immunostimulatory agent which may enhance a Th1-based response of the immune system (*see* International Publication No. WO2010123569 and U.S. Publication No. 20110223201, each of which is herein incorporated by reference in its entirety).

In some embodiments, the synthetic nanocarriers may be formulated for targeted release. In some embodiments, the synthetic nanocarrier is formulated to release the polynucleotides at a specified pH and/or after a desired time interval. As a non-limiting example, the synthetic nanoparticle may be formulated to release the RNA (*e.g.* mRNA) vaccines after 24 hours and/or at a pH of 4.5 (*see* International Publication Nos. WO2010138193 and WO2010138194 and U.S. Publication Nos. US20110020388 and US20110027217, each of which is herein incorporated by reference in its entirety).

In some embodiments, the synthetic nanocarriers may be formulated for controlled and/or sustained release of the polynucleotides described herein. As a non-limiting example, the synthetic nanocarriers for sustained release may be formulated by methods known in the art, described herein and/or as described in International Publication No. WO2010138192 and U.S. Publication No. 20100303850, each of which is herein incorporated by reference in its entirety.

In some embodiments, the RNA (*e.g.* mRNA) vaccine may be formulated for controlled and/or sustained release wherein the formulation comprises at least one polymer that is a crystalline side chain (CYSC) polymer. CYSC polymers are described in U.S. Patent No. 8,399,007, herein incorporated by reference in its entirety.

In some embodiments, the synthetic nanocarrier may be formulated for use as a vaccine. In some embodiments, the synthetic nanocarrier may encapsulate at least one polynucleotide which encodes at least one antigen. As a non-limiting example, the synthetic nanocarrier may include at least one antigen and an excipient for a vaccine dosage form (*see* International Publication No. WO2011150264 and U.S. Publication No. 20110293723, each of which is herein incorporated by reference in its entirety). As another non-limiting example, a vaccine dosage form may include at least two synthetic nanocarriers with the same or different antigens and an excipient (*see* International Publication No. WO2011150249 and U.S. Publication No. 20110293701, each of which is herein incorporated by reference in its entirety). The vaccine dosage form may be selected by methods described herein, known in the art, and/or described in International Publication No. WO2011150258 and U.S. Publication No. US20120027806, each of which is herein incorporated by reference in its entirety.

In some embodiments, the synthetic nanocarrier may comprise at least one polynucleotide which encodes at least one adjuvant. As non-limiting example, the adjuvant may comprise dimethyldioctadecylammonium-bromide, dimethyldioctadecylammonium-chloride, dimethyldioctadecylammonium-phosphate or dimethyldioctadecylammonium-acetate (DDA), and an apolar fraction or part of said apolar fraction of a total lipid extract of a

mycobacterium (*see e.g.,* U.S. Patent No. 8,241,610; herein incorporated by reference in its entirety). In other embodiments, the synthetic nanocarrier may comprise at least one polynucleotide and an adjuvant. As a non-limiting example, the synthetic nanocarrier comprising an adjuvant may be formulated by the methods described in International Publication No. WO2011150240 and U.S. Publication No. US20110293700, each of which is herein incorporated by reference in its entirety.

In some embodiments, the synthetic nanocarrier may encapsulate at least one polynucleotide which encodes a peptide, fragment, or region from a virus. As a non-limiting example, the synthetic nanocarrier may include, but is not limited to, the nanocarriers described in International Publication Nos. WO2012024621, WO201202629, and WO2012024632 and U.S. Publication Nos. US20120064110, US20120058153, and US20120058154, each of which is herein incorporated by reference in its entirety.

In some embodiments, the synthetic nanocarrier may be coupled to a polynucleotide which may be able to trigger a humoral and/or cytotoxic T lymphocyte (CTL) response (*see e.g.*, International Publication No. WO2013019669, herein incorporated by reference in its entirety).

In some embodiments, the RNA (*e.g.* mRNA) vaccine may be encapsulated in, linked to and/or associated with zwitterionic lipids. Non-limiting examples of zwitterionic lipids and methods of using zwitterionic lipids are described in U.S. Publication No. 20130216607, the content of which is herein incorporated by reference in its entirety. In some aspects, the zwitterionic lipids may be used in the liposomes and lipid nanoparticles described herein.

In some embodiments, the RNA (*e.g.* mRNA) vaccine may be formulated in colloid nanocarriers as described in U.S. Publication No. 20130197100, the content of which is herein incorporated by reference in its entirety.

In some embodiments, the nanoparticle may be optimized for oral administration. The nanoparticle may comprise at least one cationic biopolymer such as, but not limited to, chitosan or a derivative thereof. As a non-limiting example, the nanoparticle may be formulated by the methods described in U.S. Publication No. 20120282343; herein incorporated by reference in its entirety.

In some embodiments, LNPs comprise the lipid KL52 (an amino-lipid disclosed in U.S. Application Publication No. 2012/0295832 expressly incorporated herein by reference in its entirety). Activity and/or safety (as measured by examining one or more of ALT/AST, white blood cell count and cytokine induction) of LNP administration may be improved by incorporation of such lipids. LNPs comprising KL52 may be administered intravenously and/or

in one or more doses. In some embodiments, administration of LNPs comprising KL52 results in equal or improved mRNA and/or protein expression as compared to LNPs comprising MC3.

In some embodiments, RNA (*e.g.* mRNA) vaccines may be delivered using smaller LNPs. Such particles may comprise a diameter from below 0.1 μm up to 100 nm such as, but not limited to, less than 0.1 μm, less than 1.0 μm, less than 5 μm, less than 10 μm, less than 15 μm, less than 20 μm, less than 25 μm, less than 30 μm, less than 35 μm, less than 40 μm, less than 50 μm, less than 55 μm, less than 60 μm, less than 65 μm, less than 70 μm, less than 75 μm, less than 80 μm, less than 85 μm, less than 90 μm, less than 95 μm, less than 100 μm, less than 125 μm, less than 150 μm, less than 175 μm, less than 200 μm, less than 225 μm, less than 250 μm, less than 275 μm, less than 300 μm, less than 325 μm, less than 350 μm, less than 375 μm, less than 400 μm, less than 425 μm, less than 450 μm, less than 475 μm, less than 500 μm, less than 525 μm, less than 550 μm, less than 575 μm, less than 600 μm, less than 625 μm, less than 650 μm, less than 675 μm, less than 700 μm, less than 725 μm, less than 750 μm, less than 775 μm, less than 800 μm, less than 825 μm, less than 850 μm, less than 875 μm, less than 900 μm, less than 925 μm, less than 950 μm, or less than 975 μm.

In other embodiments, RNA (*e.g.*, mRNA) vaccines may be delivered using smaller LNPs which may comprise a diameter from about 1 nm to about 100 nm, from about 1 nm to about 10 nm, about 1 nm to about 20 nm, from about 1 nm to about 30 nm, from about 1 nm to about 40 nm, from about 1 nm to about 50 nm, from about 1 nm to about 60 nm, from about 1 nm to about 70 nm, from about 1 nm to about 80 nm, from about 1 nm to about 90 nm, from about 5 nm to about from 100 nm, from about 5 nm to about 10 nm, about 5 nm to about 20 nm, from about 5 nm to about 30 nm, from about 5 nm to about 40 nm, from about 5 nm to about 50 nm, from about 5 nm to about 60 nm, from about 5 nm to about 70 nm, from about 5 nm to about 80 nm, from about 5 nm to about 90 nm, about 10 to about 50 nm, from about 20 to about 50 nm, from about 30 to about 50 nm, from about 40 to about 50 nm, from about 20 to about 60 nm, from about 30 to about 60 nm, from about 40 to about 60 nm, from about 20 to about 70 nm, from about 30 to about 70 nm, from about 40 to about 70 nm, from about 50 to about 70 nm, from about 60 to about 70 nm, from about 20 to about 80 nm, from about 30 to about 80 nm, from about 40 to about 80 nm, from about 50 to about 80 nm, from about 60 to about 80 nm, from about 20 to about 90 nm, from about 30 to about 90 nm, from about 40 to about 90 nm, from about 50 to about 90 nm, from about 60 to about 90 nm, and/or from about 70 to about 90 nm.

In some embodiments, such LNPs are synthesized using methods comprising microfluidic mixers. Exemplary microfluidic mixers may include, but are not limited to a slit interdigitial micromixers including, but not limited to those manufactured by Microinnova (Allerheiligen bei Wildon, Austria) and/or a staggered herringbone micromixer (SHM) (Zhigaltsev, I.V. *et al.*, Bottom-up design and synthesis of limit size lipid nanoparticle systems with aqueous and triglyceride cores using millisecond microfluidic mixing. Langmuir. 2012. 28:3633-40) have been published (Belliveau, N.M. *et al.*, Microfluidic synthesis of highly potent limit-size lipid nanoparticles for *in vivo* delivery of siRNA. *Molecular Therapy-Nucleic Acids*. 2012. 1:e37; Chen, D. *et al.*, Rapid discovery of potent siRNA-containing lipid nanoparticles enabled by controlled microfluidic formulation. *J Am Chem Soc.* 2012. 134(16):6948-51; each of which is herein incorporated by reference in its entirety).

In some embodiments, methods of LNP generation comprising SHM, further comprise the mixing of at least two input streams wherein mixing occurs by microstructure-induced chaotic advection (MICA). According to this method, fluid streams down flow through channels present in a herringbone pattern, causing rotational flow and folding the fluids around each other. This method may also comprise a surface for fluid mixing wherein the surface changes orientations during fluid cycling. Methods of generating LNPs using SHM include those disclosed in U.S. Publication Nos. 2004/0262223 and 2012/0276209, each of which is expressly incorporated herein by reference in its entirety.

In some embodiments, the RNA (*e.g.* mRNA) vaccine of the present invention may be formulated in lipid nanoparticles created using a micromixer such as, but not limited to, a Slit Interdigital Microstructured Mixer (SIMM-V2) or a Standard Slit Interdigital Micro Mixer (SSIMM) or Caterpillar (CPMM) or Impinging-jet ((IJMM) from the Institut für Mikrotechnik Mainz GmbH, Mainz Germany).

In some embodiments, the RNA (*e.g.*, mRNA) vaccines of the present disclosure may be formulated in lipid nanoparticles created using microfluidic technology (*see* Whitesides, George M. The Origins and the Future of Microfluidics. *Nature*, 2006 442: 368-373; and Abraham *et al.* Chaotic Mixer for Microchannels. *Science*, 2002 295: 647-651; each of which is herein incorporated by reference in its entirety). As a non-limiting example, controlled microfluidic formulation includes a passive method for mixing streams of steady pressure-driven flows in micro channels at a low Reynolds number (*see e.g.*, Abraham *et al.* Chaotic Mixer for Microchannels. *Science*, 2002 295: 647651; which is herein incorporated by reference in its entirety).

In some embodiments, the RNA (*e.g.*, mRNA) vaccines of the present invention may be formulated in lipid nanoparticles created using a micromixer chip such as, but not limited to, those from Harvard Apparatus (Holliston, MA) or Dolomite Microfluidics (Royston, UK). A micromixer chip can be used for rapid mixing of two or more fluid streams with a split and recombine mechanism.

In some embodiments, the RNA (*e.g.*, mRNA) vaccines of the invention may be formulated for delivery using the drug encapsulating microspheres described in International Publication No. WO2013063468 or U.S. Patent No. 8,440,614, each of which is herein incorporated by reference in its entirety. The microspheres may comprise a compound of the formula (I), (II), (III), (IV), (V) or (VI) as described in International Publication No. WO2013063468, the content of which is herein incorporated by reference in its entirety. In other aspects, the amino acid, peptide, polypeptide, lipids are useful in delivering the RNA (*e.g.* mRNA) vaccines of the invention to cells (*see* International Publication No. WO2013063468, the contents of which is herein incorporated by reference in its entirety).

In some embodiments, the RNA (*e.g.*, mRNA) vaccines of the present disclosure may be formulated in lipid nanoparticles having a diameter from about 10 to about 100 nm such as, but not limited to, about 10 to about 20 nm, about 10 to about 30 nm, about 10 to about 40 nm, about 10 to about 50 nm, about 10 to about 60 nm, about 10 to about 70 nm, about 10 to about 80 nm, about 10 to about 90 nm, about 20 to about 30 nm, about 20 to about 40 nm, about 20 to about 50 nm, about 20 to about 60 nm, about 20 to about 70 nm, about 20 to about 80 nm, about 20 to about 90 nm, about 20 to about 100 nm, about 30 to about 40 nm, about 30 to about 50 nm, about 30 to about 60 nm, about 30 to about 70 nm, about 30 to about 80 nm, about 30 to about 90 nm, about 30 to about 100 nm, about 40 to about 50 nm, about 40 to about 60 nm, about 40 to about 70 nm, about 40 to about 80 nm, about 40 to about 90 nm, about 40 to about 100 nm, about 50 to about 60 nm, about 50 to about 70 nm about 50 to about 80 nm, about 50 to about 90 nm, about 50 to about 100 nm, about 60 to about 70 nm, about 60 to about 80 nm, about 60 to about 90 nm, about 60 to about 100 nm, about 70 to about 80 nm, about 70 to about 90 nm, about 70 to about 100 nm, about 80 to about 90 nm, about 80 to about 100 nm, and/or about 90 to about 100 nm.

In some embodiments, the lipid nanoparticles may have a diameter from about 10 to 500 nm.

In some embodiments, the lipid nanoparticle may have a diameter greater than 100 nm, greater than 150 nm, greater than 200 nm, greater than 250 nm, greater than 300 nm, greater than 350 nm, greater than 400 nm, greater than 450 nm, greater than 500 nm, greater than 550 nm,

greater than 600 nm, greater than 650 nm, greater than 700 nm, greater than 750 nm, greater than 800 nm, greater than 850 nm, greater than 900 nm, greater than 950 nm or greater than 1000 nm.

In some aspects, the lipid nanoparticle may be a limit size lipid nanoparticle described in International Publication No. WO2013059922, the content of which is herein incorporated by reference in its entirety. The limit size lipid nanoparticle may comprise a lipid bilayer surrounding an aqueous core or a hydrophobic core; where the lipid bilayer may comprise a phospholipid such as, but not limited to, diacylphosphatidylcholine, a diacylphosphatidylethanolamine, a ceramide, a sphingomyelin, a dihydrosphingomyelin, a cephalin, a cerebroside, a C8-C20 fatty acid diacylphophatidylcholine, and a 1-palmitoyl-2-oleoyl phosphatidylcholine (POPC). In other aspects, the limit size lipid nanoparticle may comprise a polyethylene glycol-lipid such as, but not limited to, DLPE-PEG, DMPE-PEG, DPPC-PEG, and DSPE-PEG.

In some embodiments, the RNA (*e.g.* mRNA) vaccines may be delivered, localized, and/or concentrated in a specific location using the delivery methods described in International Publication No. WO2013063530, the content of which is herein incorporated by reference in its entirety. As a non-limiting example, a subject may be administered an empty polymeric particle prior to, simultaneously with or after delivering the RNA (*e.g.* mRNA) vaccines to the subject. The empty polymeric particle undergoes a change in volume once in contact with the subject and becomes lodged, embedded, immobilized or entrapped at a specific location in the subject.

In some embodiments, the RNA (*e.g.* mRNA) vaccines may be formulated in an active substance release system (*see e.g.*, U.S. Publication No. US20130102545, the content of which is herein incorporated by reference in its entirety). The active substance release system may comprise 1) at least one nanoparticle bonded to an oligonucleotide inhibitor strand which is hybridized with a catalytically active nucleic acid and 2) a compound bonded to at least one substrate molecule bonded to a therapeutically active substance (*e.g.*, polynucleotides described herein), where the therapeutically active substance is released by the cleavage of the substrate molecule by the catalytically active nucleic acid.

In some embodiments, the RNA (*e.g.*, mRNA) vaccines may be formulated in a nanoparticle comprising an inner core comprising a non-cellular material and an outer surface comprising a cellular membrane. The cellular membrane may be derived from a cell or a membrane derived from a virus. As a non-limiting example, the nanoparticle may be made by the methods described in International Publication No. WO2013052167, herein incorporated by reference in its entirety. As another non-limiting example, the nanoparticle described in

International Publication No. WO2013052167, herein incorporated by reference in its entirety, may be used to deliver the RNA vaccines described herein.

In some embodiments, the RNA (*e.g.*, mRNA) vaccines may be formulated in porous nanoparticle-supported lipid bilayers (protocells). Protocells are described in International Publication No. WO2013056132, the content of which is herein incorporated by reference in its entirety.

In some embodiments, the RNA (*e.g.*, mRNA) vaccines described herein may be formulated in polymeric nanoparticles as described in or made by the methods described in US Patent Nos. 8,420,123 and 8,518,963 and European Patent No. EP2073848B1, the contents of each of which are herein incorporated by reference in their entirety. As a non-limiting example, the polymeric nanoparticle may have a high glass transition temperature such as the nanoparticles described in or nanoparticles made by the methods described in US Patent No. 8,518,963, the content of which is herein incorporated by reference in its entirety. As another non-limiting example, the polymer nanoparticle for oral and parenteral formulations may be made by the methods described in European Patent No. EP2073848B1, the content of which is herein incorporated by reference in its entirety.

In other embodiments, the RNA (*e.g.*, mRNA) vaccines described herein may be formulated in nanoparticles used in imaging. The nanoparticles may be liposome nanoparticles such as those described in U.S. Publication No. 20130129636, herein incorporated by reference in its entirety. As a non-limiting example, the liposome may comprise gadolinium(III)2-{4,7-bis-carboxymethyl-10-[(N,N-distearylamidomethyl-N′-amido-methyl]-1,4,7,10-tetra-azacyclododec-1-yl}-acetic acid and a neutral, fully saturated phospholipid component (*see e.g.*, U.S. Publication No. US20130129636, the contents of which is herein incorporated by reference in its entirety).

In some embodiments, the nanoparticles which may be used in the present invention are formed by the methods described in U.S. Patent Application No. 20130130348, the content of which is herein incorporated by reference in its entirety.

The nanoparticles of the present invention may further include nutrients such as, but not limited to, those which deficiencies can lead to health hazards from anemia to neural tube defects (*see e.g.,* the nanoparticles described in International Patent Publication No. WO2013072929, the contents of which is herein incorporated by reference in its entirety). As a non-limiting example, the nutrient may be iron in the form of ferrous, ferric salts, or elemental iron, iodine, folic acid, vitamins or micronutrients.

In some embodiments, the RNA (*e.g.*, mRNA) vaccines of the present invention may be formulated in a swellable nanoparticle. The swellable nanoparticle may be, but is not limited to, those described in U.S. Patent No. 8,440,231, the content of which is herein incorporated by reference in its entirety. As a non-limiting embodiment, the swellable nanoparticle may be used for delivery of the RNA (*e.g.*, mRNA) vaccines of the present invention to the pulmonary system (*see e.g.*, U.S. Patent No. 8,440,231, the content of which is herein incorporated by reference in its entirety).

The RNA (*e.g.*, mRNA) vaccines of the present invention may be formulated in polyanhydride nanoparticles such as, but not limited to, those described in U.S. Patent No. 8,449,916, the content of which is herein incorporated by reference in its entirety. The nanoparticles and microparticles of the present invention may be geometrically engineered to modulate macrophage and/or the immune response. In some aspects, the geometrically engineered particles may have varied shapes, sizes, and/or surface charges in order to incorporated the polynucleotides of the present invention for targeted delivery such as, but not limited to, pulmonary delivery (*see e.g.*, International Publication No. WO2013082111, the content of which is herein incorporated by reference in its entirety). Other physical features the geometrically engineering particles may have include, but are not limited to, fenestrations, angled arms, asymmetry, surface roughness, and charge, which can alter the interactions with cells and tissues. As a non-limiting example, nanoparticles of the present invention may be made by the methods described in International Publication No. WO2013082111, the content of which is herein incorporated by reference in its entirety.

In some embodiments, the nanoparticles of the present invention may be water soluble nanoparticles such as, but not limited to, those described in International Publication No. WO2013090601, the content of which is herein incorporated by reference in its entirety. The nanoparticles may be inorganic nanoparticles which have a compact and zwitterionic ligand in order to exhibit good water solubility. The nanoparticles may also have small hydrodynamic diameters (HD), stability with respect to time, pH, and salinity and a low level of non-specific protein binding.

In some embodiments, the nanoparticles of the present invention may be developed by the methods described in U.S. Publication No. US20130172406, the content of which is herein incorporated by reference in its entirety.

In some embodiments, the nanoparticles of the present invention are stealth nanoparticles or target-specific stealth nanoparticles such as, but not limited to, those described in U.S. Publication No. 20130172406, the content of which is herein incorporated

by reference in its entirety. The nanoparticles of the present invention may be made by the methods described in U.S. Publication No. 20130172406, the content of which is herein incorporated by reference in its entirety.

In other embodiments, the stealth or target-specific stealth nanoparticles may comprise a polymeric matrix. The polymeric matrix may comprise two or more polymers such as, but not limited to, polyethylenes, polycarbonates, polyanhydrides, polyhydroxyacids, polypropylfumerates, polycaprolactones, polyamides, polyacetals, polyethers, polyesters, poly(orthoesters), polycyanoacrylates, polyvinyl alcohols, polyurethanes, polyphosphazenes, polyacrylates, polymethacrylates, polycyanoacrylates, polyureas, polystyrenes, polyamines, polyesters, polyanhydrides, polyethers, polyurethanes, polymethacrylates, polyacrylates, polycyanoacrylates, or combinations thereof.

In some embodiments, the nanoparticle may be a nanoparticle-nucleic acid hybrid structure having a high density nucleic acid layer. As a non-limiting example, the nanoparticle-nucleic acid hybrid structure may made by the methods described in U.S. Publication No. 20130171646, the content of which is herein incorporated by reference in its entirety. The nanoparticle may comprise a nucleic acid such as, but not limited to, polynucleotides described herein and/or known in the art.

At least one of the nanoparticles of the present invention may be embedded in the core a nanostructure or coated with a low density porous 3-D structure or coating which is capable of carrying or associating with at least one payload within or on the surface of the nanostructure. Non-limiting examples of the nanostructures comprising at least one nanoparticle are described in International Publication No. WO2013123523, the content of which is herein incorporated by reference in its entirety.

Modes of Vaccine Administration

HSV RNA (*e.g.*, mRNA) vaccines may be administered by any route which results in a therapeutically effective outcome. These include, but are not limited, to intradermal, intramuscular, and/or subcutaneous administration. The present disclosure provides methods comprising administering RNA (*e.g.*, mRNA) vaccines to a subject in need thereof. The exact amount required will vary from subject to subject, depending on the species, age, and general condition of the subject, the severity of the disease, the particular composition, its mode of administration, its mode of activity, and the like. HSV RNA (*e.g.*, mRNA) vaccines compositions are typically formulated in dosage unit form for ease of administration and uniformity of dosage. It will be understood, however, that the total daily usage of HSV RNA

(*e.g.*, mRNA) vaccines compositions may be decided by the attending physician within the scope of sound medical judgment. The specific therapeutically effective, prophylactically effective, or appropriate imaging dose level for any particular patient will depend upon a variety of factors including the disorder being treated and the severity of the disorder; the activity of the specific compound employed; the specific composition employed; the age, body weight, general health, sex and diet of the patient; the time of administration, route of administration, and rate of excretion of the specific compound employed; the duration of the treatment; drugs used in combination or coincidental with the specific compound employed; and like factors well known in the medical arts.

In some embodiments, HSV RNA (*e.g.*, mRNA) vaccines compositions may be administered at dosage levels sufficient to deliver 0.0001 mg/kg to 100 mg/kg, 0.001 mg/kg to 0.05 mg/kg, 0.005 mg/kg to 0.05 mg/kg, 0.001 mg/kg to 0.005 mg/kg, 0.05 mg/kg to 0.5 mg/kg, 0.01 mg/kg to 50 mg/kg, 0.1 mg/kg to 40 mg/kg, 0.5 mg/kg to 30 mg/kg, 0.01 mg/kg to 10 mg/kg, 0.1 mg/kg to 10 mg/kg, or 1 mg/kg to 25 mg/kg, of subject body weight per day, one or more times a day, per week, per month, *etc.* to obtain the desired therapeutic, diagnostic, prophylactic, or imaging effect (*see e.g.*, the range of unit doses described in International Publication No WO2013078199, herein incorporated by reference in its entirety). The desired dosage may be delivered three times a day, two times a day, once a day, every other day, every third day, every week, every two weeks, every three weeks, every four weeks, every 2 months, every 3 months, every 6 months, *etc.* In certain embodiments, the desired dosage may be delivered using multiple administrations (*e.g.*, two, three, four, five, six, seven, eight, nine, ten, eleven, twelve, thirteen, fourteen, or more administrations). When multiple administrations are employed, split dosing regimens such as those described herein may be used. In exemplary embodiments, HSV RNA (*e.g.*, mRNA) vaccine compositions may be administered at dosage levels sufficient to deliver 0.0005 mg/kg to 0.01 mg/kg, *e.g.*, about 0.0005 mg/kg to about 0.0075 mg/kg, *e.g.*, about 0.0005 mg/kg, about 0.001 mg/kg, about 0.002 mg/kg, about 0.003 mg/kg, about 0.004 mg/kg, or about 0.005 mg/kg.

In some embodiments, HSV RNA (*e.g.*, mRNA) vaccine compositions may be administered once or twice (or more) at dosage levels sufficient to deliver 0.025 mg/kg to 0.250 mg/kg, 0.025 mg/kg to 0.500 mg/kg, 0.025 mg/kg to 0.750 mg/kg, or 0.025 mg/kg to 1.0 mg/kg.

In some embodiments, HSV RNA (*e.g.*, mRNA) vaccine compositions may be administered twice (*e.g.*, Day 0 and Day 7, Day 0 and Day 14, Day 0 and Day 21, Day 0 and

Day 28, Day 0 and Day 60, Day 0 and Day 90, Day 0 and Day 120, Day 0 and Day 150, Day 0 and Day 180, Day 0 and 3 months later, Day 0 and 6 months later, Day 0 and 9 months later, Day 0 and 12 months later, Day 0 and 18 months later, Day 0 and 2 years later, Day 0 and 5 years later, or Day 0 and 10 years later) at a total dose of or at dosage levels sufficient to deliver a total dose of 0.0100 mg, 0.025 mg, 0.050 mg, 0.075 mg, 0.100 mg, 0.125 mg, 0.150 mg, 0.175 mg, 0.200 mg, 0.225 mg, 0.250 mg, 0.275 mg, 0.300 mg, 0.325 mg, 0.350 mg, 0.375 mg, 0.400 mg, 0.425 mg, 0.450 mg, 0.475 mg, 0.500 mg, 0.525 mg, 0.550 mg, 0.575 mg, 0.600 mg, 0.625 mg, 0.650 mg, 0.675 mg, 0.700 mg, 0.725 mg, 0.750 mg, 0.775 mg, 0.800 mg, 0.825 mg, 0.850 mg, 0.875 mg, 0.900 mg, 0.925 mg, 0.950 mg, 0.975 mg, or 1.0 mg. Higher and lower dosages and frequency of administration are encompassed by the present disclosure. For example, a HSV RNA (*e.g.*, mRNA) vaccine composition may be administered three or four times.

In some embodiments, HSV RNA (*e.g.*, mRNA) vaccine compositions may be administered twice (*e.g.*, Day 0 and Day 7, Day 0 and Day 14, Day 0 and Day 21, Day 0 and Day 28, Day 0 and Day 60, Day 0 and Day 90, Day 0 and Day 120, Day 0 and Day 150, Day 0 and Day 180, Day 0 and 3 months later, Day 0 and 6 months later, Day 0 and 9 months later, Day 0 and 12 months later, Day 0 and 18 months later, Day 0 and 2 years later, Day 0 and 5 years later, or Day 0 and 10 years later) at a total dose of or at dosage levels sufficient to deliver a total dose of 0.010 mg, 0.025 mg, 0.100 mg, or 0.400 mg.

In some embodiments, the RNA (*e.g.*, mRNA) vaccine for use in a method of vaccinating a subject is administered the subject a single dosage of between 10 μg/kg and 400 μg/kg of the nucleic acid vaccine in an effective amount to vaccinate the subject. In some embodiments, the RNA (*e.g.*, mRNA) vaccine for use in a method of vaccinating a subject is administered to the subject via a single dosage of between 10 μg and 400 μg of the nucleic acid vaccine in an effective amount to vaccinate the subject.

A RNA (*e.g.*, mRNA) vaccine pharmaceutical composition described herein can be formulated into a dosage form described herein, such as an intranasal, intratracheal, or injectable (*e.g.*, intravenous, intraocular, intravitreal, intramuscular, intradermal, intracardiac, intraperitoneal, and subcutaneous).

HSV RNA (*e.g.*, mRNA) *vaccine formulations and methods of use*

Some aspects of the present disclosure provide formulations of the HSV RNA (*e.g.*, mRNA) vaccine, wherein the HSV RNA vaccine is formulated in an effective amount to produce an antigen specific immune response in a subject (*e.g.*, production of antibodies

specific to an anti-HSV antigenic polypeptide). "An effective amount" is a dose of a HSV RNA (*e.g.*, mRNA) vaccine effective to produce an antigen-specific immune response. Also provided herein are methods of inducing an antigen-specific immune response in a subject.

In some embodiments, the antigen-specific immune response is characterized by measuring an anti-HSV antigenic polypeptide antibody titer produced in a subject administered a HSV RNA (*e.g.*, mRNA) vaccine as provided herein. An antibody titer is a measurement of the amount of antibodies within a subject, for example, antibodies that are specific to a particular antigen (*e.g.*, an anti-HSV antigenic polypeptide) or epitope of an antigen. Antibody titer is typically expressed as the inverse of the greatest dilution that provides a positive result. Enzyme-linked immunosorbent assay (ELISA) is a common assay for determining antibody titers, for example.

In some embodiments, an antibody titer is used to assess whether a subject has had an infection or to determine whether immunizations are required. In some embodiments, an antibody titer is used to determine the strength of an autoimmune response, to determine whether a booster immunization is needed, to determine whether a previous vaccine was effective, and to identify any recent or prior infections. In accordance with the present disclosure, an antibody titer may be used to determine the strength of an immune response induced in a subject by the HSV RNA (*e.g.*, mRNA) vaccine.

In some embodiments, an anti-HSV antigenic polypeptide antibody titer produced in a subject is increased by at least 1 log relative to a control. For example, anti-HSV antigenic polypeptide antibody titer produced in a subject may be increased by at least 1.5, at least 2, at least 2.5, or at least 3 log relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased by 1, 1.5, 2, 2.5 or 3 log relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased by 1-3 log relative to a control. For example, the anti-HSV antigenic polypeptide antibody titer produced in a subject may be increased by 1-1.5, 1-2, 1-2.5, 1-3, 1.5-2, 1.5-2.5, 1.5-3, 2-2.5, 2-3, or 2.5-3 log relative to a control.

In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in a subject is increased at least 2 times relative to a control. For example, the anti-HSV antigenic polypeptide antibody titer produced in a subject may be increased at least 3 times, at least 4 times, at least 5 times, at least 6 times, at least 7 times, at least 8 times, at least 9 times, or at least 10 times relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in the subject is increased 2, 3, 4, 5 ,6, 7, 8, 9, or 10 times relative to a control. In some embodiments, the anti-HSV antigenic polypeptide antibody

titer produced in a subject is increased 2-10 times relative to a control. For example, the anti-HSV antigenic polypeptide antibody titer produced in a subject may be increased 2-10, 2-9, 2-8, 2-7, 2-6, 2-5, 2-4, 2-3, 3-10, 3-9, 3-8, 3-7, 3-6, 3-5, 3-4, 4-10, 4-9, 4-8, 4-7, 4-6, 4-5, 5-10, 5-9, 5-8, 5-7, 5-6, 6-10, 6-9, 6-8, 6-7, 7-10, 7-9, 7-8, 8-10, 8-9, or 9-10 times relative to a control.

A control, in some embodiments, is the anti-HSV antigenic polypeptide antibody titer produced in a subject who has not been administered a HSV RNA (*e.g.*, mRNA) vaccine. In some embodiments, a control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has been administered a live attenuated HSV vaccine. An attenuated vaccine is a vaccine produced by reducing the virulence of a viable (live). An attenuated virus is altered in a manner that renders it harmless or less virulent relative to live, unmodified virus. In some embodiments, a control is an anti-HSV antigenic polypeptide antibody titer produced in a subject administered inactivated HSV vaccine. In some embodiments, a control is an anti-HSV antigenic polypeptide antibody titer produced in a subject administered a recombinant or purified HSV protein vaccine. Recombinant protein vaccines typically include protein antigens that either have been produced in a heterologous expression system (*e.g.*, bacteria or yeast) or purified from large amounts of the pathogenic organism. In some embodiments, a control is an anti-HSV antigenic polypeptide antibody titer produced in a subject who has been administered a HSV virus-like particle (VLP) vaccine (*e.g.*, particles that contain viral capsid protein but lack a viral genome and, therefore, cannot replicate/produce progeny virus). In some embodiments, the control is a VLP HSV vaccine that comprises prefusion or postfusion F proteins, or that comprises a combination of the two.

In some embodiments, an effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a dose that is reduced compared to the standard of care dose of a recombinant HSV protein vaccine. A "standard of care," as provided herein, refers to a medical or psychological treatment guideline and can be general or specific. "Standard of care" specifies appropriate treatment based on scientific evidence and collaboration between medical professionals involved in the treatment of a given condition. It is the diagnostic and treatment process that a physician/ clinician should follow for a certain type of patient, illness or clinical circumstance. A "standard of care dose," as provided herein, refers to the dose of a recombinant or purified HSV protein vaccine, or a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine, that a physician/clinician or other medical professional would administer to a subject to treat or prevent HSV, or a HSV-related condition, while

following the standard of care guideline for treating or preventing HSV, or a HSV-related condition.

In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in a subject administered an effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered a standard of care dose of a recombinant or purified HSV protein vaccine, or a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, an effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a dose equivalent to an at least 2-fold reduction in a standard of care dose of a recombinant or purified HSV protein vaccine. For example, an effective amount of a HSV RNA (*e.g.*, mRNA) vaccine may be a dose equivalent to an at least 3-fold, at least 4-fold, at least 5-fold, at least 6-fold, at least 7-fold, at least 8-fold, at least 9-fold, or at least 10-fold reduction in a standard of care dose of a recombinant or purified HSV protein vaccine. In some embodiments, an effective amount of a HSV RNA vaccine is a dose equivalent to an at least at least 100-fold, at least 500-fold, or at least 1000-fold reduction in a standard of care dose of a recombinant or purified HSV protein vaccine. In some embodiments, an effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a dose equivalent to a 2-, 3-, 4-, 5-, 6-, 7-, 8-, 9-, 10-, 20-, 50-, 100-, 250-, 500-, or 1000-fold reduction in a standard of care dose of a recombinant or purified HSV protein vaccine. In some embodiments, the anti-HSV antigenic polypeptide antibody titer produced in a subject administered an effective amount of a HSV RNA vaccine is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or protein HSV protein vaccine, or a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine. In some embodiments, an effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a dose equivalent to a 2-fold to 1000-fold (*e.g.*, 2-fold to 100-fold, 10-fold to 1000-fold) reduction in the standard of care dose of a recombinant or purified HSV protein vaccine, wherein the anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, or a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a dose equivalent to a 2 to 1000-, 2 to 900-, 2 to 800-, 2 to 700-, 2 to 600-, 2 to 500-, 2 to 400-, 2 to 300-, 2 to 200-, 2 to 100-, 2 to 90-, 2 to 80-, 2 to 70-, 2 to 60-, 2 to 50-, 2 to 40-, 2 to 30-, 2 to 20-, 2 to 10-, 2 to 9-, 2 to 8-, 2 to 7-, 2 to 6-, 2 to 5-, 2 to 4-, 2 to 3-, 3 to 1000-, 3 to 900-,

3 to 800-, 3 to 700-, 3 to 600-, 3 to 500-, 3 to 400-, 3 to 3 to 00-, 3 to 200-, 3 to 100-, 3 to 90-, 3 to 80-, 3 to 70-, 3 to 60-, 3 to 50-, 3 to 40-, 3 to 30-, 3 to 20-, 3 to 10-, 3 to 9-, 3 to 8-, 3 to 7-, 3 to 6-, 3 to 5-, 3 to 4-, 4 to 1000-, 4 to 900-, 4 to 800-, 4 to 700-, 4 to 600-, 4 to 500-, 4 to 400-, 4 to 4 to 00-, 4 to 200-, 4 to 100-, 4 to 90-, 4 to 80-, 4 to 70-, 4 to 60-, 4 to 50-, 4 to 40-, 4 to 30-, 4 to 20-, 4 to 10-, 4 to 9-, 4 to 8-, 4 to 7-, 4 to 6-, 4 to 5-, 4 to 4-, 5 to 1000-, 5 to 900-, 5 to 800-, 5 to 700-, 5 to 600-, 5 to 500-, 5 to 400-, 5 to 300-, 5 to 200-, 5 to 100-, 5 to 90-, 5 to 80-, 5 to 70-, 5 to 60-, 5 to 50-, 5 to 40-, 5 to 30-, 5 to 20-, 5 to 10-, 5 to 9-, 5 to 8-, 5 to 7-, 5 to 6-, 6 to 1000-, 6 to 900-, 6 to 800-, 6 to 700-, 6 to 600-, 6 to 500-, 6 to 400-, 6 to 300-, 6 to 200-, 6 to 100-, 6 to 90-, 6 to 80-, 6 to 70-, 6 to 60-, 6 to 50-, 6 to 40-, 6 to 30-, 6 to 20-, 6 to 10-, 6 to 9-, 6 to 8-, 6 to 7-, 7 to 1000-, 7 to 900-, 7 to 800-, 7 to 700-, 7 to 600-, 7 to 500-, 7 to 400-, 7 to 300-, 7 to 200-, 7 to 100-, 7 to 90-, 7 to 80-, 7 to 70-, 7 to 60-, 7 to 50-, 7 to 40-, 7 to 30-, 7 to 20-, 7 to 10-, 7 to 9-, 7 to 8-, 8 to 1000-, 8 to 900-, 8 to 800-, 8 to 700-, 8 to 600-, 8 to 500-, 8 to 400-, 8 to 300-, 8 to 200-, 8 to 100-, 8 to 90-, 8 to 80-, 8 to 70-, 8 to 60-, 8 to 50-, 8 to 40-, 8 to 30-, 8 to 20-, 8 to 10-, 8 to 9-, 9 to 1000-, 9 to 900-, 9 to 800-, 9 to 700-, 9 to 600-, 9 to 500-, 9 to 400-, 9 to 300-, 9 to 200-, 9 to 100-, 9 to 90-, 9 to 80-, 9 to 70-, 9 to 60-, 9 to 50-, 9 to 40-, 9 to 30-, 9 to 20-, 9 to 10-, 10 to 1000-, 10 to 900-, 10 to 800-, 10 to 700-, 10 to 600-, 10 to 500-, 10 to 400-, 10 to 300-, 10 to 200-, 10 to 100-, 10 to 90-, 10 to 80-, 10 to 70-, 10 to 60-, 10 to 50-, 10 to 40-, 10 to 30-, 10 to 20-, 20 to 1000-, 20 to 900-, 20 to 800-, 20 to 700-, 20 to 600-, 20 to 500-, 20 to 400-, 20 to 300-, 20 to 200-, 20 to 100-, 20 to 90-, 20 to 80-, 20 to 70-, 20 to 60-, 20 to 50-, 20 to 40-, 20 to 30-, 30 to 1000-, 30 to 900-, 30 to 800-, 30 to 700-, 30 to 600-, 30 to 500-, 30 to 400-, 30 to 300-, 30 to 200-, 30 to 100-, 30 to 90-, 30 to 80-, 30 to 70-, 30 to 60-, 30 to 50-, 30 to 40-, 40 to 1000-, 40 to 900-, 40 to 800-, 40 to 700-, 40 to 600-, 40 to 500-, 40 to 400-, 40 to 300-, 40 to 200-, 40 to 100-, 40 to 90-, 40 to 80-, 40 to 70-, 40 to 60-, 40 to 50-, 50 to 1000-, 50 to 900-, 50 to 800-, 50 to 700-, 50 to 600-, 50 to 500-, 50 to 400-, 50 to 300-, 50 to 200-, 50 to 100-, 50 to 90-, 50 to 80-, 50 to 70-, 50 to 60-, 60 to 1000-, 60 to 900-, 60 to 800-, 60 to 700-, 60 to 600-, 60 to 500-, 60 to 400-, 60 to 300-, 60 to 200-, 60 to 100-, 60 to 90-, 60 to 80-, 60 to 70-, 70 to 1000-, 70 to 900-, 70 to 800-, 70 to 700-, 70 to 600-, 70 to 500-, 70 to 400-, 70 to 300-, 70 to 200-, 70 to 100-, 70 to 90-, 70 to 80-, 80 to 1000-, 80 to 900-, 80 to 800-, 80 to 700-, 80 to 600-, 80 to 500-, 80 to 400-, 80 to 300-, 80 to 200-, 80 to 100-, 80 to 90-, 90 to 1000-, 90 to 900-, 90 to 800-, 90 to 700-, 90 to 600-, 90 to 500-, 90 to 400-, 90 to 300-, 90 to 200-, 90 to 100-, 100 to 1000-, 100 to 900-, 100 to 800-, 100 to 700-, 100 to 600-, 100 to 500-, 100 to 400-, 100 to 300-, 100 to 200-, 200 to 1000-, 200 to 900-, 200 to 800-, 200 to 700-, 200 to 600-, 200 to 500-, 200 to 400-, 200 to 300-, 300 to 1000-, 300 to 900-, 300 to 800-, 300 to 700-, 300 to

600-, 300 to 500-, 300 to 400-, 400 to 1000-, 400 to 900-, 400 to 800-, 400 to 700-, 400 to 600-, 400 to 500-, 500 to 1000-, 500 to 900-, 500 to 800-, 500 to 700-, 500 to 600-, 600 to 1000-, 600 to 900-, 600 to 800-, 600 to 700-, 700 to 1000-, 700 to 900-, 700 to 800-, 800 to 1000-, 800 to 900-, or 900 to 1000-fold reduction in the standard of care dose of a recombinant HSV protein vaccine. In some embodiments, such as the foregoing, the anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, or a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine. In some embodiments, the effective amount is a dose equivalent to (or equivalent to and at least) a 2-, 3 -,4 -,5 -,6-, 7-, 8-, 9-, 10-, 20-, 30-, 40-, 50-, 60-, 70-, 80-, 90-, 100-, 110-, 120-, 130-, 140-, 150-, 160-, 170-, 1280-, 190-, 200-, 210-, 220-, 230-, 240-, 250-, 260-, 270-, 280-, 290-, 300-, 310-, 320-, 330-, 340-, 350-, 360-, 370-, 380-, 390-, 400-, 410-, 420-, 430-, 440-, 450-, 4360-, 470-, 480-, 490-, 500-, 510-, 520-, 530-, 540-, 550-, 560-, 5760-, 580-, 590-, 600-, 610-, 620-, 630-, 640-, 650-, 660-, 670-, 680-, 690-, 700-, 710-, 720-, 730-, 740-, 750-, 760-, 770-, 780-, 790-, 800-, 810-, 820--, 830-, 840-, 850-, 860-, 870-, 880-, 890-, 900-, 910-, 920-, 930-, 940-, 950-, 960-, 970-, 980-, 990-, or 1000-fold reduction in the standard of care dose of a recombinant HSV protein vaccine. In some embodiments, such as the foregoing, an anti-HSV antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-HSV antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant or purified HSV protein vaccine, or a live attenuated or inactivated HSV vaccine, or a HSV VLP vaccine.

In some embodiments, the effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a total dose of 50-1000 μg. In some embodiments, the effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a total dose of 50-1000, 50- 900, 50-800, 50-700, 50-600, 50-500, 50-400, 50-300, 50-200, 50-100, 50-90, 50-80, 50-70, 50-60, 60-1000, 60- 900, 60-800, 60-700, 60-600, 60-500, 60-400, 60-300, 60-200, 60-100, 60-90, 60-80, 60-70, 70-1000, 70- 900, 70-800, 70-700, 70-600, 70-500, 70-400, 70-300, 70-200, 70-100, 70-90, 70-80, 80-1000, 80-900, 80-800, 80-700, 80-600, 80-500, 80-400, 80-300, 80-200, 80-100, 80-90, 90-1000, 90-900, 90-800, 90-700, 90-600, 90-500, 90-400, 90-300, 90-200, 90-100, 100-1000, 100- 900, 100-800, 100-700, 100-600, 100-500, 100-400, 100-300, 100-200, 200-1000, 200-900, 200-800, 200-700, 200-600, 200-500, 200-400, 200-300, 300-1000, 300-900, 300-800, 300-700, 300-600, 300-500, 300-400, 400-1000, 400-900, 400-800, 400-700, 400-600, 400-500, 500-

1000, 500-900, 500-800, 500-700, 500-600, 600-1000, 600-900, 600-900, 600-700, 700-1000, 700-900, 700-800, 800-1000, 800-900, or 900-1000 μg. In some embodiments, the effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a total dose of 50, 100, 150, 200, 250, 300, 350, 400, 450, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950 or 1000 μg. In some embodiments, the effective amount is a dose of 25-500 μg administered to the subject a total of two times. In some embodiments, the effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a dose of 25-500, 25-400, 25-300, 25-200, 25-100, 25-50, 50-500, 50-400, 50-300, 50-200, 50-100, 100-500, 100-400, 100-300, 100-200, 150-500, 150-400, 150-300, 150-200, 200-500, 200-400, 200-300, 250-500, 250-400, 250-300, 300-500, 300-400, 350-500, 350-400, 400-500 or 450-500 μg administered to the subject a total of two times. In some embodiments, the effective amount of a HSV RNA (*e.g.*, mRNA) vaccine is a total dose of 25, 50, 100, 150, 200, 250, 300, 350, 400, 450, or 500 μg administered to the subject a total of two times.

Additional Embodiments

1. A herpes simplex virus (HSV) vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide having a 5' terminal cap, an open reading frame encoding at least one HSV antigenic polypeptide, and a 3' polyA tail.

2. The vaccine of paragraph 1, wherein the at least one mRNA polynucleotide is encoded by a sequence identified by any one of SEQ ID NO: 1-23 or 54-64, or a fragment of a sequence identified by any one of SEQ ID NO: 1-23 or 54-64.

3. The vaccine of paragraph 1, wherein the at least one mRNA polynucleotide comprises a sequence identified by any one of SEQ ID NO: 90-124, or a fragment of a sequence identified by any one of SEQ ID NO: 90-124.

4. The vaccine of paragraph 1, wherein the at least one antigenic polypeptide comprises a sequence identified by any one of SEQ ID NO: 24-53 or 66-77, or a fragment of a sequence identified by any one of SEQ ID NO: 24-53 or 66-77.

5. The vaccine of any one of paragraphs 1-4, wherein the 5' terminal cap is or comprises 7mG(5')ppp(5')NlmpNp.

6. The vaccine of any one of paragraphs 1-5, wherein 100% of the uracil in the open reading frame is modified to include N1-methyl pseudouridine at the 5-position of the uracil.

7. The vaccine of any one of paragraphs 1-6, wherein the vaccine is formulated in a lipid nanoparticle comprising: DLin-MC3-DMA; cholesterol; 1,2-Distearoyl-sn-glycero-3-phosphocholine (DSPC); and polyethylene glycol (PEG)2000-DMG.

8. The vaccine of paragraph 7, wherein the lipid nanoparticle further comprises trisodium citrate buffer, sucrose and water.

9. A herpes simplex virus (HSV) vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 90-124 or a fragment thereof, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 90-124 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

10. A herpes simplex virus (HSV) vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 90, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 90 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

11. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 91, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 91 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

12. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 92, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 92 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

13. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 93, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence

identified by any one of SEQ ID NO: 93 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

14. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 94, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 94 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

15. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 95, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 95 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

16. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 96, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 96 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

17. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 97, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 97 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

18. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 98, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 98 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

19. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 99, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 99 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

20. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 100, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 100 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

21. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 101, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 101 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

22. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 102, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 102 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

23. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 103, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 103 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

24. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 104, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence

identified by any one of SEQ ID NO: 104 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

25. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 105, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 105 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

26. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 106, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 106 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

27. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 107, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 107 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

28. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 108, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 108 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

29. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 109, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 109 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

30. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 110, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 110 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

31. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 111, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 111 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

32. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 112, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 112 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

33. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 113, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 113 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

34. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 114, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 114 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

35. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 115, having a 5' terminal cap

7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 115 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

36. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 116, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 116 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

37. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 117, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 117 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

38. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 118, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 118 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

39. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 119, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 119 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

40. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 120, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 120 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

41. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 121, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 121 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

42. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 122, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 122 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

43. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 123, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 123 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

44. A HSV vaccine, comprising:

at least one messenger ribonucleic acid (mRNA) polynucleotide comprising a sequence identified by any one of SEQ ID NO: 124, having a 5' terminal cap 7mG(5')ppp(5')NlmpNp and a 3' polyA tail, wherein the uracil nucleotides of the sequence identified by any one of SEQ ID NO: 124 are modified to include N1-methyl pseudouridine at the 5-position of the uracil nucleotide.

45. The vaccine of any one of paragraphs 9-44 formulated in a lipid nanoparticle comprising DLin-MC3-DMA, cholesterol, 1,2-Distearoyl-sn-glycero-3-phosphocholine (DSPC), and polyethylene glycol (PEG)2000-DMG.

This invention is not limited in its application to the details of construction and the arrangement of components set forth in the following description or illustrated in the drawings. The invention is capable of other embodiments and of being practiced or of being carried out in various ways. Also, the phraseology and terminology used herein is for the purpose of description and should not be regarded as limiting. The use of "including," "comprising," or "having," "containing," "involving," and variations thereof herein, is meant to encompass the items listed thereafter.

EXAMPLES

Example 1: Manufacture of Polynucleotides

According to the present disclosure, the manufacture of polynucleotides and/or parts or regions thereof may be accomplished utilizing the methods taught in International Publication WO2014/152027, entitled "Manufacturing Methods for Production of RNA Transcripts," the content of which is incorporated herein by reference in its entirety.

Purification methods may include those taught in International Publication WO2014/152030 and International Publication WO2014/152031, each of which is incorporated herein by reference in its entirety.

Detection and characterization methods of the polynucleotides may be performed as taught in International Publication WO2014/144039, which is incorporated herein by reference in its entirety.

Characterization of the polynucleotides of the disclosure may be accomplished using polynucleotide mapping, reverse transcriptase sequencing, charge distribution analysis, detection of RNA impurities, or any combination of two or more of the foregoing. "Characterizing" comprises determining the RNA transcript sequence, determining the purity of the RNA transcript, or determining the charge heterogeneity of the RNA transcript, for example. Such methods are taught in, for example, International Publication WO2014/144711 and International Publication WO2014/144767, the content of each of which is incorporated herein by reference in its entirety.

Example 2: Chimeric PolynucleotideSsynthesis

According to the present disclosure, two regions or parts of a chimeric polynucleotide may be joined or ligated using triphosphate chemistry. A first region or part of 100 nucleotides or less is chemically synthesized with a 5' monophosphate and terminal 3'desOH or blocked OH, for example. If the region is longer than 80 nucleotides, it may be synthesized as two strands for ligation.

If the first region or part is synthesized as a non-positionally modified region or part using *in vitro* transcription (IVT), conversion the 5'monophosphate with subsequent capping of the 3' terminus may follow.

Monophosphate protecting groups may be selected from any of those known in the art.

The second region or part of the chimeric polynucleotide may be synthesized using either chemical synthesis or IVT methods. IVT methods may include an RNA polymerase

that can utilize a primer with a modified cap. Alternatively, a cap of up to 130 nucleotides may be chemically synthesized and coupled to the IVT region or part.

For ligation methods, ligation with DNA T4 ligase, followed by treatment with DNAse should readily avoid concatenation.

The entire chimeric polynucleotide need not be manufactured with a phosphate-sugar backbone. If one of the regions or parts encodes a polypeptide, then such region or part may comprise a phosphate-sugar backbone.

Ligation is then performed using any known click chemistry, orthoclick chemistry, solulink, or other bioconjugate chemistries known to those in the art.

Synthetic route

The chimeric polynucleotide may be made using a series of starting segments. Such segments include:

(a) a capped and protected 5' segment comprising a normal 3'OH (SEG. 1);

(b) a 5' triphosphate segment, which may include the coding region of a polypeptide and a normal 3'OH (SEG. 2); and

(c) a 5' monophosphate segment for the 3' end of the chimeric polynucleotide (*e.g.*, the tail) comprising cordycepin or no 3'OH (SEG. 3).

After synthesis (chemical or IVT), segment 3 (SEG. 3) may be treated with cordycepin and then with pyrophosphatase to create the 5' monophosphate.

Segment 2 (SEG. 2) may then be ligated to SEG. 3 using RNA ligase. The ligated polynucleotide is then purified and treated with pyrophosphatase to cleave the diphosphate. The treated SEG.2-SEG. 3 construct may then be purified and SEG. 1 is ligated to the 5' terminus. A further purification step of the chimeric polynucleotide may be performed.

Where the chimeric polynucleotide encodes a polypeptide, the ligated or joined segments may be represented as: 5'UTR (SEG. 1), open reading frame or ORF (SEG. 2) and 3'UTR+PolyA (SEG. 3).

The yields of each step may be as much as 90-95%.

Example 3: PCR for cDNA Production

PCR procedures for the preparation of cDNA may be performed using 2x KAPA HIFI™ HotStart ReadyMix by Kapa Biosystems (Woburn, MA). This system includes 2x KAPA ReadyMix 12.5 µl; Forward Primer (10 µM) 0.75 µl; Reverse Primer (10 µM) 0.75 µl; Template cDNA 100 ng; and dH_20 diluted to 25.0 µl. The reaction conditions may be at 95

°C for 5 min. The reaction may be performed for 25 cycles of 98 °C for 20 sec, then 58 °C for 15 sec, then 72 °C for 45 sec, then 72 °C for 5 min, then 4 °C to termination.

The reaction may be cleaned up using Invitrogen's PURELINK™ PCR Micro Kit (Carlsbad, CA) per manufacturer's instructions (up to 5 µg). Larger reactions may require a cleanup using a product with a larger capacity. Following the cleanup, the cDNA may be quantified using the NANODROP™ and analyzed by agarose gel electrophoresis to confirm that the cDNA is the expected size. The cDNA may then be submitted for sequencing analysis before proceeding to the *in vitro* transcription reaction.

Example 4: In vitro Transcription (IVT)

The *in vitro* transcription reaction generates RNA polynucleotides. Such polynucleotides may comprise a region or part of the polynucleotides of the disclosure, including chemically modified RNA (*e.g.*, mRNA) polynucleotides. The chemically modified RNA polynucleotides can be uniformly modified polynucleotides. The *in vitro* transcription reaction utilizes a custom mix of nucleotide triphosphates (NTPs). The NTPs may comprise chemically modified NTPs, or a mix of natural and chemically modified NTPs, or natural NTPs.

A typical *in vitro* transcription reaction includes the following:

1)	Template cDNA	1.0 µg
2)	10x transcription buffer (400 mM Tris-HCl pH 8.0, 190 mM $MgCl_2$, 50 mM DTT, 10 mM Spermidine)	2.0 µl
3)	Custom NTPs (25mM each)	0.2 µl
4)	RNase Inhibitor	20 U
5)	T7 RNA polymerase	3000 U
6)	dH_20	up to 20.0 µl. and
7)	Incubation at 37 °C for 3 hr-5 hrs.	

The crude IVT mix may be stored at 4 °C overnight for cleanup the next day. 1 U of RNase-free DNase may then be used to digest the original template. After 15 minutes of incubation at 37 °C, the mRNA may be purified using Ambion's MEGACLEAR™ Kit (Austin, TX) following the manufacturer's instructions. This kit can purify up to 500 µg of RNA. Following the cleanup, the RNA polynucleotide may be quantified using the NanoDrop™ and analyzed by agarose gel electrophoresis to confirm the RNA polynucleotide is the proper size and that no degradation of the RNA has occurred.

Example 5: Enzymatic Capping

Capping of a RNA polynucleotide is performed as follows where the mixture includes: IVT RNA 60 µg-180µg and dH_20 up to 72 µl. The mixture is incubated at 65 °C for 5 minutes to denature RNA, and then is transferred immediately to ice.

The protocol then involves the mixing of 10x Capping Buffer (0.5 M Tris-HCl (pH 8.0), 60 mM KCl, 12.5 mM $MgCl_2$) (10.0 µl); 20 mM GTP (5.0 µl); 20 mM S-Adenosyl Methionine (2.5 µl); RNase Inhibitor (100 U); 2'-O-Methyltransferase (400U); Vaccinia capping enzyme (Guanylyl transferase) (40 U); dH_20 (Up to 28 µl); and incubation at 37 °C for 30 minutes for 60 µg RNA or up to 2 hours for 180 µg of RNA.

The RNA polynucleotide may then be purified using Ambion's MEGACLEAR™ Kit (Austin, TX) following the manufacturer's instructions. Following the cleanup, the RNA may be quantified using the NANODROP™ (ThermoFisher, Waltham, MA) and analyzed by agarose gel electrophoresis to confirm the RNA polynucleotide is the proper size and that no degradation of the RNA has occurred. The RNA polynucleotide product may also be sequenced by running a reverse-transcription-PCR to generate the cDNA for sequencing.

Example 6: PolyA Tailing Reaction

Without a poly-T in the cDNA, a poly-A tailing reaction must be performed before cleaning the final product. This is done by mixing capped IVT RNA (100 µl); RNase Inhibitor (20 U); 10x Tailing Buffer (0.5 M Tris-HCl (pH 8.0), 2.5 M NaCl, 100 mM $MgCl_2$)(12.0 µl); 20 mM ATP (6.0 µl); Poly-A Polymerase (20 U); dH_20 up to 123.5 µl and incubation at 37 °C for 30 min. If the poly-A tail is already in the transcript, then the tailing reaction may be skipped and proceed directly to cleanup with Ambion's MEGACLEAR™ kit (Austin, TX) (up to 500 µg). Poly-A Polymerase may be a recombinant enzyme expressed in yeast.

It should be understood that the processivity or integrity of the polyA tailing reaction may not always result in an exact size polyA tail. Hence, polyA tails of approximately between 40-200 nucleotides, *e.g.*, about 40, 50, 60, 70, 80, 90, 91, 92, 93, 94, 95, 96, 97, 98, 99, 100, 101, 102, 103, 104, 105, 106, 107, 108, 109, 110, 150-165, 155, 156, 157, 158, 159, 160, 161, 162, 163, 164 or 165 are within the scope of the present disclosure.

Example 7: Capping Assays

Protein Expression Assay

Polynucleotides (*e.g.*, mRNA) encoding a polypeptide, containing any of the caps taught herein, can be transfected into cells at equal concentrations. The amount of protein secreted into the culture medium can be assayed by ELISA at 6, 12, 24 and/or 36 hours post-transfection. Synthetic polynucleotides that secrete higher levels of protein into the medium correspond to a synthetic polynucleotide with a higher translationally-competent cap structure.

Purity Analysis Synthesis

RNA (*e.g.*, mRNA) polynucleotides encoding a polypeptide, containing any of the caps taught herein can be compared for purity using denaturing Agarose-Urea gel electrophoresis or HPLC analysis. RNA polynucleotides with a single, consolidated band by electrophoresis correspond to the higher purity product compared to polynucleotides with multiple bands or streaking bands. Chemically modified RNA polynucleotides with a single HPLC peak also correspond to a higher purity product. The capping reaction with a higher efficiency provides a more pure polynucleotide population.

Cytokine Analysis

RNA (*e.g.*, mRNA) polynucleotides encoding a polypeptide, containing any of the caps taught herein can be transfected into cells at multiple concentrations. The amount of pro-inflammatory cytokines, such as TNF-alpha and IFN-beta, secreted into the culture medium can be assayed by ELISA at 6, 12, 24, and/or 36 hours post-transfection. RNA polynucleotides resulting in the secretion of higher levels of pro-inflammatory cytokines into the medium correspond to a polynucleotides containing an immune-activating cap structure.

Capping Reaction Efficiency

RNA (*e.g.*, mRNA) polynucleotides encoding a polypeptide, containing any of the caps taught herein can be analyzed for capping reaction efficiency by LC-MS after nuclease treatment. Nuclease treatment of capped polynucleotides yield a mixture of free nucleotides and the capped 5′-5-triphosphate cap structure detectable by LC-MS. The amount of capped product on the LC-MS spectra can be expressed as a percent of total polynucleotide from the reaction and correspond to capping reaction efficiency. The cap structure with a higher capping reaction efficiency has a higher amount of capped product by LC-MS.

Example 8: Agarose Gel Electrophoresis of Modified RNA or RT PCR Products

Individual RNA polynucleotides (200-400 ng in a 20 µl volume) or reverse transcribed PCR products (200-400 ng) may be loaded into a well on a non-denaturing 1.2% Agarose E-Gel (Invitrogen, Carlsbad, CA) and run for 12-15 minutes, according to the manufacturer protocol.

Example 9: Nanodrop Modified RNA Quantification and UV Spectral Data

Chemically modified RNA polynucleotides in TE buffer (1 µl) are used for NANODROP™ UV absorbance readings to quantitate the yield of each polynucleotide from an chemical synthesis or *in vitro* transcription reaction.

Example 10: Formulation of Modified mRNA Using Lipidoids

RNA (*e.g.*, mRNA) polynucleotides may be formulated for *in vitro* experiments by mixing the polynucleotides with the lipidoid at a set ratio prior to addition to cells. *In vivo* formulation may require the addition of extra ingredients to facilitate circulation throughout the body. To test the ability of these lipidoids to form particles suitable for *in vivo* work, a standard formulation process used for siRNA-lipidoid formulations may be used as a starting point. After formation of the particle, polynucleotide is added and allowed to integrate with the complex. The encapsulation efficiency is determined using a standard dye exclusion assays.

Example 11: Immunogenicity Study

The instant study is designed to test the immunogenicity in mice of candidate HSV vaccines comprising a mRNA polynucleotide encoding one or a combination of HSV proteins.

Mice are immunized intravenously (IV), intramuscularly (IM), intranasally (IN), or intradermally (ID) with candidate HSV vaccines with and without adjuvant. A total of four immunizations are given at 3 week intervals (*i.e.,* at weeks 0, 3, 6, and 9), and sera are collected after each immunization until weeks 33–51. Serum antibody titers against glycoprotein C or glycoprotein D are determined by ELISA. Sera collected from each mouse during weeks 10–16 are pooled, and total IgGs are purified by using ammonium sulfate (Sigma) precipitation followed by DEAE (Pierce) batch purification. Following dialysis against PBS, the purified antibodies are used for immunoelectron microscopy, antibody-affinity testing, and an *in vitro* protection assay.

Example 12: HSV Rodent Challenge

The instant study is designed to test the efficacy in cotton rats of candidate HSV vaccines against a lethal challenge using a HSV vaccine comprising a chemically modified or unmodified mRNA encoding one or a combination of HSV proteins. Cotton rats are challenged with a lethal dose of HSV.

Animals are immunized intravenously (IV), intramuscularly (IM), intranasally (IN), or intradermally (ID) at week 0 and week 3 with candidate HSV vaccines with and without adjuvant. The animals are then challenged with a lethal dose of HSV on week 7 via IV, IM or ID. Endpoint is day 13 post infection, death, or euthanasia. Animals displaying severe illness as determined by >30% weight loss, extreme lethargy, or paralysis are euthanized. Body temperature and weight are assessed and recorded daily.

In experiments where a lipid nanoparticle (LNP) formulation is used, the formulation may include a cationic lipid, non-cationic lipid, PEG lipid and structural lipid in the ratios 50:10:1.5:38.5. The cationic lipid is DLin-KC2-DMA (50 mol%), the non-cationic lipid is DSPC (10 mol%), the PEG lipid is PEG-DOMG (1.5 mol%) and the structural lipid is cholesterol (38.5 mol%), for example.

Example 13: HSV Non-Human Primate Challenge

The instant study is designed to test the efficacy in African Green Monkey of candidate HSV vaccines against a non-lethal challenge using a HSV vaccine comprising a chemically modified or unmodified mRNA encoding one or a combination of HSV proteins. Animals are challenged with an attenuated dose of HSV.

Animals are immunized intravenously (IV), intramuscularly (IM), or intradermally (ID) at week 0 and week 3 with candidate HSV vaccines with and without adjuvant. The animals are then challenged with an attenuated dose of HSV on week 7 via IV, IM or ID. Endpoint is day 13 post infection. Body temperature and weight are assessed and recorded daily.

In experiments where a lipid nanoparticle (LNP) formulation is used, the formulation may include a cationic lipid, non-cationic lipid, PEG lipid and structural lipid in the ratios 50:10:1.5:38.5. The cationic lipid is DLin-KC2-DMA (50 mol%), the non-cationic lipid is DSPC (10 mol%), the PEG lipid is PEG-DOMG (1.5 mol%) and the structural lipid is cholesterol (38.5 mol%), for example.

Example 14: Microneutralization Assay

Nine serial 2-fold dilutions (1:50 –1:12,800) of simian or human serum are made in 50 µl virus growth medium (VGM) with trypsin in 96 well microtiter plates. Fifty microliters of HSV are added to the serum dilutions and allowed to incubate for 60 minutes at RT. Positive control wells of HSV without sera and negative control wells without HSV or sera are included in triplicate on each plate. While the serum-HSV mixtures incubate, a single cell suspension of cells are prepared by trypsinizing (Gibco 0.5% bovine pancrease trypsin in EDTA) a confluent monolayer and suspended cells are transferred to a 50 ml centrifuge tube, topped with sterile PBS and gently mixed. The cells are then pelleted at 200 g for 5 minutes, supernatant aspirated and cells resuspended in PBS. This procedure is repeated once and the cells are resuspended at a concentration of 3×10^5/ml in VGM with porcine trypsin. Then, 100 µl of cells are added to the serum-virus mixtures and the plates incubated at 35 °C in CO_2 for 5 days. The plates are fixed with 80% acetone in phosphate buffered saline (PBS) for 15 minutes at RT, air dried and then blocked for 30 minutes containing PBS with 0.5% gelatin and 2% FCS. An antibody to glycoprotein C or glycoprotein D is diluted in PBS with 0.5% gelatin/ 2% FCS/0.5% Tween 20 and incubated at RT for 2 hours. Wells are washed and horse radish peroxidase conjugated goat anti-mouse IgG added, followed by another 2 hour incubation. After washing, O-phenylenediamine dihydrochloride is added and the neutralization titer is defined as the titer of serum that reduced color development by 50% compared to the positive control wells.

One having ordinary skill in the art will recognize that the nucleotide sequences found in Table 1 below may be modified, for example but not limited to, for increased expression and RNA stability, and as such are covered by the present invention. Derivatives and variants thereof of the sequences found in Table 1 are considered covered by the present invention.

Each of the sequences described herein encompasses a chemically modified sequence or an unmodified sequence that includes no modified nucleotides.

Table 1: HSV Nucleic Acid Sequences

Strain	Nucleic Acid Sequence
HSV-2 gB_DX	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGAGAGGTGGTGGCTTAGTT TGCGCGCTGGTTGTCGGGGCGCTCGTAGCCGCCGTGGCGTCGGCCGCCCCTGCGGCT CCTCGCGCTAGCGGAGGCGTAGCCGCAACAGTTGCGGCGAACGGGGGTCCAGCCTC TCAGCCTCCTCCCGTCCCGAGCCCTGCGACCACCAAGGCTAGAAAGCGGAAGACCA AGAAACCGCCCAAGCGCCCCGAGGCCACCCCGCCCCCCGATGCCAACGCGACTGTC GCCGCTGGCCATGCGACGCTTCGCGCTCATCTGAGGGAGATCAAGGTTGAAAATGCT GATGCCCAATTTTACGTGTGCCCGCCCCCGACGGGCGCCACGGTTGTGCAGTTTGAA CAGCCGCGGCGCTGTCCGACGCGGCCAGAAGGCCAGAACTATACGGAGGGCATAGC

Strain	Nucleic Acid Sequence
	GGTGGTCTTTAAGGAAAACATCGCCCCGTACAAATTTAAGGCCACAATGTACTACAA AGACGTGACAGTTTCGCAAGTGTGGTTTGGCCACAGATACTCGCAGTTTATGGGAAT CTTCGAAGATAGAGCCCCTGTTCCCTTCGAGGAAGTCATCGACAAGATTAATGCCAA AGGGGTATGCCGTTCCACGGCCAAATACGTGCGCAACAATATGGAGACCACCGCCT TTCACCGGGATGATCACGAGACCGACATGGAGCTTAAGCCGGCGAAGGTCGCCACG CGTACCTCCCGGGGTTGGCACACCACAGATCTTAAGTACAATCCCTCGCGAGTTGAA GCATTCCATCGGTATGGAACTACCGTTAACTGCATCGTTGAGGAGGTGGATGCGCGG TCGGTGTACCCTTACGATGAGTTTGTGTTAGCGACCGGCGATTTTGTGTACATGTCCC CGTTTTACGGCTACCGGGAGGGGTCGCACACCGAACATACCTCGTACGCCGCTGACA GGTTCAAGCAGGTCGATGGCTTTTACGCGCGCGATCTCACCACGAAGGCCCGGGCCA CGTCACCGACGACCAGGAACTTGCTCACGACCCCCAAGTTCACCGTCGCTTGGGATT GGGTCCCAAAGCGTCCGGCGGTCTGCACGATGACCAAATGGCAGGAGGTGGACGAA ATGCTCCGCGCAGAATACGGCGGCTCCTTCCGCTTCTCGTCCGACGCCATCTCGACA ACCTTCACCACCAATCTGACCCAGTACAGTCTGTCGCGCGTTGATTTAGGAGACTGC ATTGGCCGGGATGCCCGGGAGGCCATCGACAGAATGTTTGCGCGTAAGTACAATGC CACACATATTAAGGTGGGCCAGCCGCAATACTACCTTGCCACGGGCGGCTTTCTCAT CGCGTACCAGCCCCTTCTCTCAAATACGCTCGCTGAACTGTACGTGCGGGAGTATAT GAGGGAACAGGACCGCAAGCCCCGCAATGCCACGCCTGCGCCACTACGAGAGGCGC CTTCAGCTAATGCGTCGGTGGAACGTATCAAGACCACCTCCTCAATAGAGTTCGCCC GGCTGCAATTTACGTACAACCACATCCAGCGCCACGTGAACGACATGCTGGGCCGC ATCGCTGTCGCCTGGTGCGAGCTGCAGAATCACGAGCTGACTCTTTGGAACGAGGCC CGAAAACTCAACCCCAACGCGATCGCCTCCGCAACAGTCGGTAGACGGGTGAGCGC TCGCATGCTAGGAGATGTCATGGCTGTGTCCACCTGCGTGCCCGTCGCTCCGGACAA CGTGATTGTGCAGAATTCGATGCGGGTCTTGATAATAGGCTGGAGCCTCGGTGGCCA TGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCC CCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 1)
HSV-2 gC_DX	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGCCCTTGGACGGGTAGG CCTAGCCGTGGGCCTGTGGGGCCTACTGTGGGTGGGTGTGGTCGTGGTGCTGGCCAA TGCCTCCCCCGGACGCACGATAACGGTGGGCCCGCGAGGCAACGCGAGCAATGCTG CCCCCTCCGCGTCCCCGCGGAACGCATCCGCCCCCCGAACCACACCCACGCCCCCAC AACCCCGCAAAGCGACGAAATCCAAGGCCTCCACCGCCAAACCGGCTCCGCCCCCC AAGACCGGACCCCCGAAGACATCCTCGGAGCCCGTGCGATGCAACCGCCACGACCC GCTGGCCCGGTACGGCTCGCGGGTGCAAATCCGATGCCGGTTTCCCAACTCCACGAG GACTGAGTCCCGTCTCCAGATCTGGCGTTATGCCACGGCGACGGACGCCGAAATCGG AACAGCGCCTAGCTTAGAAGAGGTGATGGTGAACGTGTCGGCCCCGCCCGGGGGCC AACTGGTGTATGACAGTGCCCCCAACCGAACGGACCCGCATGTAATCTGGGCGGAG GGCGCCGGCCCGGGCGCCAGCCCGCGCCTGTACTCGGTTGTCGGCCCGCTGGGTCGG CAGCGGCTCATCATCGAAGAGTTAACCCTGGAGACACAGGGCATGTACTATTGGGT GTGGGGCCGGACGGACCGCCCGTCCGCCTACGGGACCTGGGTCCGCGTTCGAGTATT TCGCCCTCCGTCGCTGACCATCCACCCCCACGCGGTGCTGGAGGGCCAGCCGTTTAA GGCGACGTGCACGGCCGCAACCTACTACCCGGGCAACCGCGCGGAGTTCGTCTGGTT TGAGGACGGTCGCCGCGTATTCGATCCGGCACAGATACACACGCAGACGCAGGAGA ACCCCGACGGCTTTTCCACCGTCTCCACCGTGACCTCCGCGGCCGTCGGCGGGCAGG GCCCCCCTCGCACCTTCACCTGCCAGCTGACGTGGCACCGCGACTCCGTGTCGTTCT CTCGGCGCAACGCCAGCGGCACGGCCTCGGTTCTGCCGCGGCCGACCATTACCATGG AGTTTACAGGCGACCATGCGGTCTGCACGGCCGGCTGTGTGCCCGAGGGGGTCACGT TTGCTTGGTTCCTGGGGGATGACTCCTCGCCGGCGGAAAAGGTGGCCGTCGCGTCCC AGACATCGTGCGGGCGCCCCGGCACCGCCACGATCCGCTCCACCCTGCCGGTCTCGT ACGAGCAGACCGAGTACATCTGTAGACTGGCGGGATACCCGGACGGAATTCCGGTC CTAGAGCACCACGGAAGCCACCAGCCCCCGCCGCGGGACCCAACCGAGCGGCAGGT GATCCGGGCGGTGGAGGGGGCGGGGATCGGAGTGGCTGTCCTTGTCGCGGTGGTTC TGGCCGGGACCGCGGTAGTGTACCTGACCCATGCCTCCTCGGTACGCTATCGTCGGC TGCGGTAATGATAATAGGCTGGAGCCTCGGTGGCCATGCTTCTTGCCCCTTGGGCCT CCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCCGTGGTCTTTGAATAAAGTC TGAGTGGGCGGC (SEQ ID NO: 2)
HSV-2 gD_DX	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGGGCGTTTGACCTCCGGC GTCGGGACGGCGGCCCTGCTAGTTGTCGCGGTGGGACTCCGCGTCGTCTGCGCCAAA

Strain	Nucleic Acid Sequence
HSV-2 gE_DX	TACGCCTTAGCAGACCCCTCGCTTAAGATGGCCGATCCCAATCGATTTCGCGGGAAG AACCTTCCGGTTTTGGACCAGCTGACCGACCCCCCCGGGGTGAAGCGTGTTTACCAC ATTCAGCCGAGCCTGGAGGACCCGTTCCAGCCCCCCAGCATCCCGATCACTGTGTAC TACGCAGTGCTGGAACGTGCCTGCCGCAGCGTGCTCCTACATGCCCCATCGGAGGCC CCCCAGATCGTGCGCGGGGCTTCGGACGAGGCCCGAAAGCACACGTACAACCTGAC CATCGCCTGGTATCGCATGGGAGACAATTGCGCTATCCCCATCACGGTTATGGAATA CACCGAGTGCCCCTACAACAAGTCGTTGGGGGTCTGCCCCATCCGAACGCAGCCCCG CTGGAGCTACTATGACAGCTTTAGCGCCGTCAGCGAGGATAACCTGGGATTCCTGAT GCACGCCCCCGCCTTCGAGACCGCGGGTACGTACCTGCGGCTAGTGAAGATAAACG ACTGGACGGAGATCACACAATTTATCCTGGAGCACCGGGCCCGCGCCTCCTGCAAGT ACGCTCTCCCCCTGCGCATCCCCCCGGCAGCGTGCCTCACCTCGAAGGCCTACCAAC AGGGCGTGACGGTCGACAGCATCGGGATGCTACCCCGCTTTATCCCCGAAAACCAG CGCACCGTCGCCCTATACAGCTTAAAAATCGCCGGGTGGCACGGCCCCAAGCCCCC GTACACCAGCACCCTGCTGCCGCCGGAGCTGTCCGACACCACCAACGCCACGCAAC CCGAACTCGTTCCGGAAGACCCCGAGGACTCGGCCCTCTTAGAGGATCCCGCCGGG ACGGTGTCTTCGCAGATCCCCCCAAACTGGCACATCCCGTCGATCCAGGACGTCGCA CCGCACCACGCCCCCGCCGCCCCCAGCAACCCGGGCCTGATCATCGGCGCGCTGGCC GGCAGTACCCTGGCGGTGCTGGTCATCGGCGGTATTGCGTTTTGGGTACGCCGCCGC GCTCAGATGGCCCCCAAGCGCCTACGTCTCCCCCACATCCGGGATGACGACGCGCCC CCCTCGCACCAGCCATTGTTTTACTAGTGATAATAGGCTGGAGCCTCGGTGGCCATG CTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCC GTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 3)
HSV-2 gI_DX	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGCTAGGGGGGCCGGGTT GGTTTTTTTTGTTGGAGTTTGGGTCGTAAGCTGCCTCGCGGCAGCGCCCAGAACGTC CTGGAAACGCGTAACCTCGGGCGAAGACGTGGTGTTACTCCCCGCGCCGGCGGGGC CGGAAGAACGCACTCGGGCCCACAAACTACTGTGGGCAGCGGAACCGCTGGATGCC TGCGGTCCCCTGAGGCCGTCATGGGTGGCACTGTGGCCCCCCCGACGAGTGCTTGAG ACGGTTGTCGATGCGGCGTGCATGCGCGCCCCGGAACCGCTCGCTATCGCATACAGT CCCCCGTTCCCTGCGGGCGACGAGGGACTTTATTCGGAGTTGGCGTGGCGCGATCGC GTAGCCGTGGTCAACGAGAGTTTAGTTATCTACGGGGCCCTGGAGACGGACAGTGG TCTGTACACCCTGTCAGTGGTGGGCCTATCCGACGAGGCCCGCCAAGTGGCGTCCGT GGTTCTCGTCGTCGAGCCCGCCCCTGTGCCTACCCCGACCCCCGATGACTACGACGA GGAGGATGACGCGGGCGTGAGCGAACGCACGCCCGTCAGCGTTCCCCCCCCAACAC CCCCCCGACGTCCCCCCGTCGCCCCCCCGACGCACCCTCGTGTTATCCCTGAGGTGA GCCACGTGCGGGGGGTGACGGTCCACATGGAAACCCCGGAGGCCATTCTGTTTGCG CCAGGGGAGACGTTTGGGACGAACGTCTCCATCCACGCAATTGCCCACGACGACGG TCCGTACGCCATGGACGTCGTCTGGATGCGATTTGATGTCCCGTCCTCGTGCGCCGA GATGCGGATCTATGAAGCATGTCTGTATCACCCGCAGCTGCCTGAGTGTCTGTCTCC GGCCGATGCGCCGTGCGCCGTAAGTTCGTGGGCGTACCGCCTGGCGGTCCGCAGCTA CGCCGGCTGCTCCAGGACTACGCCCCCACCTCGATGTTTTGCTGAAGCTCGCATGGA ACCGGTCCCCGGGTTGGCGTGGCTCGCATCAACTGTTAATCTGGAATTCCAGCATGC CTCTCCCCAACACGCCGGCCTCTATCTGTGTGTGGTGTATGTGGACGACCATATCCAT GCCTGGGGCCACATGACCATCTCCACAGCGGCCCAGTACCGGAATGCGGTGGTGGA ACAGCATCTCCCCCAGCGCCAGCCCGAGCCCGTAGAACCCACCCGACCGCATGTGA GAGCCCCCCCTCCCGCACCCTCCGCGAGAGGCCCGTTACGCTTAGGTGCGGTCCTGG GGGCGGCCCTGTTGCTCGCGGCCCTCGGGCTATCCGCCTGGGCGTGCATGACCTGCT GGCGCAGGCGCAGTTGGCGGGCGGTTAAAAGTCGGGCCTCGGCGACCGGCCCCACT TACATTCGAGTAGCGGATAGCGAGCTGTACGCGGACTGGAGTTCGGACTCAGAGGG CGAGCGCGACGGTTCCCTGTGGCAGGACCCTCCGGAGAGACCCGACTCACCGTCCA CAAATGGATCCGGCTTTGAGATCTTATCCCCAACGGCGCCCTCTGTATACCCCCATA GCGAAGGGCGTAAATCGCGCCGCCCGCTCACCACCTTTGGTTCAGGAAGCCCGGGA CGTCGTCACTCCCAGGCGTCCTATTCTTCCGTCTTATGGTAATGATAATAGGCTGGAG CCTCGGTGGCCATGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCT GCACCCGTACCCCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 4)

Strain	Nucleic Acid Sequence
HSV-2 SgB_DX	TTCGTGGAAGAGGACCTGCGTGTTTTCGGGGAGCTTCATTTTGTGGGGGCCCAGGTC CCCCACACAAACTACTACGACGGCATCATCGAGCTGTTTCACTACCCCCTGGGGAAC CACTGCCCCCGCGTTGTACACGTGGTCACACTGACCGCATGCCCCCGCCGCCCCGCC GTGGCGTTCACCTTGTGTCGCTCGACGCACCACGCCCACAGCCCCGCCTATCCGACC CTGGAGCTGGGTCTGGCGCGGCAGCCGCTTCTGCGGGTTCGAACGGCAACGCGCGA CTATGCCGGTCTGTATGTCCTGCGCGTATGGGTCGGCAGCGCGACGAACGCCAGCCT GTTTGTTTTGGGGGTGGCGCTCTCTGCCAACGGGACGTTTGTGTATAACGGCTCGGA CTACGGCTCCTGCGATCCGGCGCAGCTTCCCTTTTCGGCCCCGCGCCTGGGACCCTC GAGCGTATACACCCCCGGAGCCTCCCGGCCCACCCCTCCACGGACAACGACATCAC CGTCCTCCCCACGAGACCCGACCCCCGCCCCCGGGGACACAGGGACGCCTGCTCCC GCGAGCGGCGAGAGAGCCCCGCCCAATTCCACGCGATCGGCCAGCGAATCGAGACA CAGGCTAACCGTAGCCCAGGTAATCCAGATCGCCATACCGGCGTCCATCATCGCCTT TGTGTTTCTGGGCAGCTGTATCTGCTTCATCCATAGATGCCAGCGCCGATACAGGCG CCCCCGCGGCCAGATTTACAACCCCGGGGGCGTTTCCTGCGCGGTCAACGAGGCGGC CATGGCCCGCCTCGGAGCCGAGCTGCGATCCCACCCAAACACCCCCCCCAAACCCC GACGCCGTTCGTCGTCGTCCACGACCATGCCTTCCCTAACGTCGATAGCTGAGGAAT CGGAGCCAGGTCCAGTCGTGCTGCTGTCCGTCAGTCCTCGGCCCCGCAGTGGCCCGA CGGCCCCCCAAGAGGTCTAGTGATAATAGGCTGGAGCCTCGGTGGCCATGCTTCTTG CCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCCGTGGTCT TTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 5)
	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGCGCGGGGGGGGCTTAGT TTGCGCGCTGGTCGTGGGGGCGCTCGTAGCCGCGGTCGCGTCGGCGGCTCCGGCTGC CCCACGCGCTTCAGGTGGTGTCGCTGCGACCGTTGCGGCGAATGGTGGTCCCGCCAG CCAACCGCCTCCCGTCCCGAGCCCCGCGACCACTAAGGCCCGGAAGCGGAAGACCA AGAAGCCACCCAAGCGGCCCGAGGCGACTCCGCCCCCAGACGCCAACGCGACCGTC GCCGCCGGCCACGCCACTCTGCGTGCGCACCTGCGGGAAATCAAGGTCGAGAACGC GGACGCCCAGTTTTACGTGTGCCCGCCGCCGACTGGCGCCACGGTGGTGCAGTTTGA GCAACCTAGGCGCTGCCCGACGCGACCAGAGGGGCAGAACTACACCGAGGGCATAG CGGTGGTCTTTAAGGAAAACATCGCCCCGTACAAATTCAAGGCCACCATGTACTACA AAGACGTGACCGTGTCGCAGGTGTGGTTCGGCCACCGCTACTCCCAGTTTATGGGGA TATTCGAGGACCGCGCCCCCGTTCCCTTCGAAGAGGTGATTGACAAAATTAACGCCA AGGGGGTCTGCCGCAGTACGGCGAAGTACGTCCGGAACAACATGGAGACCACTGCC TTCCACCGGGACGACCACGAAACAGACATGGAGCTCAAACCGGCGAAAGTCGCCAC GCGCACGAGCCGGGGGTGGCACACCACCGACCTCAAATACAATCCTTCGCGGGTGG AAGCATTCCATCGGTATGGCACGACCGTCAACTGTATCGTAGAGGAGGTGGATGCG CGGTCGGTGTACCCCTACGATGAGTTCGTGCTGGCAACGGGCGATTTTGTGTACATG TCCCCTTTTTACGGCTACCGGGAAGGTAGTCACACCGAGCACACCAGTTACGCCGCC GACCGCTTTAAGCAAGTGGACGGCTTCTACGCGCGCGACCTCACCACAAAGGCCCG GGCCACGTCGCCGACGACCCGCAATTTGCTGACGACCCCCAAGTTTACCGTGGCCTG GGACTGGGTGCCTAAGCGACCGGCGGTCTGTACCATGACAAAGTGGCAGGAGGTGG ACGAAATGCTCCGCGCTGAATACGGTGGCTCTTTCCGCTTCTCTTCCGACGCCATCTC CACCACGTTCACCACCAACCTGACCCAATACTCGCTCTCGAGAGTCGATCTGGGAGA CTGCATTGGCCGGGATGCCCGCGAGGCAATTGACCGCATGTTCGCGCGCAAGTACA ACGCTACGCACATAAAGGTTGGCCAACCCCAGTACTACCTAGCCACGGGGGGCTTCC TCATCGCTTATCAACCCCTCCTCAGCAACACGCTCGCCGAGCTGTACGTGCGGGAAT ATATGCGGGAACAGGACCGCAAACCCCGAAACGCCACGCCCGCGCCGCTGCGGGAA GCACCGAGCGCCAACGCGTCCGTGGAGCGCATCAAGACGACATCCTCGATTGAGTTT GCTCGTCTGCAGTTTACGTATAACCACATACAGCGCCATGTAAACGACATGCTCGGG CGCATCGCCGTCGCGTGGTGCGAGCTCCAAAATCACGAGCTCACTCTGTGGAACGAG GCACGCAAGCTCAATCCCAACGCCATCGCATCCGCCACCGTAGGCCGGCGGGTGAG CGCTCGCATGCTCGGGGATGTCATGGCCGTCTCCACGTGCGTGCCCGTCGCCCCGGA CAACGTGATCGTGCAAAATAGCATGCGCGTTTCTTCGCGGCCGGGGACGTGCTACAG CCGCCCGCTGGTTAGCTTTCGGTACGAAGACCAAGGCCCGCTGATTGAGGGGCAGCT GGGTGAGAACAACGAGCTGCGCCTCACCCGCGATGCGTTAGAGCCGTGTACCGTCG GCCACCGGCGCTACTTCATCTTCGGAGGGGGATACGTATACTTCGAAGAATATGCGT ACTCTCACCAATTGAGTCGCGCCGATGTCACCACTGTTAGCACCTTCATCGACCTGA ACATCACCATGCTGGAGGACCACGAGTTCGTGCCCCTGGAGGTCTACACACGCCACG AGATCAAGGATTCCGGCCTACTGGACTACACCGAAGTCCAGAGACGAAATCAGCTG

Strain	Nucleic Acid Sequence
	CACGATCTCCGCTTTGCTGACATCGATACTGTTATCCGCGCCGACGCCAACGCCGCC ATGTTCGCAGGTCTGTGTGCGTTTTTCGAGGGTATGGGTGACTTAGGGCGCGCGGTG GGCAAGGTCGTCATGGGGGTAGTCGGGGGCGTGGTGTCGGCCGTCTCGGGCGTCTCC TCCTTTATGTCTAACCCCTGATAATAGGCTGGAGCCTCGGTGGCCATGCTTCTTGCCC CTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCCGTGGTCTTTG AATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 6)
HSV-2 SgC_DX	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGCCCTTGGACGGGTGGG CCTAGCCGTGGGCCTGTGGGGCCTGCTGTGGGTGGGTGTTGTCGTGGTGCTGGCCAA TGCCTCCCCTGGACGCACGATAACGGTGGGCCCGCGGGGGAACGCGAGCAATGCCG CCCCATCCGCGTCCCCGCGGAACGCATCCGCCCCCCGAACCACACCCACTCCCCCCC AACCCCGCAAAGCGACGAAAAGTAAGGCCTCCACCGCCAAACCGGCCCCGCCCCCC AAGACCGGGCCCCCGAAGACATCTTCTGAGCCCGTGCGCTGCAACCGCCACGACCC GCTGGCCCGGTACGGCTCGCGGGTGCAAATCCGATGTCGATTTCCCAACTCCACTCG CACGGAATCCCGCCTCCAGATCTGGCGTTATGCCACGGCGACGGACGCCGAGATTG GAACTGCGCCTAGCTTAGAGGAGGTGATGGTAAACGTGTCGGCCCCGCCCGGGGGC CAACTGGTGTATGATAGCGCACCTAACCGAACGGACCCGCACGTGATTTGGGCGGA GGGCGCCGGACCTGGCGCCTCACCGCGGCTGTACTCGGTCGTCGGGCCGCTGGGTCG GCAGAGACTTATCATCGAAGAGCTGACCCTCGAGACACAGGGCATGTATTATTGGGT GTGGGGCCGGACGGACCGCCCGTCCGCGTACGGGACCTGGGTGCGCGTTCGCGTGTT CCGCCCTCCTTCGCTGACCATCCACCCCCACGCGGTGCTGGAGGGCCAGCCGTTTAA AGCGACGTGCACCGCCGCCACCTACTACCCGGGCAACCGCGCGGAGTTCGTCTGGTT CGAGGACGGTCGCCGGGTATTCGATCCGGCCCAGATACATACGCAGACGCAGGAAA ACCCCGACGGCTTTTCCACCGTCTCCACCGTGACCTCCGCGGCCGTCGGCGGCCAGG GCCCCCCGCGCACCTTCACCTGTCAGCTGACGTGGCACCGCGACTCCGTGTCGTTCT CTCGGCGCAATGCCAGCGGCACGGCATCGGTGCTGCCACGGCCAACCATTACCATG GAGTTTACGGGCGACCATGCGGTCTGCACGGCCGGCTGTGTGCCCGAGGGGGTGAC GTTTGCCTGGTTCCTGGGGGACGACTCCTCGCCGGCCGAGAAGGTGGCCGTCGCGTC CCAGACCTCGTGCGGTCGCCCCGGCACCGCCACGATCCGCTCCACACTGCCGGTCTC GTACGAGCAGACCGAGTACATCTGCCGGCTGGCGGGATACCCGGACGGAATTCCGG TCCTAGAGCACCATGGCAGCCACCAGCCCCCGCCGCGGGACCCCACCGAACGGCAG GTGATTCGGGCAGTGGAAGGGTGATAATAGGCTGGAGCCTCGGTGGCCATGCTTCTT GCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCCGTGGTC TTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 7)
HSV-2 SgE_DX	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGCTCGCGGGGCCGGGTT GGTGTTTTTTGTTGGAGTTTGGGTCGTATCGTGCCTGGCGGCAGCACCCAGAACGTC CTGGAAACGGGTTACCTCGGGCGAGGACGTGGTGTTGCTTCCGGCGCCCGCGGGGC CGGAGGAACGCACACGGGCCCACAAACTACTGTGGGCCGCGGAACCCCTGGATGCC TGCGGTCCCCTGAGGCCGTCGTGGGTGGCGCTGTGGCCCCCGCGACGGGTGCTCGAA ACGGTCGTGGATGCGGCGTGCATGCGCGCCCCGGAACCGCTCGCCATAGCATACAG TCCCCCGTTCCCCGCGGGCGACGAGGGACTGTATTCGGAGTTGGCGTGGCGCGATCG CGTAGCCGTGGTCAACGAGAGTCTGGTCATCTACGGGGCCCTGGAGACGGACAGCG GTCTGTACACCCTGTCCGTGGTCGGCCTAAGCGACGAGGCGCGCCAAGTGGCGTCGG TGGTTCTGGTCGTGGAGCCCGCCCCTGTGCCGACCCCGACCCCCGACGACTACGACG AAGAAGACGACGCGGGCGTGAGCGAACGCACGCCGGTCAGCGTACCCCCCCCGACC CCACCCCGTCGTCCCCCCGTCGCCCCCCCTACGCACCCTCGTGTTATCCCCGAGGTGT CCCACGTGCGCGGGGTAACGGTCCATATGGAGACCCCGGAGGCCATTCTGTTTGCCC CCGGAGAGACGTTTGGGACGAACGTCTCCATCCACGCCATTGCCCATGACGACGGTC CGTACGCCATGGACGTCGTCTGGATGCGGTTTGACGTGCCGTCCTCGTGCGCCGAGA TGCGGATCTACGAAGCTTGTCTGTATCACCCGCAGCTTCCAGAATGTCTATCTCCGG CCGACGCGCCGTGCGCTGTAAGTTCCTGGGCGTACCGCCTGGCGGTCCGCAGCTACG CCGGCTGTTCCAGGACTACGCCCCCGCCGCGATGTTTTGCCGAGGCTCGCATGGAAC CGGTCCCGGGGTTGGCGTGGTTAGCCTCCACCGTCAACCTGGAATTCCAGCACGCCT CCCCTCAGCACGCCGGCCTTTACCTGTGCGTGGTGTACGTGGACGATCATATCCACG CCTGGGGCCACATGACCATCTCTACCGCGGCGCAGTACCGGAACGCGGTGGTGGAA CAGCACTTGCCCCAGCGCCAGCCTGAACCCGTCGAGCCCACCCGCCCGCACGTAAG AGCACCCCCTCCCGCGCCTTCCGCGCGCGGCCCGCTGCGCTGATAATAGGCTGGAGC CTCGGTGGCCATGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTG

Strain	Nucleic Acid Sequence
	CACCCGTACCCCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 8)
HSV-2 ICP-4	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGTCGGCGGAGCAGCGGAA GAAGAAGAAGACGACGACGACGACGCAGGGCCGCGGGGCCGAGGTCGCGATGGCG GACGAGGACGGGGGACGTCTCCGGGCCGCGGCGGAGACGACCGGCGGCCCCGGATC TCCGGATCCAGCCGACGGACCGCCGCCCACCCCGAACCCGGACCGTCGCCCCGCCG CGCGGCCCGGGTTCGGGTGGCACGGTGGGCCGGAGGAGAACGAAGACGAGGCCGA CGACGCCGCCGCCGATGCCGATGCCGACGAGGCGGCCCCGGCGTCCGGGGAGGCCG TCGACGAGCCTGCCGCGGACGGCGTCGTCTCGCCGCGGCAGCTGGCCCTGCTGGCCT CGATGGTGGACGAGGCCGTTCGCACGATCCCGTCGCCCCCCCCGGAGCGCGACGGC GCGCAAGAAGAAGCGGCCCGCTCGCCTTCTCCGCCGCGGACCCCCTCCATGCGCGCC GATTATGGCGAGGAGAACGACGACGACGACGACGACGACGATGACGACGACCGCG ACGCGGGCCGCTGGGTCCGCGGACCGGAGACGACGTCCGCGGTCCGCGGGGCGTAC CCGGACCCCATGGCCAGCCTGTCGCCGCGACCCCCGGCGCCCCGCCGACACCACCA CCACCACCACCACCGCCGCCGGCGCGCCCCCCGCCGGCGCTCGGCCGCCTCTGACTC ATCAAAATCCGGATCCTCGTCGTCGGCGTCCTCCGCCTCCTCCTCCGCCTCCTCCTCC TCGTCTGCATCCGCCTCCTCGTCTGACGACGACGACGACGACGACGCCGCCCGCGCC CCCGCCAGCGCCGCAGACCACGCCGCGGGCGGGACCCTCGGCGCGGACGACGAGGA GGCGGGGGTGCCCGCGAGGGCCCCGGGGGCGGCGCCCCGGCCGAGCCCGCCCAGG GCCGAGCCCGCCCCGGCCCGGACCCCCGCGGCGACCGCGGGCCGCCTGGAGCGCCG CCGGGCCCGCGCGGCGGTGGCCGGCCGCGACGCCACGGGCCGCTTCACGGCCGGGC GGCCCCGGCGGGTCGAGCTGGACGCCGACGCGGCCTCCGGCGCCTTCTACGCGCGC TACCGCGACGGGTACGTCAGCGGGGAGCCGTGGCCCGGGGCCGGCCCCCCGCCCCC GGGGCGCGTGCTGTACGGCGGGCTGGGCGACAGCCGCCCCGGCCTCTGGGGGGCGC CCGAGGCGGAGGAGGCGCGGGCCCGGTTCGAGGCCTCGGGCGCCCCGGCGCCCGTG TGGGCGCCCGAGCTGGGCGACGCGGCGCAGCAGTACGCCCTGATCACGCGGCTGCT GTACACGCCGGACGCGGAGGCGATGGGGTGGCTCCAGAACCCGCGCGTGGCGCCCG GGGACGTGGCGCTGGACCAGGCCTGCTTCCGGATCTCGGGCGCGGCGCGCAACAGC AGCTCCTTCATCTCCGGCAGCGTGGCGCGGGCCGTGCCCCACCTGGGGTACGCCATG GCGGCGGGCCGCTTCGGCTGGGGCCTGGCGCACGTGGCGGCCGCCGTGGCCATGAG CCGCCGCTACGACCGCGCGCAGAAGGGCTTCCTGCTGACCAGCCTGCGCCGCGCCTA CGCGCCCCTGCTGGCGCGCGAGAACGCGGCGCTGACCGGGGCGCGAACCCCCGACG ACGGCGGCGACGCCAACCGCCACGACGGCGACGACGCCCGCGGGAAGCCCGCCGCC GCCGCCGCCCCGTTGCCGTCGGCGGCGGCGTCGCCGGCCGACGAGCGCGCGGTGCC CGCCGGCTACGGCGCCGCGGGGGTGCTCGCCGCCCTGGGGCGCCTGAGCGCCGCGC CCGCCTCCGCGCCGGCCGGGGCCGACGACGACGACGACGACGACGGCGCCGGCGGT GGTGGCGGCGGCCGGCGCGCGGAGGCGGGCCGCGTGGCCGTGGAGTGCCTGGCCGC CTGCCGCGGGATCCTGGAGGCGCTGGCGGAGGGCTTCGACGGCGACCTGGCGGCCG TGCCGGGGCTGGCCGGAGCCCGGCCCGCCGCGCCCCCGCGCCCGGGGCCCGCGGGC GCGGCCGCCCCCGCCGCACGCCGACGCGCCCCGCCTGCGCGCCTGGCTGCGCGAGCT GCGGTTCGTGCGCGACGCGCTGGTGCTGATGCGCCTGCGCGGGGACCTGCGCGTGGC CGGCGGCAGCGAGGCCGCCGTGGCCGCCGTGCGCGCCGTGAGCCTGGTCGCCGGGG CCCTGGGCCCGGCGCTGCCGCGGAGCCCGCGCCTGCTGAGCTCCGCCGCCGCCGCCG CCGCGGACCTGCTCTTCCAGAACCAGAGCCTGCGCCCCCTGCTGGCCGACACCGTCG CCGCGGCCGACTCGCTCGCCGCGCCCGCCTCCGCGCCGCGGGAGGCCGCGGACGCC CCCCGCCCCGCGGCCGCCCCTCCCGCGGGGGCCGCGCCCCCCGCCCCGCCGACGCCG CCGCCGCGGCCGCCGCGCCCCGCGGCGCTGACCCGCCGGCCCGCCGAGGGCCCCGA CCCGCAGGGCGGCTGGCGCCGCCAGCCGCCGGGGCCCAGCCACACGCCGGCGCCCT CGGCCGCCGCCCTGGAGGCCTACTGCGCCCCGCGGGCCGTGGCCGAGCTCACGGAC CACCCGCTCTTCCCCGCGCCGTGGCGCCCGGCCCTCATGTTCGACCCGCGCGCGCTG GCCTCGCTGGCCGCGCGCTGCGCCGCCCCGCCCCCCGGCGGCGCGCCCGCCGCCTTC GGCCCGCTGCGCGCCTCGGGCCCGCTGCGCCGCGCGGCGGCCTGGATGCGCCAGGT GCCCGACCCGGAGGACGTGCGCGTGGTGATCCTCTACTCGCCGCTGCCGGGCGAGG ACCTGGCCGCGGGCCGCGCCGGGGGCGGGCCCCCCCCGGAGTGGTCCGCCGAGCGC GGCGGGCTGTCCTGCCTGCTGGCGGCCCTGGGCAACCGGCTCTGCGGGCCCGCCACG GCCGCCTGGGCGGGCAACTGGACCGGCGCCCCCGACGTCTCGGCGCTGGGCGCGCA GGGCGTGCTGCTGCTGTCCACGCGGGACCTGGCCTTCGCCGGCGCCGTGGAGTTCCT GGGGCTGCTGGCCGGCGCCTGCGACCGCCGCCTCATCGTCGTCAACGCCGTGCGCGC CGCGGCCTGGCCCGCCGCTGCCCCCGTGGTCTCGCGGCAGCACGCCTACCTGGCCTG

Strain	Nucleic Acid Sequence
	CGAGGTGCTGCCCGCCGTGCAGTGCGCCGTGCGCTGGCCGGCGGCGCGGGACCTGC GCCGCACCGTGCTGGCCTCCGGCCGCGTGTTCGGGCCGGGGGTCTTCGCGCGCGTGG AGGCCGCGCACGCGCGCCTGTACCCCGACGCGCCGCCGCTGCGCCTCTGCCGCGGG GCCAACGTGCGGTACCGCGTGCGCACGCGCTTCGGCCCCGACACGCTGGTGCCCATG TCCCCGCGCGAGTACCGCCGCGCCGTGCTCCCGGCGCTGGACGGCCGGGCCGCCGC CTCGGGCGCGGGCGACGCCATGGCGCCCGGCGCGCCGGACTTCTGCGAGGACGAGG CGCACTCGCACCGCGCCTGCGCGCGCTGGGGCCTGGGCGCGCCGCTGCGGCCCGTCT ACGTGGCGCTGGGGCGCGACGCCGTGCGCGGCGGCCCGGCGGAGCTGCGCGGGCCG CGGCGGGAGTTCTGCGCGCGGGCGCTGCTCGAGCCCGACGGCGACGCGCCCCCGCT GGTGCTGCGCGACGACGCGGACGCGGGCCCGCCCCCGCAGATACGCTGGGCGTCGG CCGCGGGCCGCGCGGGGACGGTGCTGGCCGCGGCGGGCGGCGGCGTGGAGGTGGTG GGGACCGCCGCGGGGCTGGCCACGCCGCCGAGGCGCGAGCCCGTGGACATGGACGC GGAGCTGGAGGACGACGACGACGGACTGTTTGGGGAGTGATTGATAATAGGCTGGAG CCTCGGTGGCCATGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCT GCACCCGTACCCCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 9)
HSV-2 SgI_DX	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGCCCGGCCGCTCGCTGCAG GGCCTGGCGATCCTGGGCCTGTGGGTCTGCGCCACCGGCCTGGTCGTCCGCGGCCCC ACGGTCAGTCTGGTCTCAGACTCACTCGTGGATGCCGGGGCCGTGGGGCCCCAGGGC TTCGTGGAAGAGGACCTGCGTGTTTTCGGGGAGCTTCATTTTGTGGGGGCCCAGGTC CCCCACACAAACTACTACGACGGCATCATCGAGCTGTTTCACTACCCCCTGGGGAAC CACTGCCCCCGCGTTGTACACGTGGTCACACTGACCGCATGCCCCCGCCGCCCCGCC GTGGCGTTCACCTTGTGTCGCTCGACGCACCACGCCCACAGCCCCGCCTATCCGACC CTGGAGCTGGGTCTGGCGCGGCAGCCGCTTCTGCGGGTTCGAACGGCAACGCGCGA CTATGCCGGTCTGTATGTCCTGCGCGTATGGGTCGGCAGCGCGACGAACGCCAGCCT GTTTGTTTTGGGGGTGGCGCTCTCTGCCAACGGGACGTTTGTGTATAACGGCTCGGA CTACGGCTCCTGCGATCCGGCGCAGCTTCCCTTTTCGGCCCCGCGCCTGGGACCCTC GAGCGTATACACCCCCGGAGCCTCCCGGCCCACCCCTCCACGGACAACGACATCCCC GTCCTCCCCTAGAGACCCGACCCCCGCCCCCGGGGACACAGGAACGCCTGCGCCCG CGAGCGGCGAGAGAGCCCCGCCCAATTCCACGCGATCGGCCAGCGAATCGAGACAC AGGCTAACCGTAGCCCAGGTAATCCAGTGATAATAGGCTGGAGCCTCGGTGGCCAT GCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCC CGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 10)
HSV-2 SgD	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGGGCGTTTGACCTCCGGC GTCGGGACGGCGGCCCTGCTAGTTGTCGCGGTGGGACTCCGCGTCGTCTGCGCCAAA TACGCCTTAGCAGACCCCTCGCTTAAGATGGCCGATCCCAATCGATTTCGCGGGAAG AACCTTCCGGTTTTGGACCAGCTGACCGACCCCCCCGGGGTGAAGCGTGTTTACCAC ATTCAGCCGAGCCTGGAGGACCCGTTCCAGCCCCCCAGCATCCCGATCACTGTGTAC TACGCAGTGCTGGAACGTGCCTGCCGCAGCGTGCTCCTACATGCCCCATCGGAGGCC CCCCAGATCGTGCGCGGGGCTTCGGACGAGGCCCGAAAGCACACGTACAACCTGAC CATCGCCTGGTATCGCATGGGAGACAATTGCGCTATCCCCATCACGGTTATGGAATA CACCGAGTGCCCCTACAACAAGTCGTTGGGGGTCTGCCCCATCCGAACGCAGCCCCG CTGGAGCTACTATGACAGCTTTAGCGCCGTCAGCGAGGATAACCTGGGATTCCTGAT GCACGCCCCCGCCTTCGAGACCGCGGGTACGTACCTGCGGCTAGTGAAGATAAACG ACTGGACGGAGATCACACAATTTATCCTGGAGCACCGGGCCCGCGCCTCCTGCAAGT ACGCTCTCCCCCTGCGCATCCCCCCGGCAGCGTGCCTCACCTCGAAGGCCTACCAAC AGGGCGTGACGGTCGACAGCATCGGGATGCTACCCCGCTTTATCCCCGAAAACCAG CGCACCGTCGCCCTATACAGCTTAAAAATCGCCGGGTGGCACGGCCCCAAGCCCCC GTACACCAGCACCCTGCTGCCGCCGGAGCTGTCCGACACCACCAACGCCACGCAAC CCGAACTCGTTCCGGAAGACCCCGAGGACTCGGCCCTCTTAGAGGATCCCGCCGGG ACGGTGTCTTCGCAGATCCCCCCAAACTGGCACATCCCGTCGATCCAGGACGTCGCG CCGCACCACGCCCCCGCCGCCCCCAGCAACCCGTGATAATAGGCTGGAGCCTCGGT GGCCATGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCG TACCCCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 11)
HSV-2 gB	ATGCGCGGGGGGGGCTTGGTTTGCGCGCTGGTCGTGGGGGCGCTGGTGGCCGCGGT GGCGTCGGCGGCCCCGGCGGCCCCCCGCGCCTCGGGCGGCGTGGCCGCGACCGTCG CGGCGAACGGGGGTCCCGCCTCCCAGCCGCCCCCCGTCCCGAGCCCCGCGACCACC AAGGCCCGGAAGCGGAAAACCAAAAAGCCGCCCAAGCGGCCCGAGGCGACCCCGC

Strain	Nucleic Acid Sequence
	CCCCCGACGCCAACGCGACCGTCGCCGCCGGCCACGCCACGCTGCGCGCGCACCTG CGGGAAATCAAGGTCGAGAACGCCGATGCCCAGTTTTACGTGTGCCCGCCCCCGAC GGGCGCCACGGTGGTGCAGTTTGAGCAGCCGCGCCGCTGCCCGACGCGCCCGGAGG GGCAGAACTACACGGAGGGCATCGCGGTGGTCTTCAAGGAGAACATCGCCCCGTAC AAATTCAAGGCCACCATGTACTACAAAGACGTGACCGTGTCGCAGGTGTGGTTCGGC CACCGCTACTCCCAGTTTATGGGGATATTCGAGGACCGCGCCCCCGTTCCCTTCGAG GAGGTGATCGACAAGATTAACGCCAAGGGGGTCTGCCGCTCCACGGCCAAGTACGT GCGGAACAACATGGAGACCACCGCGTTTCACCGGGACGACCACGAGACCGACATGG AGCTCAAGCCGGCGAAGGTCGCCACGCGCACGAGCCGGGGGTGGCACACCACCGAC CTCAAGTACAACCCCTCGCGGGTGGAGGCGTTCCATCGGTACGGCACGACGGTCAA CTGCATCGTCGAGGAGGTGGACGCGCGGTCGGTGTACCCGTACGATGAGTTTGTGCT GGCGACGGGCGACTTTGTGTACATGTCCCCGTTTTACGGCTACCGGGAGGGGTCGCA CACCGAGCACACCAGCTACGCCGCCGACCGCTTCAAGCAGGTCGACGGCTTCTACG CGCGCGACCTCACCACGAAGGCCCGGGCCACGTCGCCGACGACCCGCAACTTGCTG ACGACCCCCAAGTTTACCGTGGCCTGGGACTGGGTGCCGAAGCGACCGGCGGTCTG CACCATGACCAAGTGGCAGGAGGTGGACGAGATGCTCCGCGCCGAGTACGGCGGCT CCTTCCGCTTCTCCTCCGACGCCATCTCGACCACCTTCACCACCAACCTGACCCAGTA CTCGCTCTCGCGCGTCGACCTGGGCGACTGCATCGGCCGGGATGCCCGCGAGGCCAT CGACCGCATGTTTGCGCGCAAGTACAACGCCACGCACATCAAGGTGGGCCAGCCGC AGTACTACCTGGCCACGGGGGGCTTCCTCATCGCGTACCAGCCCCTCCTCAGCAACA CGCTCGCCGAGCTGTACGTGCGGGAGTACATGCGGGAGCAGGACCGCAAGCCCCGG AATGCCACGCCCGCGCCACTGCGGGAGGCGCCCAGCGCCAACGCGTCCGTGGAGCG CATCAAGACCACCTCCTCGATCGAGTTCGCCCGGCTGCAGTTTACGTATAACCACAT ACAGCGCCACGTGAACGACATGCTGGGGCGCATCGCCGTCGCGTGGTGCGAGCTGC AGAACCACGAGCTGACTCTCTGGAACGAGGCCCGCAAGCTCAACCCCAACGCCATC GCCTCCGCCACCGTCGGCCGGCGGGTGAGCGCGCGCATGCTCGGAGACGTCATGGC CGTCTCCACGTGCGTGCCCGTCGCCCCGGACAACGTGATCGTGCAGAACTCGATGCG CGTCAGCTCGCGGCCGGGGACGTGCTACAGCCGCCCCCTGGTCAGCTTTCGGTACGA AGACCAGGGCCCGCTGATCGAGGGGCAGCTGGGCGAGAACAACGAGCTGCGCCTCA CCCGCGACGCGCTCGAGCCGTGCACCGTGGGCCACCGGCGCTACTTCATCTTCGGCG GGGGCTACGTGTACTTCGAGGAGTACGCGTACTCTCACCAGCTGAGTCGCGCCGACG TCACCACCGTCAGCACCTTCATCGACCTGAACATCACCATGCTGGAGGACCACGAGT TTGTGCCCCTGGAGGTCTACACGCGCCACGAGATCAAGGACAGCGGCCTGCTGGACT ACACGGAGGTCCAGCGCCGCAACCAGCTGCACGACCTGCGCTTTGCCGACATCGAC ACGGTCATCCGCGCCGACGCCAACGCCGCCATGTTCGCGGGGCTGTGCGCGTTCTTC GAGGGGATGGGGGACTTGGGGCGCGCGGTCGGCAAGGTCGTCATGGGAGTAGTGGG GGGCGTGGTGTCGGCCGTCTCGGGCGTGTCCTCCTTTATGTCCAACCCCTTCGGGGC GCTTGCCGTGGGGCTGCTGGTCCTGGCCGGCCTGGTCGCGGCCTTCTTCGCCTTCCGC TACGTCCTGCAACTGCAACGCAATCCCATGAAGGCCCTGTATCCGCTCACCACCAAG GAACTCAAGACTTCCGACCCCGGGGGCGTGGGCGGGGAGGGGGAGGAAGGCGCGG AGGGGGGCGGGTTTGACGAGGCCAAGTTGGCCGAGGCCCGAGAAATGATCCGATAT ATGGCTTTGGTGTCGGCCATGGAGCGCACGGAACACAAGGCCAGAAAGAAGGGCAC GAGCGCCCTGCTCAGCTCCAAGGTCACCAACATGGTTCTGCGCAAGCGCAACAAAG CCAGGTACTCTCCGCTCCACAACGAGGACGAGGCCGGAGACGAAGACGAGCTCTAA (SEQ ID NO: 12)
HSV-2 gC	ATGGCCCTTGGACGGGTGGGCCTAGCCGTGGGCCTGTGGGGCCTGCTGTGGGTGGGT GTGGTCGTGGTGCTGGCCAATGCCTCCCCCGGACGCACGATAACGGTGGGCCCGCG GGGGAACGCGAGCAATGCCGCCCCCTCCGCGTCCCCGCGGAACGCATCCGCCCCCC GAACCACACCCACGCCCCCCCAACCCCGCAAGGCGACGAAAAGTAAGGCCTCCACC GCCAAACCGGCCCCGCCCCCCAAGACCGGGCCCCCGAAGACATCCTCGGAGCCCGT GCGATGCAACCGCCACGACCCGCTGGCCCGGTACGGCTCGCGGGTGCAAATCCGAT GCCGGTTTCCCAACTCCACCCGCACGGAGTCCCGCCTCCAGATCTGGCGTTATGCCA CGGCGACGGACGCCGAGATCGGAACGGCGCCTAGCTTAGAGGAGGTGATGGTAAAC GTGTCGGCCCCGCCCGGGGGCCAACTGGTGTATGACAGCGCCCCCAACCGAACGGA CCCGCACGTGATCTGGGCGGAGGGCGCCGGCCCGGGCGCCAGCCCGCGGCTGTACT CGGTCGTCGGGCCGCTGGGTCGGCAGCGGCTCATCATCGAAGAGCTGACCCTGGAG ACCCAGGGCATGTACTACTGGGTGTGGGGCCGGACGGACCGCCCGTCCGCGTACGG GACCTGGGTGCGCGTTCGCGTGTTCCGCCCTCCGTCGCTGACCATCCACCCCCACGC GGTGCTGGAGGGCCAGCCGTTTAAGGCGACGTGCACGGCCGCCACCTACTACCCGG

Strain	Nucleic Acid Sequence
	GCAACCGCGCGGAGTTCGTCTGGTTCGAGGACGGTCGCCGGGTATTCGATCCGGCCC AGATACACACGCAGACGCAGGAGAACCCCGACGGCTTTTCCACCGTCTCCACCGTG ACCTCCGCGGCCGTCGGCGGCCAGGGCCCCCCGCGCACCTTCACCTGCCAGCTGACG TGGCACCGCGACTCCGTGTCGTTCTCTCGGCGCAACGCCAGCGGCACGGCATCGGTG CTGCCGCGGCCAACCATTACCATGGAGTTTACGGGCGACCATGCGGTCTGCACGGCC GGCTGTGTGCCCGAGGGGGTGACGTTTGCCTGGTTCCTGGGGGACGACTCCTCGCCG GCGGAGAAGGTGGCCGTCGCGTCCCAGACATCGTGCGGGCGCCCCGGCACCGCCAC GATCCGCTCCACCCTGCCGGTCTCGTACGAGCAGACCGAGTACATCTGCCGGCTGGC GGGATACCCGGACGGAATTCCGGTCCTAGAGCACCACGGCAGCCACCAGCCCCCGC CGCGGGACCCCACCGAGCGGCAGGTGATCCGGGCGGTGGAGGGGGCGGGGATCGG AGTGGCTGTCCTTGTCGCGGTGGTTCTGGCCGGGACCGCGGTAGTGTACCTCACCCA CGCCTCCTCGGTGCGCTATCGTCGGCTGCGGTAA (SEQ ID NO: 13)
HSV-2 gD	ATGGGGCGTTTGACCTCCGGCGTCGGGACGGCGGCCCTGCTAGTTGTCGCGGTGGGA CTCCGCGTCGTCTGCGCCAAATACGCCTTAGCAGACCCCTCGCTTAAGATGGCCGAT CCCAATCGATTTCGCGGGAAGAACCTTCCGGTTTTGGACCAGCTGACCGACCCCCCC GGGGTGAAGCGTGTTTACCACATTCAGCCGAGCCTGGAGGACCCGTTCCAGCCCCCC AGCATCCCGATCACTGTGTACTACGCAGTGCTGGAACGTGCCTGCCGCAGCGTGCTC CTACATGCCCCATCGGAGGCCCCCCAGATCGTGCGCGGGGCTTCGGACGAGGCCCG AAAGCACACGTACAACCTGACCATCGCCTGGTATCGCATGGGAGACAATTGCGCTAT CCCCATCACGGTTATGGAATACACCGAGTGCCCCTACAACAAGTCGTTGGGGGTCTG CCCCATCCGAACGCAGCCCCGCTGGAGCTACTATGACAGCTTTAGCGCCGTCAGCGA GGATAACCTGGGATTCCTGATGCACGCCCCCGCCTTCGAGACCGCGGGTACGTACCT GCGGCTAGTGAAGATAAACGACTGGACGGAGATCACACAATTTATCCTGGAGCACC GGGCCCGCGCCTCCTGCAAGTACGCTCTCCCCCTGCGCATCCCCCCGGCAGCGTGCC TCACCTCGAAGGCCTACCAACAGGGCGTGACGGTCGACAGCATCGGGATGCTACCC CGCTTTATCCCCGAAAACCAGCGCACCGTCGCCCTATACAGCTTAAAAATCGCCGGG TGGCACGGCCCCAAGCCCCCGTACACCAGCACCCTGCTGCCGCCGGAGCTGTCCGAC ACCACCAACGCCACGCAACCCGAACTCGTTCCGGAAGACCCCGAGGACTCGGCCCT CTTAGAGGATCCCGCCGGGACGGTGTCTTCGCAGATCCCCCCAAACTGGCACATCCC GTCGATCCAGGACGTCGCGCCGCACCACGCCCCCGCCGCCCCCAGCAACCCGGGCC TGATCATCGGCGCGCTGGCCGGCAGTACCCTGGCGGTGCTGGTCATCGGCGGTATTG CGTTTTGGGTACGCCGCCGCGCTCAGATGGCCCCCAAGCGCCTACGTCTCCCCCACA TCCGGGATGACGACGCGCCCCCCTCGCACCAGCCATTGTTTTACTAG (SEQ ID NO: 14)
HSV-2 gE	ATGGCTCGCGGGGCCGGGTTGGTGTTTTTTGTTGGAGTTTGGGTCGTATCGTGCCTGG CGGCAGCACCCAGAACGTCCTGGAAACGGGTAACCTCGGGCGAGGACGTGGTGTTG CTTCCGGCGCCCGCGGGGCCGGAGGAACGCACCCGGGCCCACAAACTACTGTGGGC CGCGGAACCCCTGGATGCCTGCGGTCCCCTGCGCCCGTCGTGGGTGGCGCTGTGGCC CCCCCGACGGGTGCTCGAGACGGTCGTGGATGCGGCGTGCATGCGCGCCCCGGAAC CGCTCGCCATAGCATACAGTCCCCCGTTCCCCGCGGGCGACGAGGGACTGTATTCGG AGTTGGCGTGGCGCGATCGCGTAGCCGTGGTCAACGAGAGTCTGGTCATCTACGGG GCCCTGGAGACGGACAGCGGTCTGTACACCCTGTCCGTGGTCGGCCTAAGCGACGA GGCGCGCCAAGTGGCGTCGGTGGTTCTGGTCGTGGAGCCCGCCCCTGTGCCGACCCC GACCCCCGACGACTACGACGAAGAAGACGACGCGGGCGTGAGCGAACGCACGCCG GTCAGCGTTCCCCCCCCAACCCCCCCCCGTCGTCCCCCCGTCGCCCCCCCGACGCAC CCTCGTGTTATCCCCGAGGTGTCCCACGTGCGCGGGGTAACGGTCCATATGGAGACC CCGGAGGCCATTCTGTTTGCCCCCGGGGAGACGTTTGGGACGAACGTCTCCATCCAC GCCATTGCCCACGACGACGGTCCGTACGCCATGGACGTCGTCTGGATGCGGTTTGAC GTGCCGTCCTCGTGCGCCGAGATGCGGATCTACGAAGCTTGTCTGTATCACCCGCAG CTTCCAGAGTGTCTATCTCCGGCCGACGCGCCGTGCGCCGTAAGTTCCTGGGCGTAC CGCCTGGCGGTCCGCAGCTACGCCGGCTGTTCCAGGACTACGCCCCCGCCGCGATGT TTTGCCGAGGCTCGCATGGAACCGGTCCCGGGGTTGGCGTGGCTGGCCTCCACCGTC AATCTGGAATTCCAGCACGCCTCCCCCCAGCACGCCGGCCTCTACCTGTGCGTGGTG TACGTGGACGATCATATCCACGCCTGGGGCCACATGACCATCAGCACCGCGGCGCA GTACCGGAACGCGGTGGTGGAACAGCACCTCCCCCAGCGCCAGCCCGAGCCCGTCG AGCCCACCCGCCCGCACGTGAGAGCCCCCCCTCCCGCGCCCTCCGCGCGCGGCCCGC TGCGCCTCGGGGCGGTGCTGGGGGCGGCCCTGTTGCTGGCCGCCCTCGGGCTGTCCG CGTGGGCGTGCATGACCTGCTGGCGCAGGCGCTCCTGGCGGGCGGTTAAAAGCCGG GCCTCGGCGACGGGCCCCACTTACATTCGCGTGGCGGACAGCGAGCTGTACGCGGA

Strain	Nucleic Acid Sequence
	CTGGAGTTCGGACAGCGAGGGGGAGCGCGACGGGTCCCTGTGGCAGGACCCTCCGG AGAGACCCGACTCTCCCTCCACAAATGGATCCGGCTTTGAGATCTTATCACCAACGG CTCCGTCTGTATACCCCCATAGCGAGGGGCGTAAATCTCGCCGCCCGCTCACCACCT TTGGTTCGGGAAGCCCGGGCCGTCGTCACTCCCAGGCCTCCTATTCGTCCGTCCTCTG GTAA (SEQ ID NO: 15)
HSV-2 gI	ATGCCCGGCCGCTCGCTGCAGGGCCTGGCGATCCTGGGCCTGTGGGTCTGCGCCACC GGCCTGGTCGTCCGCGGCCCCACGGTCAGTCTGGTCTCAGACTCACTCGTGGATGCC GGGGCCGTGGGGCCCCAGGGCTTCGTGGAAGAGGACCTGCGTGTTTTCGGGGAGCT TCATTTTGTGGGGGCCCAGGTCCCCCACACAAACTACTACGACGGCATCATCGAGCT GTTTCACTACCCCCTGGGGAACCACTGCCCCCGCGTTGTACACGTGGTCACACTGAC CGCATGCCCCCGCCGCCCCGCCGTGGCGTTCACCTTGTGTCGCTCGACGCACCACGC CCACAGCCCCGCCTATCCGACCCTGGAGCTGGGTCTGGCGCGGCAGCCGCTTCTGCG GGTTCGAACGGCAACGCGCGACTATGCCGGTCTGTATGTCCTGCGCGTATGGGTCGG CAGCGCGACGAACGCCAGCCTGTTTGTTTTGGGGGTGGCGCTCTCTGCCAACGGGAC GTTTGTGTATAACGGCTCGGACTACGGCTCCTGCGATCCGGCGCAGCTTCCCTTTTCG GCCCCGCGCCTGGGACCCTCGAGCGTATACACCCCCGGAGCCTCCCGGCCCACCCCT CCACGGACAACGACATCCCCGTCCTCCCCCCCGAGACCCGACCCCCGCCCCCGGGGA CACAGGGACGCCCGCGCCCGCGAGCGGCGAGAGAGCCCCGCCCAATTCCACGCGAT CGGCCAGCGAATCGAGACACAGGCTAACCGTAGCCCAGGTAATCCAGATCGCCATA CCGGCGTCCATCATCGCCTTTGTGTTTCTGGGCAGCTGTATCTGCTTCATCCATAGAT GCCAGCGCCGATACAGGCGCCCCCGCGGCCAGATTTACAACCCCGGGGGCGTTTCCT GCGCGGTCAACGAGGCGGCCATGGCCCGCCTCGGAGCCGAGCTGCGATCCCACCCA AACACCCCCCCCAAACCCCGACGCCGTTCGTCGTCGTCCACGACCATGCCTTCCCTA ACGTCGATAGCTGAGGAATCGGAGCCAGGTCCAGTCGTGCTGCTGTCCGTCAGTCCT CGGCCCCGCAGTGGCCCGACGGCCCCCCAAGAGGTCTAG (SEQ ID NO: 16)
ICP0-2	Based on strain HG52 (inactivated by deletion of the nuclear localization signal and zinc-binding ring finger)	ATGGAACCCCGGCCCGGCACGAGCTCCCGGGCGGACCCCGGCCCCGAGCGGCCGCC GCGGCAGACCCCCGGCACGCAGCCCGCCGCCCCGCACGCCTGGGGGATGCTCAACG ACATGCAGTGGCTCGCCAGCAGCGACTCGGAGGAGGAGACCGAGGTGGGAATCTCT GACGACGACCTTCACCGCGACTCCACCTCCGAGGCGGGCAGCACGGACACGGAGAT GTTCGAGGCGGGCCTGATGGACGCGGCCACGCCCCCGGCCCGGCCCCCGGCCGAGC GCCAGGGCAGCCCCACGCCCGCCGACGCGCAGGGATCCTGTGGGGGTGGGCCCGTG GGTGAGGAGGAAGCGGAAGCGGGAGGGGGGGGCGACGTGAACACCCCGGTGGCGT ACCTGATAGTGGGCGTGACCGCCAGCGGGTCGTTCAGCACCATCCCGATAGTGAAC GACCCCCGGACCCGCGTGGAGGCCGAGGCGGCCGTGCGGGCCGGCACGGCCGTGGA CTTTATCTGGACGGGCAACCCGCGGACGGCCCCGCGCTCCCTGTCGCTGGGGGGACA CACGGTCCGCGCCCTGTCGCCCACCCCCCCGTGGCCCGGCACGGACGACGAGGACG ATGACCTGGCCGACGTGGACTACGTCCCGCCCGCCCCCCGAAGAGCGCCCCGGCGC GGGGGCGGCGGTGCGGGGGCGACCCGCGGAACCTCCCAGCCCGCCGCGACCCGACC GGCGCCCCCTGGCGCCCCGCGGAGCAGCAGCAGCGGCGGCGCCCCGTTGCGGGCGG GGGTGGGATCTGGGTCTGGGGGCGGCCCTGCCGTCGCGGCCGTCGTGCCGAGAGTG GCCTCTCTTCCCCCTGCGGCCGGCGGGGGGCGCGCGCAGGCGCGGCGGGTGGGCGA AGACGCCGCGGCGGCGGAGGGCAGGACGCCCCCCGCGAGACAGCCCCGCGCGGCC CAGGAGCCCCCCATAGTCATCAGCGACTCTCCCCCGCCGTCTCCGCGCCGCCCCGCG GGCCCCGGGCCGCTCTCCTTTGTCTCCTCCTCCTCCGCACAGGTGTCCTCGGGCCCCG GGGGGGGAGGTCTGCCACAGTCGTCGGGGCGCGCCGCGCGCCCCCGCGCGGCCGTC GCCCCGCGCGTCCGGAGTCCGCCCCGCGCCGCCGCCGCCCCCGTGGTGTCTGCGAGC GCGGACGCGGCCGGGCCCGCGCCGCCCGCCGTGCCGGTGGACGCGCACCGCGCGCC CCGGTCGCGCATGACCCAGGCTCAGACCGACACCCAAGCACAGAGTCTGGGCCGGG CAGGCGCGACCGACGCGCGCGGGTCGGGAGGGCCGGGCGCGGAGGGAGGATCGGG CCCCGCGGCCTCGTCCTCCGCCTCTTCCTCCGCCGCCCCGCGCTCGCCCCTCGCCCCC CAGGGGGTGGGGGCCAAGAGGGCGGCGCCGCGCCGGGCCCCGGACTCGGACTCGG GCGACCGCGGCCACGGGCCGCTCGCCCCGGCGTCCGCGGGCGCCGCGCCCCCGTCG GCGTCTCCGTCGTCCCAGGCCGCGGTCGCCGCCGCCTCCTCCTCCTCCGCCTCCTCCT CCTCCGCCTCCTCCTCCTCCGCCTCCTCCTCCTCCGCCTCCTCCTCCTCCGCCTCCTCC TCCTCCGCCTCCTCCTCCTCCGCCTCTTCCTCTGCGGGCGGGGCTGGTGGGAGCGTCG CGTCCGCGTCCGGCGCTGGGGAGAGACGAGAAACCTCCCTCGGCCCCCGCGCTGCT GCGCCGCGGGGGCCGAGGAAGTGTGCCAGGAAGACGCGCCACGCGGAGGGCGGCC CCGAGCCCGGGGCCCGCGACCCGGCGCCCGGCCTCACGCGCTACCTGCCCATCGCG GGGGTCTCGAGCGTCGTGGCCCTGGCGCCTTACGTGAACAAGACGGTCACGGGGGA

Strain	Nucleic Acid Sequence
	CTGCCTGCCCGTCCTGGACATGGAGACGGGCCACATAGGGGCCTACGTGGTCCTCGT GGACCAGACGGGGAACGTGGCGGACCTGCTGCGGGCCGCGGCCCCCGCGTGGAGCC GCCGCACCCTGCTCCCCGAGCACGCGCGCAACTGCGTGAGGCCCCCCGACTACCCG ACGCCCCCCGCGTCGGAGTGGAACAGCCTCTGGATGACCCCGGTGGGCAACATGCT CTTTGACCAGGGCACCCTGGTGGGCGCGCTGGACTTCCACGGCCTCCGGTCGCGCCA CCCGTGGTCTCGGGAGCAGGGCGCGCCCGCGCCGGCCGGCGACGCCCCCGCGGGCC ACGGGGAGTAG (SEQ ID NO: 17)
HSV-2 SgB	ATGCGCGGGGGGGGCTTGGTTTGCGCGCTGGTCGTGGGGGCGCTGGTGGCCGCGGT GGCGTCGGCGGCCCCGGCGGCCCCCCGCGCCTCGGGCGGCGTGGCCGCGACCGTCG CGGCGAACGGGGGTCCCGCCTCCCAGCCGCCCCCCGTCCCGAGCCCCGCGACCACC AAGGCCCGGAAGCGGAAAACCAAAAAGCCGCCCAAGCGGCCCGAGGCGACCCCGC CCCCCGACGCCAACGCGACCGTCGCCGCCGGCCACGCCACGCTGCGCGCGCACCTG CGGGAAATCAAGGTCGAGAACGCCGATGCCCAGTTTTACGTGTGCCCGCCCCCGAC GGGCGCCACGGTGGTGCAGTTTGAGCAGCCGCGCCGCTGCCCGACGCGCCCGGAGG GGCAGAACTACACGGAGGGCATCGCGGTGGTCTTCAAGGAGAACATCGCCCCGTAC AAATTCAAGGCCACCATGTACTACAAAGACGTGACCGTGTCGCAGGTGTGGTTCGGC CACCGCTACTCCCAGTTTATGGGGATATTCGAGGACCGCGCCCCCGTTCCCTTCGAG GAGGTGATCGACAAGATTAACGCCAAGGGGGTCTGCCGCTCCACGGCCAAGTACGT GCGGAACAACATGGAGACCACCGCGTTTCACCGGGACGACCACGAGACCGACATGG AGCTCAAGCCGGCGAAGGTCGCCACGCGCACGAGCCGGGGGTGGCACACCACCGAC CTCAAGTACAACCCCTCGCGGGTGGAGGCGTTCCATCGGTACGGCACGACGGTCAA CTGCATCGTCGAGGAGGTGGACGCGCGGTCGGTGTACCCGTACGATGAGTTTGTGCT GGCGACGGGCGACTTTGTGTACATGTCCCCGTTTTACGGCTACCGGGAGGGGTCGCA CACCGAGCACACCAGCTACGCCGCCGACCGCTTCAAGCAGGTCGACGGCTTCTACG CGCGCGACCTCACCACGAAGGCCCGGGCCACGTCGCCGACGACCCGCAACTTGCTG ACGACCCCCAAGTTTACCGTGGCCTGGGACTGGGTGCCGAAGCGACCGGCGGTCTG CACCATGACCAAGTGGCAGGAGGTGGACGAGATGCTCCGCGCCGAGTACGGCGGCT CCTTCCGCTTCTCCTCCGACGCCATCTCGACCACCTTCACCACCAACCTGACCCAGTA CTCGCTCTCGCGCGTCGACCTGGGCGACTGCATCGGCCGGGATGCCCGCGAGGCCAT CGACCGCATGTTTGCGCGCAAGTACAACGCCACGCACATCAAGGTGGGCCAGCCGC AGTACTACCTGGCCACGGGGGGCTTCCTCATCGCGTACCAGCCCCTCCTCAGCAACA CGCTCGCCGAGCTGTACGTGCGGGAGTACATGCGGGAGCAGGACCGCAAGCCCCGG AATGCCACGCCCGCGCCACTGCGGGAGGCGCCCAGCGCCAACGCGTCCGTGGAGCG CATCAAGACCACCTCCTCGATCGAGTTCGCCCGGCTGCAGTTTACGTATAACCACAT ACAGCGCCACGTGAACGACATGCTGGGGCGCATCGCCGTCGCGTGGTGCGAGCTGC AGAACCACGAGCTGACTCTCTGGAACGAGGCCCGCAAGCTCAACCCCAACGCCATC GCCTCCGCCACCGTCGGCCGGCGGGTGAGCGCGCGCATGCTCGGAGACGTCATGGC CGTCTCCACGTGCGTGCCCGTCGCCCCGGACAACGTGATCGTGCAGAACTCGATGCG CGTCAGCTCGCGGCCGGGGACGTGCTACAGCCGCCCCCTGGTCAGCTTTCGGTACGA AGACCAGGGCCCGCTGATCGAGGGGCAGCTGGGCGAGAACAACGAGCTGCGCCTCA CCCGCGACGCGCTCGAGCCGTGCACCGTGGGCCACCGGCGCTACTTCATCTTCGGCG GGGGCTACGTGTACTTCGAGGAGTACGCGTACTCTCACCAGCTGAGTCGCGCCGACG TCACCACCGTCAGCACCTTCATCGACCTGAACATCACCATGCTGGAGGACCACGAGT TTGTGCCCCTGGAGGTCTACACGCGCCACGAGATCAAGGACAGCGGCCTGCTGGACT ACACGGAGGTCCAGCGCCGCAACCAGCTGCACGACCTGCGCTTTGCCGACATCGAC ACGGTCATCCGCGCCGACGCCAACGCCGCCATGTTCGCGGGGCTGTGCGCGTTCTTC GAGGGGATGGGGGACTTGGGGCGCGCGGTCGGCAAGGTCGTCATGGGAGTAGTGGG GGGCGTGGTGTCGGCCGTCTCGGGCGTGTCCTCCTTTATGTCCAACCCC (SEQ ID NO: 18)
HSV-2 SgC	ATGGCCCTTGGACGGGTGGGCCTAGCCGTGGGCCTGTGGGGCCTGCTGTGGGTGGGT GTGGTCGTGGTGCTGGCCAATGCCTCCCCCGGACGCACGATAACGGTGGGCCCGCG GGGGAACGCGAGCAATGCCGCCCCCTCCGCGTCCCCGCGGAACGCATCCGCCCCCC GAACCACACCCACGCCCCCCCAACCCCGCAAGGCGACGAAAAGTAAGGCCTCCACC GCCAAACCGGCCCCGCCCCCCAAGACCGGGCCCCCGAAGACATCCTCGGAGCCCGT GCGATGCAACCGCCACGACCCGCTGGCCCGGTACGGCTCGCGGGTGCAAATCCGAT GCCGGTTTCCCAACTCCACCCGCACGGAGTCCCGCCTCCAGATCTGGCGTTATGCCA CGGCGACGGACGCCGAGATCGGAACGGCGCCTAGCTTAGAGGAGGTGATGGTAAAC GTGTCGGCCCCGCCCGGGGGCCAACTGGTGTATGACAGCGCCCCCAACCGAACGGA CCCGCACGTGATCTGGGCGGAGGGCGCCGGCCCGGGCGCCAGCCCGCGGCTGTACT

Strain	Nucleic Acid Sequence
	CGGTCGTCGGGCCGCTGGGTCGGCAGCGGCTCATCATCGAAGAGCTGACCCTGGAG ACCCAGGGCATGTACTACTGGGTGTGGGGCCGGACGGACCGCCCGTCCGCGTACGG GACCTGGGTGCGCGTTCGCGTGTTCCGCCCTCCGTCGCTGACCATCCACCCCCACGC GGTGCTGGAGGGCCAGCCGTTTAAGGCGACGTGCACGGCCGCCACCTACTACCCGG GCAACCGCGCGGAGTTCGTCTGGTTCGAGGACGGTCGCCGGGTATTCGATCCGGCCC AGATACACACGCAGACGCAGGAGAACCCCGACGGCTTTTCCACCGTCTCCACCGTG ACCTCCGCGGCCGTCGGCGGCCAGGGCCCCCCGCGCACCTTCACCTGCCAGCTGACG TGGCACCGCGACTCCGTGTCGTTCTCTCGGCGCAACGCCAGCGGCACGGCATCGGTG CTGCCGCGGCCAACCATTACCATGGAGTTTACGGGCGACCATGCGGTCTGCACGGCC GGCTGTGTGCCCGAGGGGGTGACGTTTGCCTGGTTCCTGGGGGACGACTCCTCGCCG GCGGAGAAGGTGGCCGTCGCGTCCCAGACATCGTGCGGGCGCCCCGGCACCGCCAC GATCCGCTCCACCCTGCCGGTCTCGTACGAGCAGACCGAGTACATCTGCCGGCTGGC GGGATACCCGGACGGAATTCCGGTCCTAGAGCACCACGGCAGCCACCAGCCCCCGC CGCGGGACCCCACCGAGCGGCAGGTGATCCGGGCGGTGGAGGGG (SEQ ID NO: 19)
HSV-2 SgD	ATGGGGCGTTTGACCTCCGGCGTCGGGACGGCGGCCCTGCTAGTTGTCGCGGTGGGA CTCCGCGTCGTCTGCGCCAAATACGCCTTAGCAGACCCCTCGCTTAAGATGGCCGAT CCCAATCGATTTCGCGGGAAGAACCTTCCGGTTTTGGACCAGCTGACCGACCCCCCC GGGGTGAAGCGTGTTTACCACATTCAGCCGAGCCTGGAGGACCCGTTCCAGCCCCCC AGCATCCCGATCACTGTGTACTACGCAGTGCTGGAACGTGCCTGCCGCAGCGTGCTC CTACATGCCCCATCGGAGGCCCCCCAGATCGTGCGCGGGGCTTCGGACGAGGCCCG AAAGCACACGTACAACCTGACCATCGCCTGGTATCGCATGGGAGACAATTGCGCTAT CCCCATCACGGTTATGGAATACACCGAGTGCCCCTACAACAAGTCGTTGGGGGTCTG CCCCATCCGAACGCAGCCCCGCTGGAGCTACTATGACAGCTTTAGCGCCGTCAGCGA GGATAACCTGGGATTCCTGATGCACGCCCCCGCCTTCGAGACCGCGGGTACGTACCT GCGGCTAGTGAAGATAAACGACTGGACGGAGATCACACAATTTATCCTGGAGCACC GGGCCCGCGCCTCCTGCAAGTACGCTCTCCCCCTGCGCATCCCCCCGGCAGCGTGCC TCACCTCGAAGGCCTACCAACAGGGCGTGACGGTCGACAGCATCGGGATGCTACCC CGCTTTATCCCCGAAAACCAGCGCACCGTCGCCCTATACAGCTTAAAAATCGCCGGG TGGCACGGCCCCAAGCCCCCGTACACCAGCACCCTGCTGCCGCCGGAGCTGTCCGAC ACCACCAACGCCACGCAACCCGAACTCGTTCCGGAAGACCCCGAGGACTCGGCCCT CTTAGAGGATCCCGCCGGGACGGTGTCTTCGCAGATCCCCCCAAACTGGCACATCCC GTCGATCCAGGACGTCGCGCCGCACCACGCCCCCGCCGCCCCCAGCAACCCG (SEQ ID NO: 20)
HSV-2 SgE	ATGGCTCGCGGGGCCGGGTTGGTGTTTTTTGTTGGAGTTTGGGTCGTATCGTGCCTGG CGGCAGCACCCAGAACGTCCTGGAAACGGGTAACCTCGGGCGAGGACGTGGTGTTG CTTCCGGCGCCCGCGGGGCCGGAGGAACGCACCCGGGCCCACAAACTACTGTGGGC CGCGGAACCCCTGGATGCCTGCGGTCCCCTGCGCCCGTCGTGGGTGGCGCTGTGGCC CCCCCGACGGGTGCTCGAGACGGTCGTGGATGCGGCGTGCATGCGCGCCCCGGAAC CGCTCGCCATAGCATACAGTCCCCCGTTCCCCGCGGGCGACGAGGGACTGTATTCGG AGTTGGCGTGGCGCGATCGCGTAGCCGTGGTCAACGAGAGTCTGGTCATCTACGGG GCCCTGGAGACGGACAGCGGTCTGTACACCCTGTCCGTGGTCGGCCTAAGCGACGA GGCGCGCCAAGTGGCGTCGGTGGTTCTGGTCGTGGAGCCCGCCCCTGTGCCGACCCC GACCCCCGACGACTACGACGAAGAAGACGACGCGGGCGTGAGCGAACGCACGCCG GTCAGCGTTCCCCCCCCAACCCCCCCCCGTCGTCCCCCCGTCGCCCCCCCGACGCAC CCTCGTGTTATCCCCGAGGTGTCCCACGTGCGCGGGGTAACGGTCCATATGGAGACC CCGGAGGCCATTCTGTTTGCCCCCGGGGAGACGTTTGGGACGAACGTCTCCATCCAC GCCATTGCCCACGACGACGGTCCGTACGCCATGGACGTCGTCTGGATGCGGTTTGAC GTGCCGTCCTCGTGCGCCGAGATGCGGATCTACGAAGCTTGTCTGTATCACCCGCAG CTTCCAGAGTGTCTATCTCCGGCCGACGCGCCGTGCGCCGTAAGTTCCTGGGCGTAC CGCCTGGCGGTCCGCAGCTACGCCGGCTGTTCCAGGACTACGCCCCCGCCGCGATGT TTTGCCGAGGCTCGCATGGAACCGGTCCCGGGGTTGGCGTGGCTGGCCTCCACCGTC AATCTGGAATTCCAGCACGCCTCCCCCCAGCACGCCGGCCTCTACCTGTGCGTGGTG TACGTGGACGATCATATCCACGCCTGGGGCCACATGACCATCAGCACCGCGGCGCA GTACCGGAACGCGGTGGTGGAACAGCACCTCCCCCAGCGCCAGCCCGAGCCCGTCG AGCCCACCCGCCCGCACGTGAGAGCCCCCCCTCCCGCGCCCTCCGCGCGCGGCCCGC TGCGC (SEQ ID NO: 21)
HSV-2 SgI	ATGCCCGGCCGCTCGCTGCAGGGCCTGGCGATCCTGGGCCTGTGGGTCTGCGCCACC GGCCTGGTCGTCCGCGGCCCCACGGTCAGTCTGGTCTCAGACTCACTCGTGGATGCC GGGGCCGTGGGGCCCCAGGGCTTCGTGGAAGAGGACCTGCGTGTTTTCGGGGAGCT

Strain	Nucleic Acid Sequence
	TCATTTTGTGGGGGCCCAGGTCCCCCACACAAACTACTACGACGGCATCATCGAGCT GTTTCACTACCCCCTGGGGAACCACTGCCCCCGCGTTGTACACGTGGTCACACTGAC CGCATGCCCCCGCCGCCCCGCCGTGGCGTTCACCTTGTGTCGCTCGACGCACCACGC CCACAGCCCCGCCTATCCGACCCTGGAGCTGGGTCTGGCGCGGCAGCCGCTTCTGCG GGTTCGAACGGCAACGCGCGACTATGCCGGTCTGTATGTCCTGCGCGTATGGGTCGG CAGCGCGACGAACGCCAGCCTGTTTGTTTTGGGGGTGGCGCTCTCTGCCAACGGGAC GTTTGTGTATAACGGCTCGGACTACGGCTCCTGCGATCCGGCGCAGCTTCCCTTTTCG GCCCCGCGCCTGGGACCCTCGAGCGTATACACCCCCGGAGCCTCCCGGCCCACCCCT CCACGGACAACGACATCCCCGTCCTCCCCCCGAGACCCGACCCCCGCCCCCGGGGA CACAGGGACGCCCGCGCCCGCGAGCGGCGAGAGAGCCCCGCCCAATTCCACGCGAT CGGCCAGCGAATCGAGACACAGGCTAACCGTAGCCCAGGTAATCCAG (SEQ ID NO: 22)
HSV-2 ICP-4; Based on strain HG52; (inactivated by deletion of nuclear localization signal and alanine substitution for key residues in the transactivation region)	ATGTCGGCGGAGCAGCGGAAGAAGAAGAAGACGACGACGACGACGCAGGGCCGCG GGGCCGAGGTCGCGATGGCGGACGAGGACGGGGGACGTCTCCGGGCCGCGGCGGA GACGACCGGCGGCCCCGGATCTCCGGATCCAGCCGACGGACCGCCGCCCACCCCGA ACCCGGACCGTCGCCCCGCCGCGCGGCCCGGGTTCGGGTGGCACGGTGGGCCGGAG GAGAACGAAGACGAGGCCGACGACGCCGCCGCCGATGCCGATGCCGACGAGGCGG CCCCGGCGTCCGGGGAGGCCGTCGACGAGCCTGCCGCGGACGGCGTCGTCTCGCCG CGGCAGCTGGCCCTGCTGGCCTCGATGGTGGACGAGGCCGTTCGCACGATCCCGTCG CCCCCCCCGGAGCGCGACGGCGCGCAAGAAGAAGCGGCCCGCTCGCCTTCTCCGCC GCGGACCCCCTCCATGCGCGCCGATTATGGCGAGGAGAACGACGACGACGACGACG ACGACGATGACGACGACCGCGACGCGGGCCGCTGGGTCCGCGGACCGGAGACGACG TCCGCGGTCCGCGGGGCGTACCCGGACCCCATGGCCAGCCTGTCGCCGCGACCCCCG GCGCCCCGCCGACACCACCACCACCACCACCACCGCCGCCGGCGCGCCCCCCGCCG GCGCTCGGCCGCCTCTGACTCATCAAAATCCGGATCCTCGTCGTCGGCGTCCTCCGC CTCCTCCTCCGCCTCCTCCTCCTCGTCTGCATCCGCCTCCTCGTCTGACGACGACGAC GACGACGACGCCGCCCGCGCCCCCGCCAGCGCCGCAGACCACGCCGCGGGCGGGAC CCTCGGCGCGGACGACGAGGAGGCGGGGGTGCCCGCGAGGGCCCCGGGGGCGGCG CCCCGGCCGAGCCCGCCCAGGGCCGAGCCCGCCCCGGCCCGGACCCCCGCGGCGAC CGCGGGCCGCCTGGAGCGCCGCCGGGCCCGCGCGGCGGTGGCCGGCCGCGACGCCA CGGGCCGCTTCACGGCCGGGCGGCCCCGGCGGGTCGAGCTGGACGCCGACGCGGCC TCCGGCGCCTTCTACGCGCGCTACCGCGACGGGTACGTCAGCGGGGAGCCGTGGCCC GGGGCCGGCCCCCCGCCCCCGGGGCGCGTGCTGTACGGCGGGCTGGGCGACAGCCG CCCCGGCCTCTGGGGGGCGCCCGAGGCGGAGGAGGCGCGGGCCCGGTTCGAGGCCT CGGGCGCCCCGGCGCCCGTGTGGGCGCCCGAGCTGGGCGACGCGGCGCAGCAGTAC GCCCTGATCACGCGGCTGCTGTACACGCCGGACGCGGAGGCGATGGGGTGGCTCCA GAACCCGCGCGTGGCGCCCGGGGACGTGGCGCTGGACCAGGCCTGCTTCCGGATCT CGGGCGCGGCGCGCAACAGCAGCTCCTTCATCTCCGGCAGCGTGGCGCGGGCCGTG CCCCACCTGGGGTACGCCATGGCGGCGGGCCGCTTCGGCTGGGGCCTGGCGCACGT GGCGGCCGCCGTGGCCATGAGCCGCCGCTACGACCGCGCGCAGAAGGGCTTCCTGC TGACCAGCCTGCGCCGCGCCTACGCGCCCCTGCTGGCGCGCGAGAACGCGGCGCTG ACCGGGGCGCGAACCCCCGACGACGGCGGCGACGCCAACCGCCACGACGGCGACG ACGCCCGCGGGAAGCCCGCCGCCGCCGCCGCCCCGTTGCCGTCGGCGGCGGCGTCG CCGGCCGACGAGCGCGCGGTGCCCGCCGGCTACGGCGCCGCGGGGGTGCTCGCCGC CCTGGGGCGCCTGAGCGCCGCGCCCGCCTCCGCGCCGGCCGGGGCCGACGACGACG ACGACGACGACGGCGCCGGCGGTGGTGGCGGCGGCCGGCGCGCGGAGGCGGGCCG CGTGGCCGTGGAGTGCCTGGCCGCCTGCCGCGGGATCCTGGAGGCGCTGGCGGAGG GCTTCGACGGCGACCTGGCGGCCGTGCCGGGGCTGGCCGGAGCCCGGCCCGCCGCG CCCCCGCGCCCGGGGCCCGCGGGCGCGGCCGCCCCGCCGCACGCCGACGCGCCCCG CCTGCGCGCCTGGCTGCGCGAGCTGCGGTTCGTGCGCGACGCGCTGGTGCTGATGCG CCTGCGCGGGGACCTGCGCGTGGCCGGCGGCAGCGAGGCCGCCGTGGCCGCCGTGC GCGCCGTGAGCCTGGTCGCCGGGGCCCTGGGCCCGGCGCTGCCGCGGAGCCCGCGC CTGCTGAGCTCCGCCGCCGCCGCCGCCGCGGACCTGCTCTTCCAGAACCAGAGCCTG CGCCCCCTGCTGGCCGACACCGTCGCCGCGGCCGACTCGCTCGCCGCGCCCGCCTCC GCGCCGCGGGAGGCCGCGGACGCCCCCCGCCCCGCGGCCGCCCCTCCCGCGGGGGC CGCGCCCCCCGCCCCGCCGACGCCGCCGCCGCGGCCGCCGCGCCCCGCGGCGCTGA CCCGCCGGCCCGCCGAGGGCCCCGACCCGCAGGGCGGCTGGCGCCGCCAGCCGCCG GGGCCCAGCCACACGCCGGCGCCCTCGGCCGCCGCCCTGGAGGCCTACTGCGCCCC GCGGGCCGTGGCCGAGCTCACGGACCACCCGCTCTTCCCCGCGCCGTGGCGCCCGGC

Strain	Nucleic Acid Sequence
	CCTCATGTTCGACCCGCGCGCGCTGGCCTCGCTGGCCGCGCGCTGCGCCGCCCCGCC CCCCGGCGGCGCGCCCGCCGCCTTCGGCCCGCTGCGCGCCTCGGGCCCGCTGCGCCG CGCGGCGGCCTGGATGCGCCAGGTGCCCGACCCGGAGGACGTGCGCGTGGTGATCC TCTACTCGCCGCTGCCGGGCGAGGACCTGGCCGCGGGCCGCGCCGGGGGCGGGCCC CCCCCGGAGTGGTCCGCCGAGCGCGGCGGGCTGTCCTGCCTGCTGGCGGCCCTGGGC AACCGGCTCTGCGGGCCCGCCACGGCCGCCTGGGCGGGCAACTGGACCGGCGCCCC CGACGTCTCGGCGCTGGGCGCGCAGGGCGTGCTGCTGCTGTCCACGCGGGACCTGGC CTTCGCCGGCGCCGTGGAGTTCCTGGGGCTGCTGGCCGGCGCCTGCGACCGCCGCCT CATCGTCGTCAACGCCGTGCGCGCCGCGGCCTGGCCCGCCGCTGCCCCCGTGGTCTC GCGGCAGCACGCCTACCTGGCCTGCGAGGTGCTGCCCGCCGTGCAGTGCGCCGTGCG CTGGCCGGCGGCGCGGGACCTGCGCCGCACCGTGCTGGCCTCCGGCCGCGTGTTCGG GCCGGGGGTCTTCGCGCGCGTGGAGGCCGCGCACGCGCGCCTGTACCCCGACGCGC CGCCGCTGCGCCTCTGCCGCGGGGCCAACGTGCGGTACCGCGTGCGCACGCGCTTCG GCCCCGACACGCTGGTGCCCATGTCCCCGCGCGAGTACCGCCGCGCCGTGCTCCCGG CGCTGGACGGCCGGGCCGCCGCCTCGGGCGCGGGCGACGCCATGGCGCCCGGCGCG CCGGACTTCTGCGAGGACGAGGCGCACTCGCACCGCGCCTGCGCGCGCTGGGGCCT GGGCGCGCCGCTGCGGCCCGTCTACGTGGCGCTGGGGCGCGACGCCGTGCGCGGCG GCCCGGCGGAGCTGCGCGGGCCGCGGCGGGAGTTCTGCGCGCGGGCGCTGCTCGAG CCCGACGGCGACGCGCCCCCGCTGGTGCTGCGCGACGACGCGGACGCGGGCCCGCC CCCGCAGATACGCTGGGCGTCGGCCGCGGGCCGCGCGGGGACGGTGCTGGCCGCGG CGGGCGGCGGCGTGGAGGTGGTGGGGACCGCCGCGGGGCTGGCCACGCCGCCGAGG CGCGAGCCCGTGGACATGGACGCGGAGCTGGAGGACGACGACGACGGACTGTTTGG GGAGTGA (SEQ ID NO: 23)
MRK_HSV-2 gB, SQ-032178, CX-000747	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGAGAGGTGGTGGCTTAGTT TGCGCGCTGGTTGTCGGGGCGCTCGTAGCCGCCGTGGCGTCGGCCGCCCCTGCGGCT CCTCGCGCTAGCGGAGGCGTAGCCGCAACAGTTGCGGCGAACGGGGGTCCAGCCTC TCAGCCTCCTCCCGTCCCGAGCCCTGCGACCACCAAGGCTAGAAAGCGGAAGACCA AGAAACCGCCCAAGCGCCCCGAGGCCACCCCGCCCCCCGATGCCAACGCGACTGTC GCCGCTGGCCATGCGACGCTTCGCGCTCATCTGAGGGAGATCAAGGTTGAAAATGCT GATGCCCAATTTTACGTGTGCCCGCCCCCGACGGGCGCCACGGTTGTGCAGTTTGAA CAGCCGCGGCGCTGTCCGACGCGGCCAGAAGGCCAGAACTATACGGAGGGCATAGC GGTGGTCTTTAAGGAAAACATCGCCCCGTACAAATTTAAGGCCACAATGTACTACAA AGACGTGACAGTTTCGCAAGTGTGGTTTGGCCACAGATACTCGCAGTTTATGGGAAT CTTCGAAGATAGAGCCCCTGTTCCCTTCGAGGAAGTCATCGACAAGATTAATGCCAA AGGGGTATGCCGTTCCACGGCCAAATACGTGCGCAACAATATGGAGACCACCGCCT TTCACCGGGATGATCACGAGACCGACATGGAGCTTAAGCCGGCGAAGGTCGCCACG CGTACCTCCCGGGGTTGGCACACCACAGATCTTAAGTACAATCCCTCGCGAGTTGAA GCATTCCATCGGTATGGAACTACCGTTAACTGCATCGTTGAGGAGGTGGATGCGCGG TCGGTGTACCCTTACGATGAGTTTGTGTTAGCGACCGGCGATTTTGTGTACATGTCCC CGTTTTACGGCTACCGGGAGGGGTCGCACACCGAACATACCTCGTACGCCGCTGACA GGTTCAAGCAGGTCGATGGCTTTTACGCGCGCGATCTCACCACGAAGGCCCGGGCCA CGTCACCGACGACCAGGAACTTGCTCACGACCCCCAAGTTCACCGTCGCTTGGGATT GGGTCCCAAAGCGTCCGGCGGTCTGCACGATGACCAAATGGCAGGAGGTGGACGAA ATGCTCCGCGCAGAATACGGCGGCTCCTTCCGCTTCTCGTCCGACGCCATCTCGACA ACCTTCACCACCAATCTGACCCAGTACAGTCTGTCGCGCGTTGATTTAGGAGACTGC ATTGGCCGGGATGCCCGGGAGGCCATCGACAGAATGTTTGCGCGTAAGTACAATGC CACACATATTAAGGTGGGCCAGCCGCAATACTACCTTGCCACGGGCGGCTTTCTCAT CGCGTACCAGCCCCTTCTCTCAAATACGCTCGCTGAACTGTACGTGCGGGAGTATAT GAGGGAACAGGACCGCAAGCCCCGCAATGCCACGCCTGCGCCACTACGAGAGGCGC CTTCAGCTAATGCGTCGGTGGAACGTATCAAGACCACCTCCTCAATAGAGTTCGCCC GGCTGCAATTTACGTACAACCACATCCAGCGCCACGTGAACGACATGCTGGGCCGC ATCGCTGTCGCCTGGTGCGAGCTGCAGAATCACGAGCTGACTCTTTGGAACGAGGCC CGAAAACTCAACCCCAACGCGATCGCCTCCGCAACAGTCGGTAGACGGGTGAGCGC TCGCATGCTAGGAGATGTCATGGCTGTGTCCACCTGCGTGCCCGTCGCTCCGGACAA CGTGATTGTGCAGAATTCGATGCGGGTCTCATCGCGGCCGGGCACCTGCTACAGCAG GCCCCTCGTCAGCTTCCGGTACGAAGACCAGGGCCCGCTGATTGAAGGGCAACTGG GAGAGAACAATGAGCTGCGCCTCACCCGCGACGCGCTCGAACCCTGCACCGTCGGA CATCGGAGATATTTCATCTTCGGAGGGGGCTACGTGTACTTCGAAGAGTATGCCTAC

Strain	Nucleic Acid Sequence
	TCTCACCAGCTGAGTAGAGCCGACGTCACTACCGTCAGCACCTTTATTGACCTGAAT ATCACCATGCTGGAGGACCACGAGTTTGTGCCCCTGGAAGTTTACACTCGCCACGAA ATCAAAGACTCCGGCCTGTTGGATTACACGGAGGTTCAGAGGCGGAACCAGCTGCA TGACCTGCGCTTTGCCGACATCGACACCGTCATCCGCGCCGATGCCAACGCTGCCAT GTTCGCGGGGCTGTGCGCGTTCTTCGAGGGGATGGGTGACTTGGGGCGCGCCGTCGG CAAGGTCGTCATGGGAGTAGTGGGGGGCGTTGTGAGTGCCGTCAGCGGCGTGTCCTC CTTCATGTCCAATCCATTCGGAGCGCTTGCTGTGGGGCTGCTGGTCCTGGCCGGGCT GGTAGCCGCCTTCTTCGCCTTTCGATATGTTCTGCAACTGCAACGCAATCCCATGAA AGCTCTATATCCGCTCACCACCAAGGAGCTAAAGACGTCAGATCCAGGAGGCGTGG GCGGGGAAGGGGAAGAGGGCGCGGAGGGCGGAGGGTTTGACGAAGCCAAATTGGC CGAGGCTCGTGAAATGATCCGATATATGGCACTAGTGTCGGCGATGGAAAGGACCG AACATAAGGCCCGAAAGAAGGGCACGTCGGCGCTGCTCTCATCCAAGGTCACCAAC ATGGTACTGCGCAAGCGCAACAAAGCCAGGTACTCTCCGCTCCATAACGAGGACGA GGCGGGAGATGAGGATGAGCTCTAATGATAATAGGCTGGAGCCTCGGTGGCCATGC TTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCCG TGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 54)
MRK_HSV-2 gC, SQ-032179, CX-000670	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGCCCTTGGACGGGTAGG CCTAGCCGTGGGCCTGTGGGGCCTACTGTGGGTGGGTGTGGTCGTGGTGCTGGCCAA TGCCTCCCCCGGACGCACGATAACGGTGGGCCCGCGAGGCAACGCGAGCAATGCTG CCCCCTCCGCGTCCCCGCGGAACGCATCCGCCCCCCGAACCACACCCACGCCCCCAC AACCCCGCAAAGCGACGAAATCCAAGGCCTCCACCGCCAAACCGGCTCCGCCCCCC AAGACCGGACCCCCGAAGACATCCTCGGAGCCCGTGCGATGCAACCGCCACGACCC GCTGGCCCGGTACGGCTCGCGGGTGCAAATCCGATGCCGGTTTCCCAACTCCACGAG GACTGAGTCCCGTCTCCAGATCTGGCGTTATGCCACGGCGACGGACGCCGAAATCGG AACAGCGCCTAGCTTAGAAGAGGTGATGGTGAACGTGTCGGCCCCGCCCGGGGGCC AACTGGTGTATGACAGTGCCCCCAACCGAACGGACCCGCATGTAATCTGGGCGGAG GGCGCCGGCCCGGGCGCCAGCCCGCGCCTGTACTCGGTTGTCGGCCCGCTGGGTCGG CAGCGGCTCATCATCGAAGAGTTAACCCTGGAGACACAGGGCATGTACTATTGGGT GTGGGGCCGGACGGACCGCCCGTCCGCCTACGGGACCTGGGTCCGCGTTCGAGTATT TCGCCCTCCGTCGCTGACCATCCACCCCCACGCGGTGCTGGAGGGCCAGCCGTTTAA GGCGACGTGCACGGCCGCAACCTACTACCCGGGCAACCGCGCGGAGTTCGTCTGGTT TGAGGACGGTCGCCGCGTATTCGATCCGGCACAGATACACACGCAGACGCAGGAGA ACCCCGACGGCTTTTCCACCGTCTCCACCGTGACCTCCGCGGCCGTCGGCGGGCAGG GCCCCCCTCGCACCTTCACCTGCCAGCTGACGTGGCACCGCGACTCCGTGTCGTTCT CTCGGCGCAACGCCAGCGGCACGGCCTCGGTTCTGCCGCGGCCGACCATTACCATGG AGTTTACAGGCGACCATGCGGTCTGCACGGCCGGCTGTGTGCCCGAGGGGGTCACGT TTGCTTGGTTCCTGGGGGATGACTCCTCGCCGGCGGAAAAGGTGGCCGTCGCGTCCC AGACATCGTGCGGGCGCCCCGGCACCGCCACGATCCGCTCCACCCTGCCGGTCTCGT ACGAGCAGACCGAGTACATCTGTAGACTGGCGGGATACCCGGACGGAATTCCGGTC CTAGAGCACCACGGAAGCCACCAGCCCCCGCCGCGGGACCCAACCGAGCGGCAGGT GATCCGGGCGGTGGAGGGGGCGGGGATCGGAGTGGCTGTCCTTGTCGCGGTGGTTC TGGCCGGGACCGCGGTAGTGTACCTGACCCATGCCTCCTCGGTACGCTATCGTCGGC TGCGGTAATGATAATAGGCTGGAGCCTCGGTGGCCATGCTTCTTGCCCCTTGGGCCT CCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCCGTGGTCTTTGAATAAAGTC TGAGTGGGCGGC (SEQ ID NO: 55)
MRK_HSV-2 gD, SQ-032180, CX-001301	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGGGCGTTTGACCTCCGGC GTCGGGACGGCGGCCCTGCTAGTTGTCGCGGTGGGACTCCGCGTCGTCTGCGCCAAA TACGCCTTAGCAGACCCCTCGCTTAAGATGGCCGATCCCAATCGATTTCGCGGGAAG AACCTTCCGGTTTTGGACCAGCTGACCGACCCCCCCGGGGTGAAGCGTGTTTACCAC ATTCAGCCGAGCCTGGAGGACCCGTTCCAGCCCCCCAGCATCCCGATCACTGTGTAC TACGCAGTGCTGGAACGTGCCTGCCGCAGCGTGCTCCTACATGCCCCATCGGAGGCC CCCCAGATCGTGCGCGGGGCTTCGGACGAGGCCCGAAAGCACACGTACAACCTGAC CATCGCCTGGTATCGCATGGGAGACAATTGCGCTATCCCCATCACGGTTATGGAATA CACCGAGTGCCCCTACAACAAGTCGTTGGGGGTCTGCCCCATCCGAACGCAGCCCCG CTGGAGCTACTATGACAGCTTTAGCGCCGTCAGCGAGGATAACCTGGGATTCCTGAT GCACGCCCCCGCCTTCGAGACCGCGGGTACGTACCTGCGGCTAGTGAAGATAAACG ACTGGACGGAGATCACACAATTTATCCTGGAGCACCGGGCCCGCGCCTCCTGCAAGT

Strain	Nucleic Acid Sequence
	ACGCTCTCCCCCTGCGCATCCCCCCGGCAGCGTGCCTCACCTCGAAGGCCTACCAAC AGGGCGTGACGGTCGACAGCATCGGGATGCTACCCCGCTTTATCCCCGAAAACCAG CGCACCGTCGCCCTATACAGCTTAAAAATCGCCGGGTGGCACGGCCCCAAGCCCCC GTACACCAGCACCCTGCTGCCGCCGGAGCTGTCCGACACCACCAACGCCACGCAAC CCGAACTCGTTCCGGAAGACCCCGAGGACTCGGCCCTCTTAGAGGATCCCGCCGGG ACGGTGTCTTCGCAGATCCCCCCAAACTGGCACATCCCGTCGATCCAGGACGTCGCA CCGCACCACGCCCCCGCCGCCCCCAGCAACCCGGGCCTGATCATCGGCGCGCTGGCC GGCAGTACCCTGGCGGTGCTGGTCATCGGCGGTATTGCGTTTTGGGTACGCCGCCGC GCTCAGATGGCCCCCAAGCGCCTACGTCTCCCCCACATCCGGGATGACGACGCGCCC CCCTCGCACCAGCCATTGTTTTACTAGTGATAATAGGCTGGAGCCTCGGTGGCCATG CTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCC GTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 56)
MRK_HSV-2 gE, SQ-032181, CX-001391	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGCTAGGGGGGCCGGGTT GGTTTTTTTTGTTGGAGTTTGGGTCGTAAGCTGCCTCGCGGCAGCGCCCAGAACGTC CTGGAAACGCGTAACCTCGGGCGAAGACGTGGTGTTACTCCCCGCGCCGGCGGGGC CGGAAGAACGCACTCGGGCCCACAAACTACTGTGGGCAGCGGAACCGCTGGATGCC TGCGGTCCCCTGAGGCCGTCATGGGTGGCACTGTGGCCCCCCCGACGAGTGCTTGAG ACGGTTGTCGATGCGGCGTGCATGCGCGCCCCGGAACCGCTCGCTATCGCATACAGT CCCCCGTTCCCTGCGGGCGACGAGGGACTTTATTCGGAGTTGGCGTGGCGCGATCGC GTAGCCGTGGTCAACGAGAGTTTAGTTATCTACGGGGCCCTGGAGACGGACAGTGG TCTGTACACCCTGTCAGTGGTGGGCCTATCCGACGAGGCCCGCCAAGTGGCGTCCGT GGTTCTCGTCGTCGAGCCCGCCCCTGTGCCTACCCCGACCCCCGATGACTACGACGA GGAGGATGACGCGGGCGTGAGCGAACGCACGCCCGTCAGCGTTCCCCCCCCAACAC CCCCCCGACGTCCCCCCGTCGCCCCCCCGACGCACCCTCGTGTTATCCCTGAGGTGA GCCACGTGCGGGGGGTGACGGTCCACATGGAAACCCCGGAGGCCATTCTGTTTGCG CCAGGGGAGACGTTTGGGACGAACGTCTCCATCCACGCAATTGCCCACGACGACGG TCCGTACGCCATGGACGTCGTCTGGATGCGATTTGATGTCCCGTCCTCGTGCGCCGA GATGCGGATCTATGAAGCATGTCTGTATCACCCGCAGCTGCCTGAGTGTCTGTCTCC GGCCGATGCGCCGTGCGCCGTAAGTTCGTGGGCGTACCGCCTGGCGGTCCGCAGCTA CGCCGGCTGCTCCAGGACTACGCCCCCACCTCGATGTTTTGCTGAAGCTCGCATGGA ACCGGTCCCCGGGTTGGCGTGGCTCGCATCAACTGTTAATCTGGAATTCCAGCATGC CTCTCCCCAACACGCCGGCCTCTATCTGTGTGTGGTGTATGTGGACGACCATATCCAT GCCTGGGGCCACATGACCATCTCCACAGCGGCCCAGTACCGGAATGCGGTGGTGGA ACAGCATCTCCCCCAGCGCCAGCCCGAGCCCGTAGAACCCACCCGACCGCATGTGA GAGCCCCCCCTCCCGCACCCTCCGCGAGAGGCCCGTTACGCTTAGGTGCGGTCCTGG GGGCGGCCCTGTTGCTCGCGGCCCTCGGGCTATCCGCCTGGGCGTGCATGACCTGCT GGCGCAGGCGCAGTTGGCGGGCGGTTAAAAGTCGGGCCTCGGCGACCGGCCCCACT TACATTCGAGTAGCGGATAGCGAGCTGTACGCGGACTGGAGTTCGGACTCAGAGGG CGAGCGCGACGGTTCCCTGTGGCAGGACCCTCCGGAGAGACCCGACTCACCGTCCA CAAATGGATCCGGCTTTGAGATCTTATCCCCAACGGCGCCCTCTGTATACCCCCATA GCGAAGGGCGTAAATCGCGCCGCCCGCTCACCACCTTTGGTTCAGGAAGCCCGGGA CGTCGTCACTCCCAGGCGTCCTATTCTTCCGTCTTATGGTAATGATAATAGGCTGGAG CCTCGGTGGCCATGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCT GCACCCGTACCCCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 57)
MRK_HSV-2 gI, SQ-032182, CX-000645	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGCCCGGCCGCTCGCTGCAG GGCCTGGCGATCCTGGGCCTGTGGGTCTGCGCCACCGGCCTGGTCGTCCGCGGCCCC ACGGTCAGTCTGGTCTCAGACTCACTCGTGGATGCCGGGGCCGTGGGGCCCCAGGGC TTCGTGGAAGAGGACCTGCGTGTTTTCGGGGAGCTTCATTTTGTGGGGGCCCAGGTC CCCCACACAAACTACTACGACGGCATCATCGAGCTGTTTCACTACCCCCTGGGGAAC CACTGCCCCCCGCGTTGTACACGTGGTCACACTGACCGCATGCCCCCGCCGCCCCGCC GTGGCGTTCACCTTGTGTCGCTCGACGCACCACGCCCACAGCCCCGCCTATCCGACC CTGGAGCTGGGTCTGGCGCGGCAGCCGCTTCTGCGGGTTCGAACGGCAACGCGCGA CTATGCCGGTCTGTATGTCCTGCGCGTATGGGTCGGCAGCGCGACGAACGCCAGCCT GTTTGTTTTGGGGGTGGCGCTCTCTGCCAACGGGACGTTTGTGTATAACGGCTCGGA CTACGGCTCCTGCGATCCGGCGCAGCTTCCCTTTTCGGCCCCGCGCCTGGGACCCTC GAGCGTATACACCCCCGGAGCCTCCCGGCCCACCCCTCCACGGACAACGACATCAC CGTCCTCCCCACGAGACCCGACCCCCGCCCCCGGGGACACAGGGACGCCTGCTCCC

Strain	Nucleic Acid Sequence
	GCGAGCGGCGAGAGAGCCCCGCCCAATTCCACGCGATCGGCCAGCGAATCGAGACA CAGGCTAACCGTAGCCCAGGTAATCCAGATCGCCATACCGGCGTCCATCATCGCCTT TGTGTTTCTGGGCAGCTGTATCTGCTTCATCCATAGATGCCAGCGCCGATACAGGCG CCCCCGCGGCCAGATTTACAACCCCGGGGGCGTTTCCTGCGCGGTCAACGAGGCGGC CATGGCCCGCCTCGGAGCCGAGCTGCGATCCCACCCAAACACCCCCCCCAAACCCC GACGCCGTTCGTCGTCGTCCACGACCATGCCTTCCCTAACGTCGATAGCTGAGGAAT CGGAGCCAGGTCCAGTCGTGCTGCTGTCCGTCAGTCCTCGGCCCCGCAGTGGCCCGA CGGCCCCCCAAGAGGTCTAGTGATAATAGGCTGGAGCCTCGGTGGCCATGCTTCTTG CCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCCGTGGTCT TTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 58)
MRK_HSV-2 SgB, SQ-032210, CX-000655	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGCGCGGGGGGGGCTTAGT TTGCGCGCTGGTCGTGGGGGCGCTCGTAGCCGCGGTCGCGTCGGCGGCTCCGGCTGC CCCACGCGCTTCAGGTGGTGTCGCTGCGACCGTTGCGGCGAATGGTGGTCCCGCCAG CCAACCGCCTCCCGTCCCGAGCCCCGCGACCACTAAGGCCCGGAAGCGGAAGACCA AGAAGCCACCCAAGCGGCCCGAGGCGACTCCGCCCCCAGACGCCAACGCGACCGTC GCCGCCGGCCACGCCACTCTGCGTGCGCACCTGCGGGAAATCAAGGTCGAGAACGC GGACGCCCAGTTTTACGTGTGCCCGCCGCCGACTGGCGCCACGGTGGTGCAGTTTGA GCAACCTAGGCGCTGCCCGACGCGACCAGAGGGGCAGAACTACACCGAGGGCATAG CGGTGGTCTTTAAGGAAAACATCGCCCCGTACAAATTCAAGGCCACCATGTACTACA AAGACGTGACCGTGTCGCAGGTGTGGTTCGGCCACCGCTACTCCCAGTTTATGGGGA TATTCGAGGACCGCGCCCCCGTTCCCTTCGAAGAGGTGATTGACAAAATTAACGCCA AGGGGGTCTGCCGCAGTACGGCGAAGTACGTCCGGAACAACATGGAGACCACTGCC TTCCACCGGGACGACCACGAAACAGACATGGAGCTCAAACCGGCGAAAGTCGCCAC GCGCACGAGCCGGGGGTGGCACACCACCGACCTCAAATACAATCCTTCGCGGGTGG AAGCATTCCATCGGTATGGCACGACCGTCAACTGTATCGTAGAGGAGGTGGATGCG CGGTCGGTGTACCCCTACGATGAGTTCGTGCTGGCAACGGGCGATTTTGTGTACATG TCCCCTTTTTACGGCTACCGGGAAGGTAGTCACACCGAGCACACCAGTTACGCCGCC GACCGCTTTAAGCAAGTGGACGGCTTCTACGCGCGCGACCTCACCACAAAGGCCCG GGCCACGTCGCCGACGACCCGCAATTTGCTGACGACCCCCAAGTTTACCGTGGCCTG GGACTGGGTGCCTAAGCGACCGGCGGTCTGTACCATGACAAAGTGGCAGGAGGTGG ACGAAATGCTCCGCGCTGAATACGGTGGCTCTTTCCGCTTCTCTTCCGACGCCATCTC CACCACGTTCACCACCAACCTGACCCAATACTCGCTCTCGAGAGTCGATCTGGGAGA CTGCATTGGCCGGGATGCCCGCGAGGCAATTGACCGCATGTTCGCGCGCAAGTACA ACGCTACGCACATAAAGGTTGGCCAACCCCAGTACTACCTAGCCACGGGGGGCTTCC TCATCGCTTATCAACCCCTCCTCAGCAACACGCTCGCCGAGCTGTACGTGCGGGAAT ATATGCGGGAACAGGACCGCAAACCCCGAAACGCCACGCCCGCGCCGCTGCGGGAA GCACCGAGCGCCAACGCGTCCGTGGAGCGCATCAAGACGACATCCTCGATTGAGTTT GCTCGTCTGCAGTTTACGTATAACCACATACAGCGCCATGTAAACGACATGCTCGGG CGCATCGCCGTCGCGTGGTGCGAGCTCCAAAATCACGAGCTCACTCTGTGGAACGAG GCACGCAAGCTCAATCCCAACGCCATCGCATCCGCCACCGTAGGCCGGCGGGTGAG CGCTCGCATGCTCGGGGATGTCATGGCCGTCTCCACGTGCGTGCCCGTCGCCCCGGA CAACGTGATCGTGCAAAATAGCATGCGCGTTTCTTCGCGGCCGGGGACGTGCTACAG CCGCCCGCTGGTTAGCTTTCGGTACGAAGACCAAGGCCCGCTGATTGAGGGGCAGCT GGGTGAGAACAACGAGCTGCGCCTCACCCGCGATGCGTTAGAGCCGTGTACCGTCG GCCACCGGCGCTACTTCATCTTCGGAGGGGGATACGTATACTTCGAAGAATATGCGT ACTCTCACCAATTGAGTCGCGCCGATGTCACCACTGTTAGCACCTTCATCGACCTGA ACATCACCATGCTGGAGGACCACGAGTTCGTGCCCCTGGAGGTCTACACACGCCACG AGATCAAGGATTCCGGCCTACTGGACTACACCGAAGTCCAGAGACGAAATCAGCTG CACGATCTCCGCTTTGCTGACATCGATACTGTTATCCGCGCCGACGCCAACGCCGCC ATGTTCGCAGGTCTGTGTGCGTTTTTCGAGGGTATGGGTGACTTAGGGCGCGCGGTG GGCAAGGTCGTCATGGGGGTAGTCGGGGGCGTGGTGTCGGCCGTCTCGGGCGTCTCC TCCTTTATGTCTAACCCCTGATAATAGGCTGGAGCCTCGGTGGCCATGCTTCTTGCCC CTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCCGTGGTCTTTG AATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 59)
MRK_HSV-2 SgC, SQ-032835, CX-000616	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGCACTGGGAAGAGTGGG ATTGGCCGTCGGACTGTGGGGACTGCTGTGGGTGGGAGTCGTCGTCGTCCTGGCTAA CGCCTCACCCGGTCGGACTATCACTGTGGGACCCAGGGGGAACGCCTCTAACGCCGC

Strain	Nucleic Acid Sequence
	GCCCTCAGCTAGCCCCAGGAATGCCAGCGCTCCCAGGACCACCCCGACTCCTCCGCA ACCCCGCAAGGCGACCAAGTCCAAGGCGTCCACTGCCAAGCCAGCGCCTCCGCCTA AGACTGGCCCCCCTAAGACCTCCAGCGAACCTGTGCGGTGCAACCGGCACGACCCT CTGGCACGCTACGGATCGCGGGTCCAAATCCGGTGTCGGTTCCCGAACAGCACTCGG ACCGAATCGCGGCTCCAGATTTGGAGATACGCAACTGCCACTGATGCCGAGATCGG CACTGCCCCAAGCCTTGAGGAGGTCATGGTCAACGTGTCAGCTCCTCCTGGAGGCCA GCTGGTGTACGACTCCGCTCCGAACCGAACCGACCCGCACGTCATCTGGGCCGAAG GAGCCGGTCCTGGTGCATCGCCGAGGTTGTACTCGGTAGTGGGTCCCCTGGGGAGAC AGCGGCTGATCATCGAAGAACTGACTCTGGAGACTCAGGGCATGTACTATTGGGTGT GGGGCAGAACCGATAGACCATCCGCATACGGAACCTGGGTGCGCGTGAGAGTGTTC AGACCCCCGTCCTTGACAATCCACCCGCATGCGGTGCTCGAAGGGCAGCCCTTCAAG GCCACTTGCACTGCGGCCACTTACTACCCTGGAAACCGGGCCGAATTCGTGTGGTTC GAGGATGGACGGAGGGTGTTCGACCCGGCGCAGATTCATACGCAGACTCAGGAAAA CCCGGACGGCTTCTCCACCGTGTCCACTGTGACTTCGGCCGCTGTGGGAGGACAAGG ACCGCCACGCACCTTCACCTGTCAGCTGACCTGGCACCGCGACAGCGTGTCCTTTAG CCGGCGGAACGCATCAGGCACTGCCTCCGTGTTGCCTCGCCCAACCATTACCATGGA GTTCACCGGAGATCACGCCGTGTGCACTGCTGGCTGCGTCCCCGAAGGCGTGACCTT CGCCTGGTTTCTCGGGGACGACTCATCCCCGGCGGAAAAGGTGGCCGTGGCCTCTCA GACCAGCTGCGGTAGACCGGGAACCGCCACCATCCGCTCCACTCTGCCGGTGTCGTA CGAGCAGACCGAGTACATTTGTCGCCTGGCCGGATACCCGGACGGTATCCCAGTGCT CGAACACCACGGCAGCCATCAGCCTCCGCCGAGAGATCCTACCGAGCGCCAGGTCA TCCGGGCCGTGGAAGGATGATAATAGGCTGGAGCCTCGGTGGCCATGCTTCTTGCCC CTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCCCGTGGTCTTTG AATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 60)
MRK_HSV-2 SgE, SQ-032211, CX-003794	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGCTCGCGGGGCCGGGTT GGTGTTTTTTGTTGGAGTTTGGGTCGTATCGTGCCTGGCGGCAGCACCCAGAACGTC CTGGAAACGGGTTACCTCGGGCGAGGACGTGGTGTTGCTTCCGGCGCCCGCGGGGC CGGAGGAACGCACACGGGCCCACAAACTACTGTGGGCCGCGGAACCCCTGGATGCC TGCGGTCCCCTGAGGCCGTCGTGGGTGGCGCTGTGGCCCCCGCGACGGGTGCTCGAA ACGGTCGTGGATGCGGCGTGCATGCGCGCCCCGGAACCGCTCGCCATAGCATACAG TCCCCCGTTCCCCGCGGGCGACGAGGGACTGTATTCGGAGTTGGCGTGGCGCGATCG CGTAGCCGTGGTCAACGAGAGTCTGGTCATCTACGGGGCCCTGGAGACGGACAGCG GTCTGTACACCCTGTCCGTGGTCGGCCTAAGCGACGAGGCGCGCCAAGTGGCGTCGG TGGTTCTGGTCGTGGAGCCCGCCCCTGTGCCGACCCCGACCCCCGACGACTACGACG AAGAAGACGACGCGGGCGTGAGCGAACGCACGCCGGTCAGCGTACCCCCCCCGACC CCACCCCGTCGTCCCCCCGTCGCCCCCCCTACGCACCCTCGTGTTATCCCCGAGGTGT CCCACGTGCGCGGGGTAACGGTCCATATGGAGACCCCGGAGGCCATTCTGTTTGCCC CCGGAGAGACGTTTGGGACGAACGTCTCCATCCACGCCATTGCCCATGACGACGGTC CGTACGCCATGGACGTCGTCTGGATGCGGTTTGACGTGCCGTCCTCGTGCGCCGAGA TGCGGATCTACGAAGCTTGTCTGTATCACCCGCAGCTTCCAGAATGTCTATCTCCGG CCGACGCGCCGTGCGCTGTAAGTTCCTGGGCGTACCGCCTGGCGGTCCGCAGCTACG CCGGCTGTTCCAGGACTACGCCCCCGCCGCGATGTTTTGCCGAGGCTCGCATGGAAC CGGTCCCGGGGTTGGCGTGGTTAGCCTCCACCGTCAACCTGGAATTCCAGCACGCCT CCCCTCAGCACGCCGGCCTTTACCTGTGCGTGGTGTACGTGGACGATCATATCCACG CCTGGGGCCACATGACCATCTCTACCGCGGCGCAGTACCGGAACGCGGTGGTGGAA CAGCACTTGCCCCAGCGCCAGCCTGAACCCGTCGAGCCCACCCGCCCGCACGTAAG AGCACCCCCTCCCGCGCCTTCCGCGCGCGGCCCGCTGCGCTGATAATAGGCTGGAGC CTCGGTGGCCATGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTG CACCCGTACCCCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 61)
MRK_HSV-2 SgI, SQ-032323, CX-002683	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGCCCGGCCGCTCGCTGCAG GGCCTGGCGATCCTGGGCCTGTGGGTCTGCGCCACCGGCCTGGTCGTCCGCGGCCCC ACGGTCAGTCTGGTCTCAGACTCACTCGTGGATGCCGGGGCCGTGGGGCCCCAGGGC TTCGTGGAAGAGGACCTGCGTGTTTTCGGGGAGCTTCATTTTGTGGGGGCCCAGGTC CCCCACACAAACTACTACGACGGCATCATCGAGCTGTTTCACTACCCCCTGGGGAAC CACTGCCCCCGCGTTGTACACGTGGTCACACTGACCGCATGCCCCCGCCGCCCCGCC GTGGCGTTCACCTTGTGTCGCTCGACGCACCACGCCCACAGCCCCGCCTATCCGACC CTGGAGCTGGGTCTGGCGCGGCAGCCGCTTCTGCGGGTTCGAACGGCAACGCGCGA

Strain	Nucleic Acid Sequence
	CTATGCCGGTCTGTATGTCCTGCGCGTATGGGTCGGCAGCGCGACGAACGCCAGCCT GTTTGTTTTGGGGGTGGCGCTCTCTGCCAACGGGACGTTTGTGTATAACGGCTCGGA CTACGGCTCCTGCGATCCGGCGCAGCTTCCCTTTTCGGCCCCGCGCCTGGGACCCTC GAGCGTATACACCCCCGGAGCCTCCCGGCCCACCCCTCCACGGACAACGACATCCCC GTCCTCCCCTAGAGACCCGACCCCCGCCCCCGGGGACACAGGAACGCCTGCGCCCG CGAGCGGCGAGAGAGCCCCGCCCAATTCCACGCGATCGGCCAGCGAATCGAGACAC AGGCTAACCGTAGCCCAGGTAATCCAGTGATAATAGGCTGGAGCCTCGGTGGCCAT GCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACCCC CGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 62)
MRK_HSV-2 SgD, SQ-032172, CX-004714	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGGGCGTTTGACCTCCGGC GTCGGGACGGCGGCCCTGCTAGTTGTCGCGGTGGGACTCCGCGTCGTCTGCGCCAAA TACGCCTTAGCAGACCCCTCGCTTAAGATGGCCGATCCCAATCGATTTCGCGGGAAG AACCTTCCGGTTTTGGACCAGCTGACCGACCCCCCCGGGGTGAAGCGTGTTTACCAC ATTCAGCCGAGCCTGGAGGACCCGTTCCAGCCCCCCAGCATCCCGATCACTGTGTAC TACGCAGTGCTGGAACGTGCCTGCCGCAGCGTGCTCCTACATGCCCCATCGGAGGCC CCCCAGATCGTGCGCGGGGCTTCGGACGAGGCCCGAAAGCACACGTACAACCTGAC CATCGCCTGGTATCGCATGGGAGACAATTGCGCTATCCCCATCACGGTTATGGAATA CACCGAGTGCCCCTACAACAAGTCGTTGGGGGTCTGCCCCATCCGAACGCAGCCCCG CTGGAGCTACTATGACAGCTTTAGCGCCGTCAGCGAGGATAACCTGGGATTCCTGAT GCACGCCCCCGCCTTCGAGACCGCGGGTACGTACCTGCGGCTAGTGAAGATAAACG ACTGGACGGAGATCACACAATTTATCCTGGAGCACCGGGCCCGCGCCTCCTGCAAGT ACGCTCTCCCCCTGCGCATCCCCCCGGCAGCGTGCCTCACCTCGAAGGCCTACCAAC AGGGCGTGACGGTCGACAGCATCGGGATGCTACCCCGCTTTATCCCCGAAAACCAG CGCACCGTCGCCCTATACAGCTTAAAAATCGCCGGGTGGCACGGCCCCAAGCCCCC GTACACCAGCACCCTGCTGCCGCCGGAGCTGTCCGACACCACCAACGCCACGCAAC CCGAACTCGTTCCGGAAGACCCCGAGGACTCGGCCCTCTTAGAGGATCCCGCCGGG ACGGTGTCTTCGCAGATCCCCCCAAACTGGCACATCCCGTCGATCCAGGACGTCGCG CCGCACCACGCCCCCGCCGCCCCCAGCAACCCGTGATAATAGGCTGGAGCCTCGGT GGCCATGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCG TACCCCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 63)
MRK_HSV-2 ICP-0, SQ-032521, CX-004422	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGAACCGCGGCCTGGTAC TTCATCCCGCGCCGATCCTGGACCGGAACGGCCACCTCGCCAGACCCCTGGAACGCA GCCTGCAGCCCCTCACGCCTGGGGGATGCTGAATGATATGCAGTGGCTGGCCTCAAG CGACTCCGAGGAAGAGACAGAGGTCGGCATCTCCGACGATGATCTCCATCGGGATT CTACTTCGGAAGCGGGCTCCACCGACACAGAGATGTTCGAGGCCGGCCTGATGGAT GCTGCGACCCCTCCCGCAAGACCGCCTGCCGAACGCCAAGGCTCGCCGACCCCTGCT GACGCCCAGGGTTCGTGCGGTGGAGGCCCTGTGGGGGAGGAGGAAGCTGAAGCCGG AGGCGGTGGAGATGTCAACACCCCGGTGGCCTACCTGATCGTGGGCGTGACTGCCA GCGGATCCTTCTCGACCATCCCCATTGTCAACGATCCCCGCACTCGGGTCGAAGCGG AGGCCGCAGTGCGGGCTGGAACTGCCGTGGACTTCATTTGGACTGGCAATCCCAGG ACCGCTCCCCGGTCACTGTCCCTGGGAGGACACACCGTCCGCGCCCTGTCACCAACT CCCCCGTGGCCTGGAACCGATGACGAGGACGACGACCTGGCCGATGTGGACTACGT GCCCCCTGCCCCAAGACGGGCTCCACGGAGAGGAGGCGGAGGCGCCGGTGCCACCA GGGGCACCAGCCAACCCGCTGCCACCCGGCCTGCTCCTCCTGGGGCCCCGAGATCCT CCTCATCCGGCGGGGCACCTCTGAGAGCAGGAGTGGGCTCAGGCTCCGGAGGAGGA CCCGCCGTGGCAGCTGTGGTCCCGCGAGTGGCCTCCTTGCCTCCGGCCGCAGGAGGC GGCCGGGCCCAGGCCAGAAGGGTGGGGGAGGACGCGGCAGCCGCCGAAGGGCGCA CTCCTCCAGCGCGCCAACCAAGAGCAGCGCAAGAGCCTCCGATCGTGATCTCCGATA GCCCCCCACCGTCACCTCGCAGACCAGCCGGACCCGGGCCTCTGTCGTTCGTGAGCT CCAGCTCGGCCCAGGTGTCGAGCGGACCTGGCGGTGGTGGACTCCCTCAGAGCAGC GGCAGAGCTGCCAGACCTCGCGCCGCCGTGGCCCCGAGGGTCAGGTCGCCGCCGAG AGCAGCTGCCGCCCCAGTGGTGTCCGCCTCAGCCGACGCCGCCGGTCCCGCGCCTCC TGCTGTGCCAGTGGACGCCCATAGAGCGCCGCGGAGCAGAATGACTCAGGCACAGA CTGACACCCAGGCCCAGTCGCTCGGTAGGGCTGGAGCCACCGACGCCAGAGGATCG GGCGGACCCGGAGCCGAAGGAGGGTCCGGTCCCGCCGCTTCCTCCTCCGCGTCCTCA TCAGCCGCTCCGCGCTCACCGCTCGCACCCCAGGGTGTCGGAGCAAAGCGAGCAGC TCCTCGCCGGGCCCCTGACTCCGACTCAGGAGATCGGGGCCACGGACCACTCGCGCC

Strain	Nucleic Acid Sequence
	TGCCAGCGCTGGAGCGGCTCCTCCATCGGCTTCCCCATCCTCGCAAGCAGCCGTGGC CGCCGCATCCTCAAGCTCGGCGTCCTCTAGCTCAGCGAGCTCCTCCAGCGCCTCGTC CTCGTCCGCCTCCAGCAGCTCAGCCTCCTCGTCCTCGGCCTCCTCATCGTCCGCCTCC TCCTCCGCTGGAGGTGCCGGAGGATCGGTCGCATCCGCTTCCGGCGCAGGGGAGCG CCGAGAAACGTCCCTGGGTCCGCGGGCAGCTGCTCCGAGGGGTCCTCGCAAGTGCG CGCGGAAAACTCGGCACGCGGAGGGAGGACCGGAACCTGGCGCGAGAGATCCTGC GCCTGGACTGACCCGGTACCTCCCCATTGCCGGGGTGTCCAGCGTGGTGGCACTTGC CCCGTACGTCAACAAGACCGTGACCGGGGACTGTCTCCCCGTGCTCGACATGGAGAC TGGACACATTGGCGCGTATGTGGTCCTGGTGGATCAGACCGGTAATGTGGCCGACCT TTTGAGAGCAGCGGCCCCAGCATGGTCCCGCAGAACCCTGCTGCCTGAGCACGCCA GGAATTGCGTGCGGCCGCCGGACTACCCGACTCCGCCCGCCAGCGAATGGAACTCA CTGTGGATGACTCCCGTGGGCAACATGCTGTTCGATCAGGGGACCCTGGTCGGAGCC CTGGATTTTCACGGCCTGCGCTCCAGACATCCGTGGTCTAGGGAACAGGGTGCTCCT GCTCCCGCGGGTGATGCCCCTGCTGGCCACGGCGAATAGTGATAATAGGCTGGAGC CTCGGTGGCCATGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTG CACCCGTACCCCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 64)
MRK_HSV-2 ICP-4, SQ-032440, CX-002146	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAAATAAGAG AGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGTCGGCCGAGCAGCGCAA GAAGAAGAAAACGACCACCACTACCCAGGGCAGAGGAGCCGAAGTCGCCATGGCC GATGAAGATGGCGGGAGGCTGCGGGCCGCCGCTGAAACCACCGGAGGACCGGGATC CCCTGACCCTGCGGACGGCCCACCTCCCACACCGAACCCGGACAGACGGCCTGCTG CAAGGCCCGGTTTCGGATGGCACGGGGGACCCGAAGAGAACGAGGACGAAGCCGA TGACGCCGCGGCGGATGCAGACGCCGACGAGGCGGCTCCCGCTTCGGGAGAAGCGG TGGACGAACCGGCCGCCGATGGAGTGGTCAGCCCCCGCCAGCTCGCGCTGCTCGCGT CCATGGTGGATGAAGCCGTGAGAACTATCCCCTCACCTCCGCCGGAACGGGATGGA GCTCAAGAGGAAGCCGCCAGAAGCCCGTCCCCTCCGAGAACTCCATCCATGCGGGC CGACTACGGCGAAGAGAATGACGACGATGATGACGACGATGATGACGATGACCGCG ATGCCGGACGGTGGGTCCGCGGACCTGAGACTACCTCCGCCGTGCGCGGAGCCTAC CCTGATCCGATGGCCTCACTTAGCCCCCGGCCACCCGCCCCCCGCCGCCACCACCAC CATCATCACCACCGCAGAAGAAGGGCTCCCAGGCGCAGATCAGCAGCTTCCGACAG CTCGAAGTCCGGCTCCTCGTCCTCCGCCAGCAGCGCATCCTCGTCAGCGTCCTCATC GTCCAGCGCCTCGGCGAGCTCCTCCGACGATGACGACGACGACGATGCCGCCAGAG CTCCGGCATCAGCCGCGGACCATGCCGCCGGAGGAACCCTCGGTGCCGACGACGAG GAGGCCGGCGTGCCTGCCCGCGCTCCGGGAGCTGCTCCTAGGCCTTCACCACCCCGG GCGGAGCCAGCCCCTGCCAGAACGCCAGCAGCCACCGCTGGGCGATTGGAGAGGCG GAGAGCCCGGGCCGCCGTGGCCGGTCGGGATGCCACCGGCCGCTTCACTGCCGGAC GCCCTCGGCGCGTCGAACTGGACGCAGACGCCGCCTCGGGCGCGTTCTACGCCCGCT ATCGGGACGGTTATGTGTCCGGCGAGCCTTGGCCTGGTGCCGGTCCTCCTCCGCCTG GGAGAGTGCTCTACGGGGGTCTGGGTGATTCTCGGCCAGGGTTGTGGGGAGCCCCC GAGGCGGAGGAAGCCAGAGCCCGCTTCGAAGCATCCGGAGCACCGGCCCCTGTGTG GGCGCCGGAACTGGGCGACGCCGCCCAACAATACGCCCTGATCACACGCCTGCTCT ACACTCCGGACGCCGAAGCCATGGGCTGGCTGCAGAACCCGAGAGTGGCCCCGGGT GATGTGGCCCTGGACCAGGCATGCTTCAGGATTAGCGGAGCCGCGAGAAACTCGAG CAGCTTTATCTCAGGATCTGTGGCCCGAGCCGTGCCGCACCTGGGCTACGCGATGGC CGCCGGACGCTTCGGATGGGGGCTGGCCCATGTCGCTGCCGCGGTGGCGATGTCCCG GCGGTACGACCGGGCTCAGAAGGGTTTCCTCCTCACCAGCCTCCGGAGGGCATACGC CCCGTTGCTGGCTCGGGAGAACGCCGCTCTGACTGGCGCCCGCACTCCTGATGACGG TGGCGACGCCAACCGCCACGACGGCGACGATGCACGGGGAAAGCCCGCGGCCGCCG CCGCCCCCCTTCCTAGCGCAGCCGCTTCGCCTGCCGACGAACGGGCTGTCCCTGCCG GATACGGAGCCGCCGGTGTGCTGGCGGCCCTTGGGAGACTGTCAGCCGCGCCTGCTT CAGCGCCGGCCGGAGCCGACGATGACGACGACGACGATGGAGCCGGAGGAGGGGG CGGCGGTCGGAGAGCAGAAGCCGGCAGGGTGGCAGTCGAATGCCTTGCTGCCTGTC GCGGGATCCTCGAGGCGTTGGCCGAAGGCTTCGACGGCGACCTGGCGGCAGTGCCT GGCCTGGCCGGCGCCCGCCCCGCTGCCCCTCCACGGCCCGGTCCGGCCGGGGCCGC AGCCCCTCCGCATGCTGACGCGCCTCGCCTCAGAGCATGGCTGAGAGAATTGAGATT TGTGCGGGATGCGCTGGTCCTTATGCGCCTGAGGGGGGATCTGAGGGTGGCCGGAG GTTCCGAGGCGGCCGTGGCTGCTGTGCGGGCCGTGTCCCTGGTGGCCGGTGCGCTGG GTCCCGCTCTGCCGCGGTCCCCTAGATTGCTTTCCTCAGCGGCCGCCGCCGCAGCCG ATCTGCTCTTTCAGAACCAAAGCCTCAGGCCGCTGCTGGCCGACACTGTCGCCGCTG

Strain	Nucleic Acid Sequence
	CGGACTCCCTCGCTGCCCCAGCCTCGGCCCCAAGAGAGGCTGCCGATGCCCCTCGCC CCGCCGCGGCCCCGCCTGCCGGAGCAGCGCCGCCTGCACCCCCTACTCCCCCCCCGC GACCGCCACGCCCAGCCGCTCTTACCAGAAGGCCAGCTGAGGGTCCTGACCCGCAG GGCGGCTGGCGCAGACAGCCCCCGGGACCTTCCCACACTCCCGCCCCATCTGCGGCT GCCCTTGAAGCATACTGTGCCCCGAGAGCTGTGGCGGAGCTGACCGACCACCCTCTG TTCCCTGCACCTTGGCGGCCTGCCCTGATGTTTGACCCGAGAGCGTTGGCCTCCCTGG CGGCCAGATGTGCGGCCCCGCCTCCCGGAGGAGCCCCAGCTGCATTCGGACCTCTGC GGGCATCCGGACCACTGCGGCGCGCTGCTGCATGGATGCGGCAAGTGCCGGACCCT GAGGACGTTCGCGTGGTCATTCTTTACTCCCCCCTGCCGGGAGAAGATCTCGCCGCC GGCCGCGCGGGAGGAGGCCCTCCACCCGAGTGGTCCGCTGAACGGGGAGGCCTGTC CTGCCTGCTGGCTGCCCTGGGAAACCGCCTGTGCGGACCAGCTACTGCCGCCTGGGC TGGAAACTGGACCGGCGCACCCGATGTGTCAGCCCTCGGAGCGCAGGGAGTGCTGC TGCTGTCAACTCGCGACCTGGCATTCGCCGGAGCTGTGGAGTTCCTGGGTCTGCTTG CCGGCGCGTGCGACCGGAGATTGATCGTCGTGAACGCTGTCAGAGCGGCCGCTTGG CCTGCCGCTGCTCCGGTGGTCAGCCGGCAGCACGCATATCTGGCCTGCGAGGTGCTG CCCGCCGTGCAGTGTGCCGTGCGGTGGCCAGCGGCCAGAGACTTGCGACGGACCGT GCTGGCCTCCGGTAGGGTCTTTGGCCCCGGAGTGTTCGCCCGCGTGGAGGCCGCCCA TGCCAGACTGTACCCCGACGCACCGCCCCTGAGACTGTGCCGGGGAGCCAACGTGC GGTACAGAGTCCGCACCCGCTTCGGACCCGATACTCTGGTGCCAATGTCACCGCGGG AATATAGGAGAGCCGTGCTCCCGGCACTGGACGGCAGAGCCGCCGCATCCGGTGCT GGGGACGCGATGGCACCCGGAGCCCCCGACTTTTGCGAGGATGAAGCCCACAGCCA TCGGGCCTGTGCCAGATGGGGCCTGGGTGCCCCTCTTCGCCCCGTGTACGTGGCCCT GGGGAGAGATGCCGTCCGCGGTGGACCAGCCGAGCTGAGAGGCCCACGCCGGGAAT TTTGCGCTCGGGCCCTGCTCGAGCCCGATGGAGATGCGCCTCCCCTTGTGCTGCGCG ACGACGCTGACGCCGGCCCACCTCCGCAAATCCGGTGGGCCAGCGCCGCCGGTCGA GCAGGAACGGTGTTGGCAGCAGCCGGAGGAGGAGTCGAAGTGGTCGGAACCGCGG CTGGACTGGCAACCCCGCCAAGGCGCGAACCTGTGGATATGGACGCCGAGCTGGAG GATGACGACGATGGCCTTTTCGGCGAGTGATGATAATAGGCTGGAGCCTCGGTGGCC ATGCTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCGTACC CCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC (SEQ ID NO: 65)
HSV mRNA Sequences	
Strain	**Nucleic Acid Sequence**
HSV-2 gB_DX	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGAGAGGUGGUGGCUU AGUUUGCGCGCUGGUUGUCGGGGCGCUCGUAGCCGCCGUGGCGUCGGCCGCCCCU GCGGCUCCUCGCGCUAGCGGAGGCGUAGCCGCAACAGUUGCGGCGAACGGGGGUC CAGCCUCUCAGCCUCCUCCCGUCCCGAGCCCUGCGACCACCAAGGCUAGAAAGCG GAAGACCAAGAAACCGCCCAAGCGCCCCGAGGCCACCCCGCCCCCCGAUGCCAACG CGACUGUCGCCGCUGGCCAUGCGACGCUUCGCGCUCAUCUGAGGGAGAUCAAGGU UGAAAAUGCUGAUGCCCAAUUUUACGUGUGCCCGCCCCCGACGGGCGCCACGGUU GUGCAGUUUGAACAGCCGCGGCGCUGUCCGACGCGGCCAGAAGGCCAGAACUAUA CGGAGGGCAUAGCGGUGGUCUUUAAGGAAAACAUCGCCCCGUACAAAUUUAAGGC CACAAUGUACUACAAAGACGUGACAGUUUCGCAAGUGUGGUUUGGCCACAGAUAC UCGCAGUUUAUGGGAAUCUUCGAAGAUAGAGCCCCUGUUCCCUUCGAGGAAGUCA UCGACAAGAUUAAUGCCAAAGGGGUAUGCCGUUCCACGGCCAAAUACGUGCGCAA CAAUAUGGAGACCACCGCCUUUCACCGGGAUGAUCACGAGACCGACAUGGAGCUU AAGCCGGCGAAGGUCGCCACGCGUACCUCCCGGGGUUGGCACACCACAGAUCUUA AGUACAAUCCCUCGCGAGUUGAAGCAUUCCAUCGGUAUGGAACUACCGUUAACUG CAUCGUUGAGGAGGUGGAUGCGCGGUCGGUGUACCCUUACGAUGAGUUUGUGUU AGCGACCGGCGAUUUUGUGUACAUGUCCCCGUUUUACGGCUACCGGGAGGGGUCG CACACCGAACAUACCUCGUACGCCGCUGACAGGUUCAAGCAGGUCGAUGGCUUUU ACGCGCGCGAUCUCACCACGAAGGCCCGGGCCACGUCACCGACGACCAGGAACUU GCUCACGACCCCCAAGUUCACCGUCGCUUGGGAUUGGGUCCCAAAGCGUCCGGCG GUCUGCACGAUGACCAAAUGGCAGGAGGUGGACGAAAUGCUCCGCGCAGAAUACG GCGGCUCCUUCCGCUUCUCGUCCGACGCCAUCUCGACAACCUUCACCACCAAUCU GACCCAGUACAGUCUGUCGCGCGUUGAUUUAGGAGACUGCAUUGGCCGGGAUGCC CGGGAGGCCAUCGACAGAAUGUUUGCGCGUAAGUACAAUGCCACACAUAUUAAGG UGGGCCAGCCGCAAUACUACCUUGCCACGGGCGGCUUUCUCAUCGCGUACCAGCC

Strain	Nucleic Acid Sequence
	CCUUCUCUCAAAUACGCUCGCUGAACUGUACGUGCGGGAGUAUAUGAGGGAACAG GACCGCAAGCCCCGCAAUGCCACGCCUGCGCCACUACGAGAGGCGCCUUCAGCUA AUGCGUCGGUGGAACGUAUCAAGACCACCUCCUCAAUAGAGUUCGCCCGGCUGCA AUUUACGUACAACCACAUCCAGCGCCACGUGAACGACAUGCUGGGCCGCAUCGCU GUCGCCUGGUGCGAGCUGCAGAAUCACGAGCUGACUCUUUGGAACGAGGCCCGAA AACUCAACCCCAACGCGAUCGCCUCCGCAACAGUCGGUAGACGGGUGAGCGCUCG CAUGCUAGGAGAUGUCAUGGCUGUGUCCACCUGCGUGCCCGUCGCUCCGGACAAC GUGAUUGUGCAGAAUUCGAUGCGGGUCUUGAUAAUAGGCUGGAGCCUCGGUGGC CAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCUCCUCCCCUUCCUGCACCCGU ACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGGCGGC (SEQ ID NO: 90)
HSV-2 gC_DX	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGCCCUUGGACGGGU AGGCCUAGCCGUGGGCCUGUGGGGCCUACUGUGGGUGGGGUGUGGUCGUGGUGCU GGCCAAUGCCUCCCCCGGACGCACGAUAACGGUGGGCCCGCGAGGCAACGCGAGC AAUGCUGCCCCCUCCGCGUCCCCGCGGAACGCAUCCGCCCCCCGAACCACACCCAC GCCCCCACAACCCCGCAAAGCGACGAAAUCCAAGGCCUCCACCGCCAAACCGGCUC CGCCCCCCAAGACCGGACCCCCGAAGACAUCCUCGGAGCCCGUGCGAUGCAACCGC CACGACCCGCUGGCCCGGUACGGCUCGCGGGUGCAAAUCCGAUGCCGGUUUCCCA ACUCCACGAGGACUGAGUCCCGUCUCCAGAUCUGGCGUUAUGCCACGGCGACGGA CGCCGAAAUCGGAACAGCGCCUAGCUUAGAAGAGGUGAUGGUGAACGUGUCGGCC CCGCCCGGGGGCCAACUGGUGUAUGACAGUGCCCCCAACCGAACGGACCCGCAUG UAAUCUGGGCGGAGGGCGCCGGCCCGGGCGCCAGCCCGCGCCUGUACUCGGUUGU CGGCCCGCUGGGUCGGCAGCGGCUCAUCAUCGAAGAGUUAACCCUGGAGACACAG GGCAUGUACUAUUGGGUGUGGGGCCGGACGGACCGCCCGUCCGCCUACGGGACCU GGGUCCGCGUUCGAGUAUUUCGCCCUCCGUCGCUGACCAUCCACCCCCACGCGGU GCUGGAGGGCCAGCCGUUUAAGGCGACGUGCACGGCCGCAACCUACUACCCGGGC AACCGCGCGGAGUUCGUCUGGUUUGAGGACGGUCGCCGCGUAUUCGAUCCGGCAC AGAUACACACGCAGACGCAGGAGAACCCCGACGGCUUUUCCACCGUCUCCACCGU GACCUCCGCGGCCGUCGGCGGGCAGGGCCCCCCUCGCACCUUCACCUGCCAGCUGA CGUGGCACCGCGACUCCGUGUCGUUCUCUCGGCGCAACGCCAGCGGCACGGCCUC GGUUCUGCCGCGGCCGACCAUUACCAUGGAGUUUACAGGCGACCAUGCGGUCUGC ACGGCCGGCUGUGUGCCCGAGGGGGUCACGUUUGCUUGGUUCCUGGGGGAUGACU CCUCGCCGGCGGAAAAGGUGGCCGUCGCGUCCCAGACAUCGUGCGGGCGCCCCGG CACCGCCACGAUCCGCUCCACCCUGCCGGUCUCGUACGAGCAGACCGAGUACAUC UGUAGACUGGCGGGAUACCCGGACGGAAUUCCGGUCCUAGAGCACCACGGAAGCC ACCAGCCCCCGCCGCGGGACCCAACCGAGCGGCAGGUGAUCCGGGCGGUGGAGGG GGCGGGGAUCGGAGUGGCUGUCCUUGUCGCGGUGGUUCUGGCCGGGACCGCGGUA GUGUACCUGACCCAUGCCUCCUCGGUACGCUAUCGUCGGCUGCGGUAAUGAUAAU AGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCCAGCCCCU CCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGGCG GC (SEQ ID NO: 91)
HSV-2 gD_DX	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGGGCGUUUGACCUC CGGCGUCGGGACGGCGGCCCUGCUAGUUGUCGCGGUGGGACUCCGCGUCGUCUGC GCCAAAUACGCCUUAGCAGACCCCUCGCUUAAGAUGGCCGAUCCCAAUCGAUUUC GCGGGAAGAACCUUCCGGUUUUGGACCAGCUGACCGACCCCCCCGGGGUGAAGCG UGUUUACCACAUUCAGCCGAGCCUGGAGGACCCGUUCCAGCCCCCCAGCAUCCCG AUCACUGUGUACUACGCAGUGCUGGAACGUGCCUGCCGCAGCGUGCUCCUACAUG CCCCAUCGGAGGCCCCCCAGAUCGUGCGCGGGGCUUCGGACGAGGCCCGAAAGCA CACGUACAACCUGACCAUCGCCUGGUAUCGCAUGGGAGACAAUUGCGCUAUCCCC AUCACGGUUAUGGAAUACACCGAGUGCCCCUACAACAAGUCGUUGGGGGUCUGCC CCAUCCGAACGCAGCCCCGCUGGAGCUACUAUGACAGCUUUAGCGCCGUCAGCGA GGAUAACCUGGGAUUCCUGAUGCACGCCCCCGCCUUCGAGACCGCGGGUACGUAC CUGCGGCUAGUGAAGAUAAACGACUGGACGGAGAUCACACAAUUUAUCCUGGAGC ACCGGGCCCGCGCCUCCUGCAAGUACGCUCUCCCCCCUGCGCAUCCCCCCGGCAGCG UGCCUCACCUCGAAGGCCUACCAACAGGGCGUGACGGUCGACAGCAUCGGGAUGC UACCCCGCUUUAUCCCCGAAAACCAGCGCACCGUCGCCCUAUACAGCUUAAAAAU CGCCGGGUGGCACGGCCCCAAGCCCCCGUACACCAGCACCCUGCUGCCGCCGGAGC UGUCCGACACCACCAACGCCACGCAACCCGAACUCGUUCCGGAAGACCCCGAGGA

Strain	Nucleic Acid Sequence
	CUCGGCCCUCUUAGAGGAUCCCGCCGGGACGGUGUCUUCGCAGAUCCCCCCAAAC UGGCACAUCCCGUCGAUCCAGGACGUCGCACCGCACCACGCCCCCGCCGCCCCCAG CAACCCGGGCCUGAUCAUCGGCGCGCUGGCCGGCAGUACCCUGGCGGUGCUGGUC AUCGGCGGUAUUGCGUUUUGGGUACGCCGCCGCGCUCAGAUGGCCCCCAAGCGCC UACGUCUCCCCCCACAUCCGGGAUGACGACGCGCCCCCCUCGCACCAGCCAUUGUU UUACUAGUGAUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCC UCCCCCCAGCCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAA GUCUGAGUGGGCGGC (SEQ ID NO: 92)
HSV-2 gE_DX	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGCUAGGGGGGCCGG GUUGGUUUUUUUUGUUGGAGUUUGGGUCGUAAGCUGCCUCGCGGCAGCGCCCAG AACGUCCUGGAAACGCGUAACCUCGGGCGAAGACGUGGUGUUACUCCCCGCGCCG GCGGGGCCGGAAGAACGCACUCGGGCCCACAAACUACUGUGGGCAGCGGAACCGC UGGAUGCCUGCGGUCCCCUGAGGCCGUCAUGGGUGGCACUGUGGCCCCCCCGACG AGUGCUUGAGACGGUUGUCGAUGCGGCGUGCAUGCGCGCCCCGGAACCGCUCGCU AUCGCAUACAGUCCCCCGUUCCCUGCGGGCGACGAGGGACUUUAUUCGGAGUUGG CGUGGCGCGAUCGCGUAGCCGUGGUCAACGAGAGUUUAGUUAUCUACGGGGCCCU GGAGACGGACAGUGGUCUGUACACCCUGUCAGUGGUGGGCCUAUCCGACGAGGCC CGCCAAGUGGCGUCCGUGGUUCUCGUCGUCGAGCCCGCCCCUGUGCCUACCCCGA CCCCCGAUGACUACGACGAGGAGGAUGACGCGGGCGUGAGCGAACGCACGCCCGU CAGCGUUCCCCCCCCCAACACCCCCCCGACGUCCCCCCGUCGCCCCCCCGACGCACC CUCGUGUUAUCCCUGAGGUGAGCCACGUGCGGGGGGUGACGGUCCACAUGGAAAC CCCGGAGGCCAUUCUGUUUGCGCCAGGGGAGACGUUUGGGACGAACGUCUCCAUC CACGCAAUUGCCCACGACGACGGUCCGUACGCCAUGGACGUCGUCUGGAUGCGAU UUGAUGUCCCGUCCUCGUGCGCCGAGAUGCGGAUCUAUGAAGCAUGUCUGUAUCA CCCGCAGCUGCCUGAGUGUCUGUCUCCGGCCGAUGCGCCGUGCGCCGUAAGUUCG UGGGCGUACCGCCUGGCGGUCCGCAGCUACGCCGGCUGCUCCAGGACUACGCCCC CACCUCGAUGUUUUGCUGAAGCUCGCAUGGAACCGGUCCCCGGGUUGGCGUGGCU CGCAUCAACUGUUAAUCUGGAAUUCCAGCAUGCCUCUCCCCAACACGCCGGCCUC UAUCUGUGUGUGGUGUAUGUGGACGACCAUAUCCAUGCCUGGGGCCACAUGACCA UCUCCACAGCGGCCCAGUACCGGAAUGCGGUGGUGGAACAGCAUCUCCCCCAGCG CCAGCCCGAGCCCGUAGAACCCACCCGACCGCAUGUGAGAGCCCCCCCUCCCGCAC CCUCCGCGAGAGGCCCGUUACGCUUAGGUGCGGUCCUGGGGGCGGCCCUGUUGCU CGCGGCCCUCGGGCUAUCCGCCUGGGCGUGCAUGACCUGCUGGCGCAGGCGCAGU UGGCGGGCGGUUAAAAGUCGGGCCUCGGCGACCGGCCCCACUUACAUUCGAGUAG CGGAUAGCGAGCUGUACGCGGACUGGAGUUCGGACUCAGAGGGCGAGCGCGACGG UUCCCUGUGGCAGGACCCUCCGGAGAGACCCGACUCACCGUCCACAAAUGGAUCC GGCUUUGAGAUCUUAUCCCCAACGGCGCCCUCUGUAUACCCCCAUAGCGAAGGGC GUAAAUCGCGCCGCCCGCUCACCACCUUUGGUUCAGGAAGCCCGGGACGUCGUCA CUCCCAGGCGUCCUAUUCUUCCGUCUUAUGGUAAUGAUAAUAGGCUGGAGCCUCG GUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCUCCUCCCCUUCCUGCA CCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGGCGGC (SEQ ID NO: 93)
HSV-2 gI_DX	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGCCCGGCCGCUCGCUG CAGGGCCUGGCGAUCCUGGGCCUGUGGGUCUGCGCCACCGGCCUGGUCGUCCGCG GCCCCACGGUCAGUCUGGUCUCAGACUCACUCGUGGAUGCCGGGGCCGUGGGGCC CCAGGGCUUCGUGGAAGAGGACCUGCGUGUUUUCGGGGAGCUUCAUUUUGUGGG GGCCCAGGUCCCCCACACAAACUACUACGACGGCAUCAUCGAGCUGUUUCACUAC CCCCUGGGGAACCACUGCCCCCGCGUUGUACACGUGGUCACACUGACCGCAUGCC CCCGCCGCCCCCGCCGUGGCGUUCACCUUGUGUCGCUCGACGCACCACGCCCACAGC CCCGCCUAUCCGACCCUGGAGCUGGGUCUGGCGCGGCAGCCGCUUCUGCGGGUUC GAACGGCAACGCGCGACUAUGCCGGUCUGUAUGUCCUGCGCGUAUGGGUCGGCAG CGCGACGAACGCCAGCCUGUUUGUUUUGGGGGUGGCGCUCUCUGCCAACGGGACG UUUGUGUAUAACGGCUCGGACUACGGCUCCUGCGAUCCGGCGCAGCUUCCCUUUU CGGCCCCGCGCCUGGGACCCUCGAGCGUAUACACCCCCGGAGCCUCCCGGCCCACC CCUCCACGGACAACGACAUCACCGUCCUCCCCACGAGACCCGACCCCCGCCCCCGG GGACACAGGGACGCCUGCUCCCGCGAGCGGCGAGAGAGCCCCGCCCAAUUCCACG CGAUCGGCCAGCGAAUCGAGACACAGGCUAACCGUAGCCCAGGUAAUCCAGAUCG CCAUACCGGCGUCCAUCAUCGCCUUUGUGUUUCUGGGCAGCUGUAUCUGCUUCAU

Strain	Nucleic Acid Sequence
	CCAUAGAUGCCAGCGCCGAUACAGGCGCCCCCGCGGCCAGAUUUACAACCCCGGG GGCGUUUCCUGCGCGGUCAACGAGGCGGCCAUGGCCCGCCUCGGAGCCGAGCUGC GAUCCCACCCAAACACCCCCCCCAAACCCCGACGCCGUUCGUCGUCGUCCACGACC AUGCCUUCCCUAACGUCGAUAGCUGAGGAAUCGGAGCCAGGUCCAGUCGUGCUGC UGUCCGUCAGUCCUCGGCCCCGCAGUGGCCCGACGGCCCCCCAAGAGGUCUAGUG AUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAG CCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGU GGGCGGC (SEQ ID NO: 94)
HSV-2 SgB_DX	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGCGCGGGGGGGGCUU AGUUUGCGCGCUGGUCGUGGGGGCGCUCGUAGCCGCGGUCGCGUCGGCGGCUCCG GCUGCCCCACGCGCUUCAGGUGGUGUCGCUGCGACCGUUGCGGCGAAUGGUGGUC CCGCCAGCCAACCGCCUCCCGUCCCGAGCCCCGCGACCACUAAGGCCCGGAAGCGG AAGACCAAGAAGCCACCCAAGCGGCCCGAGGCGACUCCGCCCCCAGACGCCAACG CGACCGUCGCCGCCGGCCACGCCACUCUGCGUGCGCACCUGCGGGAAAUCAAGGU CGAGAACGCGGACGCCCAGUUUUACGUGUGCCCGCCGCCGACUGGCGCCACGGUG GUGCAGUUUGAGCAACCUAGGCGCUGCCCGACGCGACCAGAGGGGCAGAACUACA CCGAGGGCAUAGCGGUGGUCUUUAAGGAAAACAUCGCCCCGUACAAAUUCAAGGC CACCAUGUACUACAAAGACGUGACCGUGUCGCAGGUGUGGUUCGGCCACCGCUAC UCCCAGUUUAUGGGGAUAUUCGAGGACCGCGCCCCCGUUCCCUUCGAAGAGGUGA UUGACAAAAUUAACGCCAAGGGGGUCUGCCGCAGUACGGCGAAGUACGUCCGGAA CAACAUGGAGACCACUGCCUUCCACCGGGACGACCACGAAACAGACAUGGAGCUC AAACCGGCGAAAGUCGCCACGCGCACGAGCCGGGGGUGGCACACCACCGACCUCA AAUACAAUCCUUCGCGGGUGGAAGCAUUCCAUCGGUAUGGCACGACCGUCAACUG UAUCGUAGAGGAGGUGGAUGCGCGGUCGGUGUACCCCUACGAUGAGUUCGUGCU GGCAACGGGCGAUUUUGUGUACAUGUCCCCUUUUUACGGCUACCGGGAAGGUAGU CACACCGAGCACACCAGUUACGCCGCCGACCGCUUUAAGCAAGUGGACGGCUUCU ACGCGCGCGACCUCACCACAAAGGCCCGGGCCACGUCGCCGACGACCCGCAAUUU GCUGACGACCCCCAAGUUUACCGUGGCCUGGGACUGGGUGCCUAAGCGACCGGCG GUCUGUACCAUGACAAAGUGGCAGGAGGUGGACGAAAUGCUCCGCGCUGAAUACG GUGGCUCUUUCCGCUUCUCUUCCGACGCCAUCUCCACCACGUUCACCACCAACCU GACCCAAUACUCGCUCUCGAGAGUCGAUCUGGGAGACUGCAUUGGCCGGGAUGCC CGCGAGGCAAUUGACCGCAUGUUCGCGCGCAAGUACAACGCUACGCACAUAAAGG UUGGCCAACCCCAGUACUACCUAGCCACGGGGGGCUUCCUCAUCGCUUAUCAACC CCUCCUCAGCAACACGCUCGCCGAGCUGUACGUGCGGGAAUAUAUGCGGGAACAG GACCGCAAACCCCGAAACGCCACGCCCGCGCCGCUGCGGGAAGCACCGAGCGCCA ACGCGUCCGUGGAGCGCAUCAAGACGACAUCCUCGAUUGAGUUUGCUCGUCUGCA GUUUACGUAUAACCACAUACAGCGCCAUGUAAACGACAUGCUCGGGCGCAUCGCC GUCGCGUGGUGCGAGCUCCAAAAUCACGAGCUCACUCUGUGGAACGAGGCACGCA AGCUCAAUCCCAACGCCAUCGCAUCCGCCACCGUAGGCCGGCGGGUGAGCGCUCG CAUGCUCGGGGAUGUCAUGGCCGUCUCCACGUGCGUGCCCGUCGCCCCGGACAAC GUGAUCGUGCAAAAUAGCAUGCGCGUUUCUUCGCGGCCGGGGACGUGCUACAGCC GCCCGCUGGUUAGCUUUCGGUACGAAGACCAAGGCCCGCUGAUUGAGGGGCAGCU GGGUGAGAACAACGAGCUGCGCCUCACCCGCGAUGCGUUAGAGCCGUGUACCGUC GGCCACCGGCGCUACUUCAUCUUCGGAGGGGGAUACGUAUACUUCGAAGAAUAUG CGUACUCUCACCAAUUGAGUCGCGCCGAUGUCACCACUGUUAGCACCUUCAUCGA CCUGAACAUCACCAUGCUGGAGGACCACGAGUUCGUGCCCCUGGAGGUCUACACA CGCCACGAGAUCAAGGAUUCCGGCCUACUGGACUACACCGAAGUCCAGAGACGAA AUCAGCUGCACGAUCUCCGCUUUGCUGACAUCGAUACUGUUAUCCGCGCCGACGC CAACGCCGCCAUGUUCGCAGGUCUGUGUGCGUUUUUCGAGGGUAUGGGUGACUUA GGGCGCGCGGUGGGCAAGGUCGUCAUGGGGGUAGUCGGGGGCGUGGUGUCGGCC GUCUCGGGCGUCUCCUCCUUUAUGUCUAACCCCUGAUAAUAGGCUGGAGCCUCGG UGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCUCCUCCCCUUCCUGCAC CCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGGCGGC (SEQ ID NO: 95)
HSV-2 SgC_DX	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGCCCUUGGACGGGU GGGCCUAGCCGUGGGCCUGUGGGGCCUGCUGUGGGUGGGUGUUGUCGUGGUGCU GGCCAAUGCCUCCCCUGGACGCACGAUAACGGUGGGCCCGCGGGGGAACGCGAGC AAUGCCGCCCCAUCCGCGUCCCCGCGGAACGCAUCCGCCCCCCGAACCACACCCAC

Strain	Nucleic Acid Sequence
HSV-2 SgE_DX	UCCCCCCCAACCCCGCAAAGCGACGAAAAGUAAGGCCUCCACCGCCAAACCGGCCC CGCCCCCCAAGACCGGGCCCCCGAAGACAUCUUCUGAGCCCGUGCGCUGCAACCGC CACGACCCGCUGGCCCGGUACGGCUCGCGGGUGCAAAUCCGAUGUCGAUUUCCCA ACUCCACUCGCACGGAAUCCCGCCUCCAGAUCUGGCGUUAUGCCACGGCGACGGA CGCCGAGAUUGGAACUGCGCCUAGCUUAGAGGAGGUGAUGGUAAACGUGUCGGCC CCGCCCGGGGGCCAACUGGUGUAUGAUAGCGCACCUAACCGAACGGACCCGCACG UGAUUUGGGCGGAGGGCGCCGGACCUGGCGCCUCACCGCGGCUGUACUCGGUCGU CGGGCCGCUGGGUCGGCAGAGACUUAUCAUCGAAGAGCUGACCCUCGAGACACAG GGCAUGUAUUAUUGGGUGUGGGGCCGGACGGACCGCCCGUCCGCGUACGGGACCU GGGUGCGCGUUCGCGUGUUCCGCCCUCCUUCGCUGACCAUCCACCCCCACGCGGU GCUGGAGGGCCAGCCGUUUAAAGCGACGUGCACCGCCGCCACCUACUACCCGGGC AACCGCGCGGAGUUCGUCUGGUUCGAGGACGGUCGCCGGGUAUUCGAUCCGGCCC AGAUACAUACGCAGACGCAGGAAAACCCCGACGGCUUUUCCACCGUCUCCACCGU GACCUCCGCGGCCGUCGGCGGCCAGGGCCCCCCGCGCACCUUCACCUGUCAGCUGA CGUGGCACCGCGACUCCGUGUCGUUCUCUCGGCGCAAUGCCAGCGGCACGGCAUC GGUGCUGCCACGGCCAACCAUUACCAUGGAGUUUACGGGCGACCAUGCGGUCUGC ACGGCCGGCUGUGUGCCCGAGGGGGUGACGUUUGCCUGGUUCCUGGGGGACGACU CCUCGCCGGCCGAGAAGGUGGCCGUCGCGUCCCAGACCUCGUGCGGUCGCCCCGG CACCGCCACGAUCCGCUCCACACUGCCGGUCUCGUACGAGCAGACCGAGUACAUC UGCCGGCUGGCGGGAUACCCGGACGGAAUUCCGGUCCUAGAGCACCAUGGCAGCC ACCAGCCCCCGCCGCGGGACCCCACCGAACGGCAGGUGAUUCGGGCAGUGGAAGG GUGAUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCC CAGCCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUG AGUGGGCGGC (SEQ ID NO: 96)
HSV-2 SgE_DX	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGCUCGCGGGGCCGG GUUGGUGUUUUUUGUUGGAGUUUGGGUCGUAUCGUGCCUGGCGGCAGCACCCAG AACGUCCUGGAAACGGGUUACCUCGGGCGAGGACGUGGUGUUGCUUCCGGCGCCC GCGGGGCCGGAGGAACGCACACGGGCCCACAAACUACUGUGGGCCGCGGAACCCC UGGAUGCCUGCGGUCCCCUGAGGCCGUCGUGGGUGGCGCUGUGGCCCCCGCGACG GGUGCUCGAAACGGUCGUGGAUGCGGCGUGCAUGCGCGCCCCGGAACCGCUCGCC AUAGCAUACAGUCCCCCGUUCCCCGCGGGCGACGAGGGACUGUAUUCGGAGUUGG CGUGGCGCGAUCGCGUAGCCGUGGUCAACGAGAGUCUGGUCAUCUACGGGGCCCU GGAGACGGACAGCGGUCUGUACACCCUGUCCGUGGUCGGCCUAAGCGACGAGGCG CGCCAAGUGGCGUCGGUGGUUCUGGUCGUGGAGCCCGCCCCUGUGCCGACCCCGA CCCCCGACGACUACGACGAAGAAGACGACGCGGGCGUGAGCGAACGCACGCCGGU CAGCGUACCCCCCCCGACCCCACCCCGUCGUCCCCCCGUCGCCCCCCCUACGCACC CUCGUGUUAUCCCCGAGGUGUCCCACGUGCGCGGGGUAACGGUCCAUAUGGAGAC CCCGGAGGCCAUUCUGUUUGCCCCCGGAGAGACGUUUGGGACGAACGUCUCCAUC CACGCCAUUGCCCAUGACGACGGUCCGUACGCCAUGGACGUCGUCUGGAUGCGGU UUGACGUGCCGUCCUCGUGCGCCGAGAUGCGGAUCUACGAAGCUUGUCUGUAUCA CCCGCAGCUUCCAGAAUGUCUAUCUCCGGCCGACGCGCCGUGCGCUGUAAGUUCC UGGGCGUACCGCCUGGCGGUCCGCAGCUACGCCGGCUGUUCCAGGACUACGCCCC CGCCGCGAUGUUUUGCCGAGGCUCGCAUGGAACCGGUCCCGGGGUUGGCGUGGUU AGCCUCCACCGUCAACCUGGAAUUCCAGCACGCCUCCCCCUCAGCACGCCGGCCUUU ACCUGUGCGUGGUGUACGUGGACGAUCAUAUCCACGCCUGGGGCCACAUGACCAU CUCUACCGCGGCGCAGUACCGGAACGCGGUGGUGGAACAGCACUUGCCCCAGCGC CAGCCUGAACCCGUCGAGCCCACCCGCCCGCACGUAAGAGCACCCCCUCCCGCGCC UUCCGCGCGCGGCCCGCUGCGCUGAUAAUAGGCUGGAGCCUCGGUGGCCAUGCUU CUUGCCCCUUGGGCCUCCCCCCAGCCCCUCCUCCCCUUCCUGCACCCGUACCCCCG UGGUCUUUGAAUAAAGUCUGAGUGGGCGGC (SEQ ID NO: 97)
HSV-2 ICP-4	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGUCGGCGGAGCAGCG GAAGAAGAAGAAGACGACGACGACGACGCAGGGCCGCGGGGCCGAGGUCGCGAUG GCGGACGAGGACGGGGGACGUCUCCGGGCCGCGGCGGAGACGACCGGCGGCCCCG GAUCUCCGGAUCCAGCCGACGGACCGCCGCCCACCCCGAACCCGGACCGUCGCCCC GCCGCGCGGCCCGGGUUCGGGUGGCACGGUGGGCCGGAGGAGAACGAAGACGAGG CCGACGACGCCGCCGCCGAUGCCGAUGCCGACGAGGCGGCCCCGGCGUCCGGGGA GGCCGUCGACGAGCCUGCCGCGGACGGCGUCGUCUCGCCGCGGCAGCUGGCCCUG

Strain	Nucleic Acid Sequence
	CUGGCCUCGAUGGUGGACGAGGCCGUUCGCACGAUCCCGUCGCCCCCCCCGGAGC
	GCGACGGCGCGCAAGAAGAAGCGGCCCGCUCGCCUUCUCCGCCGCGGACCCCCUCC
	AUGCGCGCCGAUUAUGGCGAGGAGAACGACGACGACGACGACGACGACGAUGACG
	ACGACCGCGACGCGGGCCGCUGGGUCCGCGGACCGGAGACGACGUCCGCGGUCCG
	CGGGGCGUACCCGGACCCCAUGGCCAGCCUGUCGCCGCGACCCCCGGCGCCCCGCC
	GACACCACCACCACCACCACCACCGCCGCCGGCGCGCCCCCCGCCGGCGCUCGGCC
	GCCUCUGACUCAUCAAAAUCCGGAUCCUCGUCGUCGGCGUCCUCCGCCUCCUCCU
	CCGCCUCCUCCUCCUCGUCUGCAUCCGCCUCCUCGUCUGACGACGACGACGACGAC
	GACGCCGCCCGCGCCCCCGCCAGCGCCGCAGACCACGCCGCGGGCGGGACCCUCGG
	CGCGGACGACGAGGAGGCGGGGGUGCCCGCGAGGGCCCCGGGGGCGGCGCCCCGG
	CCGAGCCCGCCCAGGGCCGAGCCCGCCCCGGCCCGGACCCCCGCGGCGACCGCGGG
	CCGCCUGGAGCGCCGCCGGGCCCGCGCGGCGGUGGCCGGCCGCGACGCCACGGGCC
	GCUUCACGGCCGGGCGGCCCCGGCGGGUCGAGCUGGACGCCGACGCGGCCUCCGG
	CGCCUUCUACGCGCGCUACCGCGACGGGUACGUCAGCGGGGAGCCGUGGCCCGGG
	GCCGGCCCCCCGCCCCCGGGGCGCGUGCUGUACGGCGGGCUGGGCGACAGCCGCCC
	CGGCCUCUGGGGGGCGCCCGAGGCGGAGGAGGCGCGGGCCCGGUUCGAGGCCUCG
	GGCGCCCCGGCGCCCGUGUGGGCGCCCGAGCUGGGCGACGCGGCGCAGCAGUACG
	CCCUGAUCACGCGGCUGCUGUACACGCCGGACGCGGAGGCGAUGGGGUGGCUCCA
	GAACCCGCGCGUGGCGCCCGGGGACGUGGCGCUGGACCAGGCCUGCUUCCGGAUC
	UCGGGCGCGGCGCGCAACAGCAGCUCCUUCAUCUCCGGCAGCGUGGCGCGGGCCG
	UGCCCCACCUGGGGUACGCCAUGGCGGCGGGCCGCUUCGGCUGGGGCCUGGCGCA
	CGUGGCGGCCGCCGUGGCCAUGAGCCGCCGCUACGACCGCGCGCAGAAGGGCUUC
	CUGCUGACCAGCCUGCGCCGCGCCUACGCGCCCCUGCUGGCGCGCGAGAACGCGG
	CGCUGACCGGGGCGCGAACCCCCGACGACGGCGGCGACGCCAACCGCCACGACGG
	CGACGACGCCCGCGGGAAGCCCGCCGCCGCCGCCCCGUUGCCGUCGGCGGCGG
	CGUCGCCGGCCGACGAGCGCGCGGUGCCCGCCGGCUACGGCGCCGCGGGGGUGCU
	CGCCGCCCUGGGGCGCCUGAGCGCCGCGCCCGCCUCCGCGCCGGCCGGGGCCGACG
	ACGACGACGACGACGACGGCGCCGGCGGUGGUGGCGGCGGCCGGCGCGCGGAGGC
	GGGCCGCGUGGCCGUGGAGUGCCUGGCCGCCUGCCGCGGGAUCCUGGAGGCGCUG
	GCGGAGGGCUUCGACGGCGACCUGGCGGCCGUGCCGGGGCUGGCCGGAGCCCGGC
	CCGCCGCGCCCCCGCGCCCGGGGCCCGCGGGCGCGGCCGCCCCGCCGCACGCCGAC
	GCGCCCCGCCUGCGCGCCUGGCUGCGCGAGCUGCGGUUCGUGCGCGACGCGCUGG
	UGCUGAUGCGCCUGCGCGGGGACCUGCGCGUGGCCGGCGGCAGCGAGGCCGCCGU
	GGCCGCCGUGCGCGCCGUGAGCCUGGUCGCCGGGGCCCUGGGCCCGGCGCUGCCG
	CGGAGCCCGCGCCUGCUGAGCUCCGCCGCCGCCGCCGCCGCGGACCUGCUCUUCCA
	GAACCAGAGCCUGCGCCCCCUGCUGGCCGACACCGUCGCCGCGGCCGACUCGCUCG
	CCGCGCCCGCCUCCGCGCCGCGGGAGGCCGCGGACGCCCCCCGCCCCGCGGCCGCC
	CCUCCCGCGGGGGCCGCGCCCCCCGCCCCGCCGACGCCGCCGCCGCGGCCGCCGCG
	CCCCGCGGCGCUGACCCGCCGGCCCGCCGAGGGCCCCGACCCGCAGGGCGGCUGGC
	GCCGCCAGCCGCCGGGGCCCAGCCACACGCCGGCGCCCUCGGCCGCCGCCCUGGAG
	GCCUACUGCGCCCCGCGGGCCGUGGCCGAGCUCACGGACCACCCGCUCUUCCCCGC
	GCCGUGGCGCCCGGCCCUCAUGUUCGACCCGCGCGCGCUGGCCUCGCUGGCCGCGC
	GCUGCGCCGCCCCGCCCCCCGGCGGCGCGCCCGCCGCCUUCGGCCCGCUGCGCGCC
	UCGGGCCCGCUGCGCCGCGCGGCGGCCUGGAUGCGCCAGGUGCCCGACCCGGAGG
	ACGUGCGCGUGGUGAUCCUCUACUCGCCGCUGCCGGGCGAGGACCUGGCCGCGGG
	CCGCGCCGGGGGCGGGCCCCCCCCGGAGUGGUCCGCCGAGCGCGGCGGGCUGUCC
	UGCCUGCUGGCGGCCCUGGGCAACCGGCUCUGCGGGCCCGCCACGGCCGCCUGGG
	CGGGCAACUGGACCGGCGCCCCCGACGUCUCGGCGCUGGGCGCGCAGGGCGUGCU
	GCUGCUGUCCACGCGGGACCUGGCCUUCGCCGGCGCCGUGGAGUUCCUGGGGCUG
	CUGGCCGGCGCCUGCGACCGCCGCCUCAUCGUCGUCAACGCCGUGCGCGCCGCGGC
	CUGGCCCGCCGCUGCCCCCGUGGUCUCGCGGCAGCACGCCUACCUGGCCUGCGAG
	GUGCUGCCCGCCGUGCAGUGCGCCGUGCGCUGGCCGGCGGCGCGGGACCUGCGCC
	GCACCGUGCUGGCCUCCGGCCGCGUGUUCGGGCCGGGGGUCUUCGCGCGCGUGGA
	GGCCGCGCACGCGCGCCUGUACCCCGACGCGCCGCCGCUGCGCCUCUGCCGCGGGG
	CCAACGUGCGGUACCGCGUGCGCACGCGCUUCGGCCCCGACACGCUGGUGCCCAU
	GUCCCCGCGCGAGUACCGCCGCGCCGUGCUCCCGGCGCUGGACGGCCGGGCCGCCG
	CCUCGGGCGCGGGCGACGCCAUGGCGCCCGGCGCGCCGGACUUCUGCGAGGACGA
	GGCGCACUCGCACCGCGCCUGCGCGCGCUGGGGCCUGGGCGCGCCGCUGCGGCCC
	GUCUACGUGGCGCUGGGGCGCGACGCCGUGCGCGGCGGCCCGGCGGAGCUGCGCG

Strain	Nucleic Acid Sequence
	GGCCGCGGCGGGAGUUCUGCGCGCGGGCGCUGCUCGAGCCCGACGGCGACGCGCC CCCGCUGGUGCUGCGCGACGACGCGGACGCGGGCCCGCCCCCGCAGAUACGCUGG GCGUCGGCCGCGGGCCGCGCGGGGACGGUGCUGGCCGCGGCGGGCGGCGGCGUGG AGGUGGUGGGGACCGCCGCGGGGCUGGCCACGCCGCCGAGGCGCGAGCCCGUGGA CAUGGACGCGGAGCUGGAGGACGACGACGACGGACUGUUUGGGGAGUGAUGAUA AUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCC CUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGG CGGC (SEQ ID NO: 98)
HSV-2 SgI_DX	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGCCCGGCCGCUCGCUG CAGGGCCUGGCGAUCCUGGGCCUGUGGGUCUGCGCCACCGGCCUGGUCGUCCGCG GCCCCACGGUCAGUCUGGUCUCAGACUCACUCGUGGAUGCCGGGGCCGUGGGGCC CCAGGGCUUCGUGGAAGAGGACCUGCGUGUUUUCGGGGAGCUUCAUUUUGUGGG GGCCCAGGUCCCCCACACAAACUACUACGACGGCAUCAUCGAGCUGUUUCACUAC CCCCUGGGGAACCACUGCCCCCGCGUUGUACACGUGGUCACACUGACCGCAUGCC CCCGCCGCCCCCGCCGUGGCGUUCACCUUGUGUCGCUCGACGCACCACGCCCACAGC CCCGCCUAUCCGACCCUGGAGCUGGGUCUGGCGCGGCAGCCGCUUCUGCGGGUUC GAACGGCAACGCGCGACUAUGCCGGUCUGUAUGUCCUGCGCGUAUGGGUCGGCAG CGCGACGAACGCCAGCCUGUUUGUUUUGGGGGUGGCGCUCUCUGCCAACGGGACG UUUGUGUAUAACGGCUCGGACUACGGCUCCUGCGAUCCGGCGCAGCUUCCCUUUU CGGCCCCGCGCCUGGGACCCUCGAGCGUAUACACCCCCGGAGCCUCCCGGCCCACC CCUCCACGGACAACGACAUCCCCGUCCUCCCCUAGAGACCCGACCCCCGCCCCCGG GGACACAGGAACGCCUGCGCCCGCGAGCGGCGAGAGAGCCCCGCCCAAUUCCACG CGAUCGGCCAGCGAAUCGAGACACAGGCUAACCGUAGCCCAGGUAAUCCAGUGAU AAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCC CCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGG GCGGC (SEQ ID NO: 99)
HSV-2 SgD	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGGGCGUUUGACCUC CGGCGUCGGGACGGCGGCCCUGCUAGUUGUCGCGGUGGGACUCCGCGUCGUCUGC GCCAAAUACGCCUUAGCAGACCCCUCGCUUAAGAUGGCCGAUCCCAAUCGAUUUC GCGGGAAGAACCUUCCGGUUUUGGACCAGCUGACCGACCCCCCCGGGGUGAAGCG UGUUUACCACAUUCAGCCGAGCCUGGAGGACCCGUUCCAGCCCCCCAGCAUCCCG AUCACUGUGUACUACGCAGUGCUGGAACGUGCCUGCCGCAGCGUGCUCCUACAUG CCCCAUCGGAGGCCCCCCAGAUCGUGCGCGGGGCUUCGGACGAGGCCCGAAAGCA CACGUACAACCUGACCAUCGCCUGGUAUCGCAUGGGAGACAAUUGCGCUAUCCCC AUCACGGUUAUGGAAUACACCGAGUGCCCCUACAACAAGUCGUUGGGGGUCUGCC CCAUCCGAACGCAGCCCCGCUGGAGCUACUAUGACAGCUUUAGCGCCGUCAGCGA GGAUAACCUGGGAUUCCUGAUGCACGCCCCCGCCUUCGAGACCGCGGGUACGUAC CUGCGGCUAGUGAAGAUAAACGACUGGACGGAGAUCACACAAUUUAUCCUGGAGC ACCGGGCCCGCGCCUCCUGCAAGUACGCUCUCCCCCUGCGCAUCCCCCCGGCAGCG UGCCUCACCUCGAAGGCCUACCAACAGGGCGUGACGGUCGACAGCAUCGGGAUGC UACCCCGCUUUAUCCCCGAAAACCAGCGCACCGUCGCCCUAUACAGCUUAAAAAU CGCCGGGUGGCACGGCCCCAAGCCCCCGUACACCAGCACCCUGCUGCCGCCGGAGC UGUCCGACACCACCAACGCCACGCAACCCGAACUCGUUCCGGAAGACCCCGAGGA CUCGGCCCUCUUAGAGGAUCCCGCCGGGACGGUGUCUUCGCAGAUCCCCCCAAAC UGGCACAUCCCGUCGAUCCAGGACGUCGCGCCGCACCACGCCCCCGCCGCCCCCAG CAACCCGUGAUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCC UCCCCCCAGCCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAA GUCUGAGUGGGCGGC (SEQ ID NO: 100)
HSV-2 gB	AUGCGCGGGGGGGGCUUGGUUUGCGCGCUGGUCGUGGGGGCGCUGGUGGCCGCGG UGGCGUCGGCGGCCCCGGCGGCCCCCCGCGCCUCGGGCGGCGUGGCCGCGACCGUC GCGGCGAACGGGGGUCCCGCCUCCCAGCCGCCCCCCGUCCCGAGCCCCGCGACCAC CAAGGCCCGGAAGCGGAAAACCAAAAAGCCGCCCAAGCGGCCCGAGGCGACCCCG CCCCCCGACGCCAACGCGACCGUCGCCGCCGGCCACGCCACGCUGCGCGCGCACCU GCGGGAAAUCAAGGUCGAGAACGCCGAUGCCCAGUUUUACGUGUGCCCGCCCCCG ACGGGCGCCACGGUGGUGCAGUUUGAGCAGCCGCGCCGCUGCCCGACGCGCCCGG AGGGGCAGAACUACACGGAGGGCAUCGCGGUGGUCUUCAAGGAGAACAUCGCCCC GUACAAAUUCAAGGCCACCAUGUACUACAAAGACGUGACCGUGUCGCAGGUGUGG

Strain	Nucleic Acid Sequence
	UUCGGCCACCGCUACUCCCAGUUUAUGGGGAUAUUCGAGGACCGCGCCCCCGUUC CCUUCGAGGAGGUGAUCGACAAGAUUAACGCCAAGGGGGUCUGCCGCUCCACGGC CAAGUACGUGCGGAACAACAUGGAGACCACCGCGUUUCACCGGGACGACCACGAG ACCGACAUGGAGCUCAAGCCGGCGAAGGUCGCCACGCGCACGAGCCGGGGGUGGC ACACCACCGACCUCAAGUACAACCCCUCGCGGGUGGAGGCGUUCCAUCGGUACGG CACGACGGUCAACUGCAUCGUCGAGGAGGUGGACGCGCGGUCGGUGUACCCGUAC GAUGAGUUUGUGCUGGCGACGGGCGACUUUGUGUACAUGUCCCCGUUUUACGGCU ACCGGGAGGGGUCGCACACCGAGCACACCAGCUACGCCGCCGACCGCUUCAAGCA GGUCGACGGCUUCUACGCGCGCGACCUCACCACGAAGGCCCGGGCCACGUCGCCG ACGACCCGCAACUUGCUGACGACCCCCAAGUUUACCGUGGCCUGGGACUGGGUGC CGAAGCGACCGGCGGUCUGCACCAUGACCAAGUGGCAGGAGGUGGACGAGAUGCU CCGCGCCGAGUACGGCGGCUCCUUCCGCUUCUCCUCCGACGCCAUCUCGACCACCU UCACCACCAACCUGACCCAGUACUCGCUCUCGCGCGUCGACCUGGGCGACUGCAU CGGCCGGGAUGCCCGCGAGGCCAUCGACCGCAUGUUUGCGCGCAAGUACAACGCC ACGCACAUCAAGGUGGGCCAGCCGCAGUACUACCUGGCCACGGGGGGCUUCCUCA UCGCGUACCAGCCCCUCCUCAGCAACACGCUCGCCGAGCUGUACGUGCGGGAGUA CAUGCGGGAGCAGGACCGCAAGCCCCGGAAUGCCACGCCCGCGCCACUGCGGGAG GCGCCCAGCGCCAACGCGUCCGUGGAGCGCAUCAAGACCACCUCCUCGAUCGAGU UCGCCCGGCUGCAGUUUACGUAUAACCACAUACAGCGCCACGUGAACGACAUGCU GGGGCGCAUCGCCGUCGCGUGGUGCGAGCUGCAGAACCACGAGCUGACUCUCUGG AACGAGGCCCGCAAGCUCAACCCCAACGCCAUCGCCUCCGCCACCGUCGGCCGGCG GGUGAGCGCGCGCAUGCUCGGAGACGUCAUGGCCGUCUCCACGUGCGUGCCCGUC GCCCCGGACAACGUGAUCGUGCAGAACUCGAUGCGCGUCAGCUCGCGGCCGGGGA CGUGCUACAGCCGCCCCCUGGUCAGCUUUCGGUACGAAGACCAGGGCCCGCUGAU CGAGGGGCAGCUGGGCGAGAACAACGAGCUGCGCCUCACCCGCGACGCGCUCGAG CCGUGCACCGUGGGCCACCGGCGCUACUUCAUCUUCGGCGGGGGCUACGUGUACU UCGAGGAGUACGCGUACUCUCACCAGCUGAGUCGCGCCGACGUCACCACCGUCAG CACCUUCAUCGACCUGAACAUCACCAUGCUGGAGGACCACGAGUUUGUGCCCCUG GAGGUCUACACGCGCCACGAGAUCAAGGACAGCGGCCUGCUGGACUACACGGAGG UCCAGCGCCGCAACCAGCUGCACGACCUGCGCUUUGCCGACAUCGACACGGUCAU CCGCGCCGACGCCAACGCCGCCAUGUUCGCGGGGCUGUGCGCGUUCUUCGAGGGG AUGGGGGACUUGGGGCGCGCGGUCGGCAAGGUCGUCAUGGGAGUAGUGGGGGGC GUGGUGUCGGCCGUCUCGGGCGUGUCCUCCUUUAUGUCCAACCCCUUCGGGGCGC UUGCCGUGGGGCUGCUGGUCCUGGCCGGCCUGGUCGCGGCCUUCUUCGCCUUCCG CUACGUCCUGCAACUGCAACGCAAUCCCAUGAAGGCCCUGUAUCCGCUCACCACC AAGGAACUCAAGACUUCCGACCCCGGGGGCGUGGGCGGGGAGGGGGAGGAAGGCG CGGAGGGGGGCGGGUUUGACGAGGCCAAGUUGGCCGAGGCCCGAGAAAUGAUCCG AUAUAUGGCUUUGGUGUCGGCCAUGGAGCGCACGGAACACAAGGCCAGAAAGAA GGGCACGAGCGCCCUGCUCAGCUCCAAGGUCACCAACAUGGUUCUGCGCAAGCGC AACAAAGCCAGGUACUCUCCGCUCCACAACGAGGACGAGGCCGGAGACGAAGACG AGCUCUAA (SEQ ID NO: 101)
HSV-2 gC	AUGGCCCUUGGACGGGUGGGCCUAGCCGUGGGCCUGUGGGGCCUGCUGUGGGUGG GUGUGGUCGUGGUGCUGGCCAAUGCCUCCCCCGGACGCACGAUAACGGUGGGCCC GCGGGGGAACGCGAGCAAUGCCGCCCCCUCCGCGUCCCCGCGGAACGCAUCCGCCC CCCGAACCACACCCACGCCCCCCCAACCCCGCAAGGCGACGAAAAGUAAGGCCUCC ACCGCCAAACCGGCCCCGCCCCCCAAGACCGGGCCCCCGAAGACAUCCUCGGAGCC CGUGCGAUGCAACCGCCACGACCCGCUGGCCCGGUACGGCUCGCGGGUGCAAAUC CGAUGCCGGUUUCCCAACUCCACCCGCACGGAGUCCCGCCUCCAGAUCUGGCGUU AUGCCACGGCGACGGACGCCGAGAUCGGAACGGCGCCUAGCUUAGAGGAGGUGAU GGUAAACGUGUCGGCCCCGCCCGGGGGCCAACUGGUGUAUGACAGCGCCCCCAAC CGAACGGACCCGCACGUGAUCUGGGCGGAGGGCGCCGGCCCGGGCGCCAGCCCGC GGCUGUACUCGGUCGUCGGGCCGCUGGGUCGGCAGCGGCUCAUCAUCGAAGAGCU GACCCUGGAGACCCAGGGCAUGUACUACUGGGUGUGGGGCCGGACGGACCGCCCG UCCGCGUACGGGACCUGGGUGCGCGUUCGCGUGUUCCGCCCUCCGUCGCUGACCA UCCACCCCCACGCGGUGCUGGAGGGCCAGCCGUUUAAGGCGACGUGCACGGCCGC CACCUACUACCCGGGCAACCGCGCGGAGUUCGUCUGGUUCGAGGACGGUCGCCGG GUAUUCGAUCCGGCCCAGAUACACACGCAGACGCAGGAGAACCCCGACGGCUUUU CCACCGUCUCCACCGUGACCUCCGCGGCCGUCGGCGGCCAGGGCCCCCCGCGCACC UUCACCUGCCAGCUGACGUGGCACCGCGACUCCGUGUCGUUCUCUCGGCGCAACG

Strain	Nucleic Acid Sequence
	CCAGCGGCACGGCAUCGGUGCUGCCGCGGCCAACCAUUACCAUGGAGUUUACGGG CGACCAUGCGGUCUGCACGGCCGGCUGUGUGCCCGAGGGGGUGACGUUUGCCUGG UUCCUGGGGGGACGACUCCUCGCCGGCGGAGAAGGUGGCCGUCGCGUCCCAGACAU CGUGCGGGCGCCCCGGCACCGCCACGAUCCGCUCCACCCUGCCGGUCUCGUACGAG CAGACCGAGUACAUCUGCCGGCUGGCGGGAUACCCGGACGGAAUUCCGGUCCUAG AGCACCACGGCAGCCACCAGCCCCCGCCGCGGGACCCCACCGAGCGGCAGGUGAUC CGGGCGGUGGAGGGGGCGGGGAUCGGAGUGGCUGUCCUUGUCGCGGUGGUUCUG GCCGGGACCGCGGUAGUGUACCUCACCCACGCCUCCUCGGUGCGCUAUCGUCGGC UGCGGUAA (SEQ ID NO: 102)
HSV-2 gD	AUGGGGCGUUUGACCUCCGGCGUCGGGACGGCGGCCCUGCUAGUUGUCGCGGUGG GACUCCGCGUCGUCUGCGCCAAAUACGCCUUAGCAGACCCCUCGCUUAAGAUGGC CGAUCCCAAUCGAUUUCGCGGGAAGAACCUUCCGGUUUUGGACCAGCUGACCGAC CCCCCCGGGGUGAAGCGUGUUUACCACAUUCAGCCGAGCCUGGAGGACCCGUUCC AGCCCCCCAGCAUCCCGAUCACUGUGUACUACGCAGUGCUGGAACGUGCCUGCCG CAGCGUGCUCCUACAUGCCCCAUCGGAGGCCCCCCAGAUCGUGCGCGGGGCUUCG GACGAGGCCCGAAAGCACACGUACAACCUGACCAUCGCCUGGUAUCGCAUGGGAG ACAAUUGCGCUAUCCCCAUCACGGUUAUGGAAUACACCGAGUGCCCCUACAACAA GUCGUUGGGGGUCUGCCCCAUCCGAACGCAGCCCCGCUGGAGCUACUAUGACAGC UUUAGCGCCGUCAGCGAGGAUAACCUGGGAUUCCUGAUGCACGCCCCCGCCUUCG AGACCGCGGGUACGUACCUGCGGCUAGUGAAGAUAAACGACUGGACGGAGAUCAC ACAAUUUAUCCUGGAGCACCGGGCCCGCGCCUCCUGCAAGUACGCUCUCCCCCUG CGCAUCCCCCCGGCAGCGUGCCUCACCUCGAAGGCCUACCAACAGGGCGUGACGG UCGACAGCAUCGGGAUGCUACCCCGCUUUAUCCCCGAAAACCAGCGCACCGUCGC CCUAUACAGCUUAAAAAUCGCCGGGUGGCACGGCCCCAAGCCCCCGUACACCAGC ACCCUGCUGCCGCCGGAGCUGUCCGACACCACCAACGCCACGCAACCCGAACUCGU UCCGGAAGACCCCGAGGACUCGGCCCUCUUAGAGGAUCCCGCCGGGACGGUGUCU UCGCAGAUCCCCCCAAACUGGCACAUCCCGUCGAUCCAGGACGUCGCGCCGCACC ACGCCCCCGCCGCCCCCAGCAACCCGGGCCUGAUCAUCGGCGCGCUGGCCGGCAGU ACCCUGGCGGUGCUGGUCAUCGGCGGUAUUGCGUUUUGGGUACGCCGCCGCGCUC AGAUGGCCCCCAAGCGCCUACGUCUCCCCCCACAUCCGGGAUGACGACGCGCCCCCC UCGCACCAGCCAUUGUUUUACUAG (SEQ ID NO: 103)
HSV-2 gE	AUGGCUCGCGGGGCCGGGUUGGUGUUUUUUGUUGGAGUUUGGGUCGUAUCGUGC CUGGCGGCAGCACCCAGAACGUCCUGGAAACGGGUAACCUCGGGCGAGGACGUGG UGUUGCUUCCGGCGCCCGCGGGGCCGGAGGAACGCACCCGGGCCCACAAACUACU GUGGGCCGCGGAACCCCUGGAUGCCUGCGGUCCCCUGCGCCCGUCGUGGGUGGCG CUGUGGCCCCCCCGACGGGUGCUCGAGACGGUCGUGGAUGCGGCGUGCAUGCGCG CCCCGGAACCGCUCGCCAUAGCAUACAGUCCCCCGUUCCCCGCGGGCGACGAGGG ACUGUAUUCGGAGUUGGCGUGGCGCGAUCGCGUAGCCGUGGUCAACGAGAGUCUG GUCAUCUACGGGGCCCUGGAGACGGACAGCGGUCUGUACACCCUGUCCGUGGUCG GCCUAAGCGACGAGGCGCGCCAAGUGGCGUCGGUGGUUCUGGUCGUGGAGCCCGC CCCUGUGCCGACCCCGACCCCCGACGACUACGACGAAGAAGACGACGCGGGCGUG AGCGAACGCACGCCGGUCAGCGUUCCCCCCCCAACCCCCCCCCGUCGUCCCCCCGU CGCCCCCCCGACGCACCCUCGUGUUAUCCCCGAGGUGUCCCACGUGCGCGGGGUA ACGGUCCAUAUGGAGACCCCGGAGGCCAUUCUGUUUGCCCCCGGGGAGACGUUUG GGACGAACGUCUCCAUCCACGCCAUUGCCCACGACGACGGUCCGUACGCCAUGGA CGUCGUCUGGAUGCGGUUUGACGUGCCGUCCUCGUGCGCCGAGAUGCGGAUCUAC GAAGCUUGUCUGUAUCACCCGCAGCUUCCAGAGUGUCUAUCUCCGGCCGACGCGC CGUGCGCCGUAAGUUCCUGGGCGUACCGCCUGGCGGUCCGCAGCUACGCCGGCUG UUCCAGGACUACGCCCCCGCCGCGAUGUUUUGCCGAGGCUCGCAUGGAACCGGUC CCGGGGUUGGCGUGGCUGGCCUCCACCGUCAAUCUGGAAUUCCAGCACGCCUCCC CCCAGCACGCCGGCCUCUACCUGUGCGUGGUGUACGUGGACGAUCAUAUCCACGC CUGGGGCCACAUGACCAUCAGCACCGCGGCGCAGUACCGGAACGCGGUGGUGGAA CAGCACCUCCCCCAGCGCCAGCCCGAGCCCGUCGAGCCCACCCGCCCGCACGUGAG AGCCCCCCCUCCCGCGCCCUCCGCGCGCGGCCCGCUGCGCCUCGGGGCGGUGCUGG GGGCGGCCCUGUUGCUGGCCGCCCUCGGGCUGUCCGCGUGGGCGUGCAUGACCUG CUGGCGCAGGCGCUCCUGGCGGGCGGUUAAAAGCCGGGCCUCGGCGACGGGCCCC ACUUACAUUCGCGUGGCGGACAGCGAGCUGUACGCGGACUGGAGUUCGGACAGCG AGGGGGAGCGCGACGGGUCCCUGUGGCAGGACCCUCCGGAGAGACCCGACUCUCC CUCCACAAAUGGAUCCGGCUUUGAGAUCUUAUCACCAACGGCUCCGUCUGUAUAC

Strain	Nucleic Acid Sequence
	CCCCAUAGCGAGGGGCGUAAAUCUCGCCGCCCGCUCACCACCUUUGGUUCGGGAA GCCCGGGCCGUCGUCACUCCCAGGCCUCCUAUUCGUCCGUCCUCUGGUAA (SEQ ID NO: 104)
HSV-2 gI	AUGCCCGGCCGCUCGCUGCAGGGCCUGGCGAUCCUGGGCCUGUGGGUCUGCGCCA CCGGCCUGGUCGUCCGCGGCCCCACGGUCAGUCUGGUCUCAGACUCACUCGUGGA UGCCGGGGCCGUGGGGCCCCAGGGCUUCGUGGAAGAGGACCUGCGUGUUUUCGGG GAGCUUCAUUUUGUGGGGGCCCAGGUCCCCCACACAAACUACUACGACGGCAUCA UCGAGCUGUUUCACUACCCCCUGGGGAACCACUGCCCCCGCGUUGUACACGUGGU CACACUGACCGCAUGCCCCCGCCGCCCCGCCGUGGCGUUCACCUUGUGUCGCUCGA CGCACCACGCCCACAGCCCCGCCUAUCCGACCCUGGAGCUGGGUCUGGCGCGGCA GCCGCUUCUGCGGGUUCGAACGGCAACGCGCGACUAUGCCGGUCUGUAUGUCCUG CGCGUAUGGGUCGGCAGCGCGACGAACGCCAGCCUGUUUGUUUUGGGGGUGGCGC UCUCUGCCAACGGGACGUUUGUGUAUAACGGCUCGGACUACGGCUCCUGCGAUCC GGCGCAGCUUCCCUUUUCGGCCCCGCGCCUGGGACCCUCGAGCGUAUACACCCCC GGAGCCUCCCGGCCCACCCCUCCACGGACAACGACAUCCCCGUCCUCCCCCCGAGA CCCGACCCCCGCCCCCGGGGACACAGGGACGCCCGCGCCCGCGAGCGGCGAGAGAG CCCCGCCCAAUUCCACGCGAUCGGCCAGCGAAUCGAGACACAGGCUAACCGUAGC CCAGGUAAUCCAGAUCGCCAUACCGGCGUCCAUCAUCGCCUUUGUGUUUCUGGGC AGCUGUAUCUGCUUCAUCCAUAGAUGCCAGCGCCGAUACAGGCGCCCCCGCGGCC AGAUUUACAACCCCGGGGGCGUUUCCUGCGCGGUCAACGAGGCGGCCAUGGCCCG CCUCGGAGCCGAGCUGCGAUCCCACCCAAACACCCCCCCCAAACCCCGACGCCGUU CGUCGUCGUCCACGACCAUGCCUUCCCUAACGUCGAUAGCUGAGGAAUCGGAGCC AGGUCCAGUCGUGCUGCUGUCCGUCAGUCCUCGGCCCCGCAGUGGCCCGACGGCC CCCCAAGAGGUCUAG (SEQ ID NO: 105)
ICP0-2	Based on strain HG52 (inactivated by deletion of the nuclear localization signal and zinc-binding ring finger)	AUGGAACCCCGGCCCGGCACGAGCUCCCGGGCGGACCCCGGCCCCGAGCGGCCGCC GCGGCAGACCCCCGGCACGCAGCCCGCCGCCCCGCACGCCUGGGGGAUGCUCAACG ACAUGCAGUGGCUCGCCAGCAGCGACUCGGAGGAGGAGACCGAGGUGGGAAUCUC UGACGACGACCUUCACCGCGACUCCACCUCCGAGGCGGGCAGCACGGACACGGAG AUGUUCGAGGCGGGCCUGAUGGACGCGGCCACGCCCCCGGCCCGGCCCCCGGCCG AGCGCCAGGGCAGCCCCACGCCCGCCGACGCGCAGGGAUCCUGUGGGGGUGGGCC CGUGGGUGAGGAGGAAGCGGAAGCGGGAGGGGGGGGCGACGUGAACACCCCGGU GGCGUACCUGAUAGUGGGCGUGACCGCCAGCGGGUCGUUCAGCACCAUCCCGAUA GUGAACGACCCCCGGACCCGCGUGGAGGCCGAGGCGGCCGUGCGGGCCGGCACGG CCGUGGACUUUAUCUGGACGGGCAACCCGCGGACGGCCCCGCGCUCCCUGUCGCU GGGGGGACACACGGUCCGCGCCCUGUCGCCCACCCCCCCGUGGCCCGGCACGGACG ACGAGGACGAUGACCUGGCCGACGUGGACUACGUCCCGCCCGCCCCCCGAAGAGC GCCCCGGCGCGGGGGCGGCGGUGCGGGGGCGACCCGCGGAACCUCCCAGCCCGCC GCGACCCGACCGGCGCCCCCUGGCGCCCCGCGGAGCAGCAGCAGCGGCGGCGCCCC GUUGCGGGCGGGGGUGGGAUCUGGGUCUGGGGGCGGCCCUGCCGUCGCGGCCGUC GUGCCGAGAGUGGCCUCUCUUCCCCCUGCGGCCGGCGGGGGGCGCGCGCAGGCGC GGCGGGUGGGCGAAGACGCCGCGGCGGCGGAGGGCAGGACGCCCCCCGCGAGACA GCCCCGCGCGGCCCAGGAGCCCCCCAUAGUCAUCAGCGACUCUCCCCCCGCCGUCUC CGCGCCGCCCCGCGGGCCCCGGGCCGCUCUCCUUUGUCUCCUCCUCCUCCGCACAG GUGUCCUCGGGCCCCGGGGGGGGGAGGUCUGCCACAGUCGUCGGGGCGCGCCGCGC GCCCCCGCGCGGCCGUCGCCCCGCGCGUCCGGAGUCCGCCCCGCGCCGCCGCCGCC CCCGUGGUGUCUGCGAGCGCGGACGCGGCCGGGCCCGCGCCGCCCGCCGUGCCGG UGGACGCGCACCGCGCGCCCCGGUCGCGCAUGACCCAGGCUCAGACCGACACCCA AGCACAGAGUCUGGGCCGGGCAGGCGCGACCGACGCGCGCGGGUCGGGAGGGCCG GGCGCGGAGGGAGGAUCGGGCCCCGCGGCCUCGUCCUCCGCCUCUUCCUCCGCCG CCCCGCGCUCGCCCCUCGCCCCCCAGGGGGUGGGGGCCAAGAGGGCGGCGCCGCGC CGGGCCCCGGACUCGGACUCGGGCGACCGCGGCCACGGGCCGCUCGCCCCGGCGUC CGCGGGCGCCGCGCCCCCGUCGGCGUCUCCGUCGUCCCAGGCCGCGGUCGCCGCCG CCUCCUCCUCCUCCGCCUCCUCCUCCUCCGCCUCCUCCUCCUCCGCCUCCUCCUCC UCCGCCUCCUCCUCCUCCGCCUCCUCCUCCUCCGCCUCCUCCUCCUCCGCCUCUUC CUCUGCGGGCGGGGCUGGUGGGAGCGUCGCGUCCGCGUCCGGCGCUGGGGAGAGA CGAGAAACCUCCCUCGGCCCCCGCGCUGCUGCGCCGCGGGGGCCGAGGAAGUGUG CCAGGAAGACGCGCCACGCGGAGGGCGGCCCCGAGCCCGGGGCCCGCGACCCGGC GCCCGGCCUCACGCGCUACCUGCCCAUCGCGGGGGUCUCGAGCGUCGUGGCCCUG GCGCCUUACGUGAACAAGACGGUCACGGGGGACUGCCUGCCCGUCCUGGACAUGG

Strain	Nucleic Acid Sequence
	AGACGGGCCACAUAGGGGCCUACGUGGUCCUCGUGGACCAGACGGGGAACGUGGC GGACCUGCUGCGGGCCGCGGCCCCCGCGUGGAGCCGCCGCACCCUGCUCCCCGAGC ACGCGCGCAACUGCGUGAGGCCCCCCGACUACCCGACGCCCCCCGCGUCGGAGUG GAACAGCCUCUGGAUGACCCCGGUGGGCAACAUGCUCUUUGACCAGGGCACCCUG GUGGGCGCGCUGGACUUCCACGGCCUCCGGUCGCGCCACCCGUGGUCUCGGGAGC AGGGCGCGCCCGCGCCGGCCGGCGACGCCCCCGCGGGCCACGGGGAGUAG (SEQ ID NO: 106)
HSV-2 SgB	AUGCGCGGGGGGGGCUUGGUUUGCGCGCUGGUCGUGGGGGCGCUGGUGGCCGCGG UGGCGUCGGCGGCCCCGGCGGCCCCCCGCGCCUCGGGCGGCGUGGCCGCGACCGUC GCGGCGAACGGGGGUCCCGCCUCCCAGCCGCCCCCCGUCCCGAGCCCCGCGACCAC CAAGGCCCGGAAGCGGAAAACCAAAAAGCCGCCCAAGCGGCCCGAGGCGACCCCG CCCCCCGACGCCAACGCGACCGUCGCCGCCGGCCACGCCACGCUGCGCGCGCACCU GCGGGAAAUCAAGGUCGAGAACGCCGAUGCCCAGUUUUACGUGUGCCCGCCCCCG ACGGGCGCCACGGUGGUGCAGUUUGAGCAGCCGCGCCGCUGCCCGACGCGCCCGG AGGGGCAGAACUACACGGAGGGCAUCGCGGUGGUCUUCAAGGAGAACAUCGCCCC GUACAAAUUCAAGGCCACCAUGUACUACAAAGACGUGACCGUGUCGCAGGUGUGG UUCGGCCACCGCUACUCCCAGUUUAUGGGGAUAUUCGAGGACCGCGCCCCCGUUC CCUUCGAGGAGGUGAUCGACAAGAUUAACGCCAAGGGGGUCUGCCGCUCCACGGC CAAGUACGUGCGGAACAACAUGGAGACCACCGCGUUUCACCGGGACGACCACGAG ACCGACAUGGAGCUCAAGCCGGCGAAGGUCGCCACGCGCACGAGCCGGGGGUGGC ACACCACCGACCUCAAGUACAACCCCUCGCGGGUGGAGGCGUUCCAUCGGUACGG CACGACGGUCAACUGCAUCGUCGAGGAGGUGGACGCGCGGUCGGUGUACCCGUAC GAUGAGUUUGUGCUGGCGACGGGCGACUUUGUGUACAUGUCCCCGUUUUACGGCU ACCGGGAGGGGUCGCACACCGAGCACACCAGCUACGCCGCCGACCGCUUCAAGCA GGUCGACGGCUUCUACGCGCGCGACCUCACCACGAAGGCCCGGGCCACGUCGCCG ACGACCCGCAACUUGCUGACGACCCCCAAGUUUACCGUGGCCUGGGACUGGGUGC CGAAGCGACCGGCGGUCUGCACCAUGACCAAGUGGCAGGAGGUGGACGAGAUGCU CCGCGCCGAGUACGGCGGCUCCUUCCGCUUCUCCUCCGACGCCAUCUCGACCACCU UCACCACCAACCUGACCCAGUACUCGCUCUCGCGCGUCGACCUGGGCGACUGCAU CGGCCGGGAUGCCCGCGAGGCCAUCGACCGCAUGUUUGCGCGCAAGUACAACGCC ACGCACAUCAAGGUGGGCCAGCCGCAGUACUACCUGGCCACGGGGGGCUUCCUCA UCGCGUACCAGCCCCUCCUCAGCAACACGCUCGCCGAGCUGUACGUGCGGGAGUA CAUGCGGGAGCAGGACCGCAAGCCCCGGAAUGCCACGCCCGCGCCACUGCGGGAG GCGCCCAGCGCCAACGCGUCCGUGGAGCGCAUCAAGACCACCUCCUCGAUCGAGU UCGCCCGGCUGCAGUUUACGUAUAACCACAUACAGCGCCACGUGAACGACAUGCU GGGGCGCAUCGCCGUCGCGUGGUGCGAGCUGCAGAACCACGAGCUGACUCUCUGG AACGAGGCCCGCAAGCUCAACCCCAACGCCAUCGCCUCCGCCACCGUCGGCCGGCG GGUGAGCGCGCGCAUGCUCGGAGACGUCAUGGCCGUCUCCACGUGCGUGCCCGUC GCCCCGGACAACGUGAUCGUGCAGAACUCGAUGCGCGUCAGCUCGCGGCCGGGGA CGUGCUACAGCCGCCCCCUGGUCAGCUUUCGGUACGAAGACCAGGGCCCGCUGAU CGAGGGGCAGCUGGGCGAGAACAACGAGCUGCGCCUCACCCGCGACGCGCUCGAG CCGUGCACCGUGGGCCACCGGCGCUACUUCAUCUUCGGCGGGGGCUACGUGUACU UCGAGGAGUACGCGUACUCUCACCAGCUGAGUCGCGCCGACGUCACCACCGUCAG CACCUUCAUCGACCUGAACAUCACCAUGCUGGAGGACCACGAGUUUGUGCCCCUG GAGGUCUACACGCGCCACGAGAUCAAGGACAGCGGCCUGCUGGACUACACGGAGG UCCAGCGCCGCAACCAGCUGCACGACCUGCGCUUUGCCGACAUCGACACGGUCAU CCGCGCCGACGCCAACGCCGCCAUGUUCGCGGGGCUGUGCGCGUUCUUCGAGGGG AUGGGGGACUUGGGGCGCGCGGUCGGCAAGGUCGUCAUGGGAGUAGUGGGGGGC GUGGUGUCGGCCGUCUCGGGCGUGUCCUCCUUUAUGUCCAACCCC (SEQ ID NO: 107)
HSV-2 SgC	AUGGCCCUUGGACGGGUGGGCCUAGCCGUGGGCCUGUGGGGCCUGCUGUGGGUGG GUGUGGUCGUGGUGCUGGCCAAUGCCUCCCCCGGACGCACGAUAACGGUGGGCCC GCGGGGGAACGCGAGCAAUGCCGCCCCCUCCGCGUCCCCGCGGAACGCAUCCGCCC CCCGAACCACACCCACGCCCCCCCAACCCCGCAAGGCGACGAAAAGUAAGGCCUCC ACCGCCAAACCGGCCCCGCCCCCCAAGACCGGGCCCCCGAAGACAUCCUCGGAGCC CGUGCGAUGCAACCGCCACGACCCGCUGGCCCGGUACGGCUCGCGGGUGCAAAUC CGAUGCCGGUUUCCCAACUCCACCCGCACGGAGUCCCGCCUCCAGAUCUGGCGUU AUGCCACGGCGACGGACGCCGAGAUCGGAACGGCGCCUAGCUUAGAGGAGGUGAU GGUAAACGUGUCGGCCCCGCCCGGGGGCCAACUGGUGUAUGACAGCGCCCCCAAC

Strain	Nucleic Acid Sequence
	CGAACGGACCCGCACGUGAUCUGGGCGGAGGGCGCCGGCCCGGGCGCCAGCCCGC GGCUGUACUCGGUCGUCGGGCCGCUGGGUCGGCAGCGGCUCAUCAUCGAAGAGCU GACCCUGGAGACCCAGGGCAUGUACUACUGGGUGUGGGGCCGGACGGACCGCCCG UCCGCGUACGGGACCUGGGUGCGCGUUCGCGUGUUCCGCCCUCCGUCGCUGACCA UCCACCCCCACGCGGUGCUGGAGGGCCAGCCGUUUAAGGCGACGUGCACGGCCGC CACCUACUACCCGGGCAACCGCGCGGAGUUCGUCUGGUUCGAGGACGGUCGCCGG GUAUUCGAUCCGGCCCAGAUACACACGCAGACGCAGGAGAACCCCGACGGCUUUU CCACCGUCUCCACCGUGACCUCCGCGGCCGUCGGCGGCCAGGGCCCCCCGCGCACC UUCACCUGCCAGCUGACGUGGCACCGCGACUCCGUGUCGUUCUCUCGGCGCAACG CCAGCGGCACGGCAUCGGUGCUGCCGCGGCCAACCAUUACCAUGGAGUUUACGGG CGACCAUGCGGUCUGCACGGCCGGCUGUGUGCCCGAGGGGGUGACGUUUGCCUGG UUCCUGGGGGACGACUCCUCGCCGGCGGAGAAGGUGGCCGUCGCGUCCCAGACAU CGUGCGGGCGCCCCGGCACCGCCACGAUCCGCUCCACCCUGCCGGUCUCGUACGAG CAGACCGAGUACAUCUGCCGGCUGGCGGGAUACCCGGACGGAAUUCCGGUCCUAG AGCACCACGGCAGCCACCAGCCCCCGCCGCGGGACCCCACCGAGCGGCAGGUGAUC CGGGCGGUGGAGGGG (SEQ ID NO: 108)
HSV-2 SgD	AUGGGGCGUUUGACCUCCGGCGUCGGGACGGCGGCCCUGCUAGUUGUCGCGGUGG GACUCCGCGUCGUCUGCGCCAAAUACGCCUUAGCAGACCCCUCGCUUAAGAUGGC CGAUCCCAAUCGAUUUCGCGGGAAGAACCUUCCGGUUUUGGACCAGCUGACCGAC CCCCCCGGGGUGAAGCGUGUUUACCACAUUCAGCCGAGCCUGGAGGACCCGUUCC AGCCCCCCAGCAUCCCGAUCACUGUGUACUACGCAGUGCUGGAACGUGCCUGCCG CAGCGUGCUCCUACAUGCCCCAUCGGAGGCCCCCCAGAUCGUGCGCGGGGCUUCG GACGAGGCCCGAAAGCACACGUACAACCUGACCAUCGCCUGGUAUCGCAUGGGAG ACAAUUGCGCUAUCCCCAUCACGGUUAUGGAAUACACCGAGUGCCCCUACAACAA GUCGUUGGGGGUCUGCCCCAUCCGAACGCAGCCCCGCUGGAGCUACUAUGACAGC UUUAGCGCCGUCAGCGAGGAUAACCUGGGAUUCCUGAUGCACGCCCCCGCCUUCG AGACCGCGGGUACGUACCUGCGGCUAGUGAAGAUAAACGACUGGACGGAGAUCAC ACAAUUUAUCCUGGAGCACCGGGCCCGCGCCUCCUGCAAGUACGCUCUCCCCCUG CGCAUCCCCCCGGCAGCGUGCCUCACCUCGAAGGCCUACCAACAGGGCGUGACGG UCGACAGCAUCGGGAUGCUACCCCGCUUUAUCCCCGAAAACCAGCGCACCGUCGC CCUAUACAGCUUAAAAAUCGCCGGGUGGCACGGCCCCAAGCCCCCGUACACCAGC ACCCUGCUGCCGCCGGAGCUGUCCGACACCACCAACGCCACGCAACCCGAACUCGU UCCGGAAGACCCCGAGGACUCGGCCCUCUUAGAGGAUCCCGCCGGGACGGUGUCU UCGCAGAUCCCCCCAAACUGGCACAUCCCGUCGAUCCAGGACGUCGCGCCGCACC ACGCCCCCGCCGCCCCCAGCAACCCG (SEQ ID NO: 109)
HSV-2 SgE	AUGGCUCGCGGGGCCGGGUUGGUGUUUUUUGUUGGAGUUUGGGUCGUAUCGUGC CUGGCGGCAGCACCCAGAACGUCCUGGAAACGGGUAACCUCGGGCGAGGACGUGG UGUUGCUUCCGGCGCCCGCGGGGCCGGAGGAACGCACCCGGGCCCACAAACUACU GUGGGCCGCGGAACCCCUGGAUGCCUGCGGUCCCCUGCGCCCGUCGUGGGUGGCG CUGUGGCCCCCCCGACGGGUGCUCGAGACGGUCGUGGAUGCGGCGUGCAUGCGCG CCCCGGAACCGCUCGCCAUAGCAUACAGUCCCCCGUUCCCCGCGGGCGACGAGGG ACUGUAUUCGGAGUUGGCGUGGCGCGAUCGCGUAGCCGUGGUCAACGAGAGUCUG GUCAUCUACGGGGCCCUGGAGACGGACAGCGGUCUGUACACCCUGUCCGUGGUCG GCCUAAGCGACGAGGCGCGCCAAGUGGCGUCGGUGGUUCUGGUCGUGGAGCCCGC CCCUGUGCCGACCCCGACCCCCGACGACUACGACGAAGAAGACGACGCGGGCGUG AGCGAACGCACGCCGGUCAGCGUUCCCCCCCCCAACCCCCCCCCGUCGUCCCCCCGU CGCCCCCCCGACGCACCCUCGUGUUAUCCCCGAGGUGUCCCACGUGCGCGGGGUA ACGGUCCAUAUGGAGACCCCGGAGGCCAUUCUGUUUGCCCCCGGGGAGACGUUUG GGACGAACGUCUCCAUCCACGCCAUUGCCCACGACGACGGUCCGUACGCCAUGGA CGUCGUCUGGAUGCGGUUUGACGUGCCGUCCUCGUGCGCCGAGAUGCGGAUCUAC GAAGCUUGUCUGUAUCACCCGCAGCUUCCAGAGUGUCUAUCUCCGGCCGACGCGC CGUGCGCCGUAAGUUCCUGGGCGUACCGCCUGGCGGUCCGCAGCUACGCCGGCUG UUCCAGGACUACGCCCCCGCCGCGAUGUUUUGCCGAGGCUCGCAUGGAACCGGUC CCGGGGUUGGCGUGGCUGGCCUCCACCGUCAAUCUGGAAUUCCAGCACGCCUCCC CCCAGCACGCCGGCCUCUACCUGUGCGUGGUGUACGUGGACGAUCAUAUCCACGC CUGGGGCCACAUGACCAUCAGCACCGCGGCGCAGUACCGGAACGCGGUGGUGGAA CAGCACCUCCCCCAGCGCCAGCCCGAGCCCGUCGAGCCCACCCGCCCGCACGUGAG AGCCCCCCCUCCCGCGCCCUCCGCGCGCGGCCCGCUGCGC (SEQ ID NO: 110)
HSV-2 SgI	AUGCCCGGCCGCUCGCUGCAGGGCCUGGCGAUCCUGGGCCUGUGGGUCUGCGCCA

Strain	Nucleic Acid Sequence
	CCGGCCUGGUCGUCCGCGGCCCCACGGUCAGUCUGGUCUCAGACUCACUCGUGGA UGCCGGGGCCGUGGGGCCCCAGGGCUUCGUGGAAGAGGACCUGCGUGUUUUCGGG GAGCUUCAUUUUGUGGGGGCCCAGGUCCCCCACACAAACUACUACGACGGCAUCA UCGAGCUGUUUCACUACCCCCUGGGGAACCACUGCCCCCGCGUUGUACACGUGGU CACACUGACCGCAUGCCCCCGCCGCCCCGCCGUGGCGUUCACCUUGUGUCGCUCGA CGCACCACGCCCACAGCCCCGCCUAUCCGACCCUGGAGCUGGGUCUGGCGCGGCA GCCGCUUCUGCGGGUUCGAACGGCAACGCGCGACUAUGCCGGUCUGUAUGUCCUG CGCGUAUGGGUCGGCAGCGCGACGAACGCCAGCCUGUUUGUUUUGGGGGUGGCGC UCUCUGCCAACGGGACGUUUGUGUAUAACGGCUCGGACUACGGCUCCUGCGAUCC GGCGCAGCUUCCCUUUUCGGCCCCGCGCCUGGGACCCUCGAGCGUAUACACCCCC GGAGCCUCCCGGCCCACCCCUCCACGGACAACGACAUCCCCGUCCUCCCCCCGAGA CCCGACCCCCGCCCCCGGGGACACAGGGACGCCCGCGCCCGCGAGCGGCGAGAGAG CCCCGCCCAAUUCCACGCGAUCGGCCAGCGAAUCGAGACACAGGCUAACCGUAGC CCAGGUAAUCCAG (SEQ ID NO: 111)
HSV-2 ICP-4; Based on strain HG52; (inactivated by deletion of nuclear localization signal and alanine substitution for key residues in the transactivation region)	AUGUCGGCGGAGCAGCGGAAGAAGAAGAAGACGACGACGACGACGCAGGGCCGCG GGGCCGAGGUCGCGAUGGCGGACGAGGACGGGGGACGUCUCCGGGCCGCGGCGGA GACGACCGGCGGCCCCGGAUCUCCGGAUCCAGCCGACGGACCGCCGCCCACCCCGA ACCCGGACCGUCGCCCCGCCGCGCGGCCCGGGUUCGGGUGGCACGGUGGGCCGGA GGAGAACGAAGACGAGGCCGACGACGCCGCCGCCGAUGCCGAUGCCGACGAGGCG GCCCCGGCGUCCGGGGAGGCCGUCGACGAGCCUGCCGCGGACGGCGUCGUCUCGC CGCGGCAGCUGGCCCUGCUGGCCUCGAUGGUGGACGAGGCCGUUCGCACGAUCCC GUCGCCCCCCCCCGGAGCGCGACGGCGCGCAAGAAGAAGCGGCCCGCUCGCCUUCU CCGCCGCGGACCCCCUCCAUGCGCGCCGAUUAUGGCGAGGAGAACGACGACGACG ACGACGACGACGAUGACGACGACCGCGACGCGGGCCGCUGGGUCCGCGGACCGGA GACGACGUCCGCGGUCCGCGGGGCGUACCCGGACCCCAUGGCCAGCCUGUCGCCG CGACCCCCGGCGCCCCGCCGACACCACCACCACCACCACCACCGCCGCCGGCGCGC CCCCCGCCGGCGCUCGGCCGCCUCUGACUCAUCAAAAUCCGGAUCCUCGUCGUCG GCGUCCUCCGCCUCCUCCUCCGCCUCCUCCUCCUCGUCUGCAUCCGCCUCCUCGUC UGACGACGACGACGACGACGCCGCCCGCGCCCCCGCCAGCGCCGCAGACCACG CCGCGGGCGGGACCCUCGGCGCGGACGACGAGGAGGCGGGGGUGCCCGCGAGGGC CCCGGGGGCGGCGCCCCGGCCGAGCCCGCCCAGGGCCGAGCCCGCCCCGGCCCGGA CCCCCGCGGCGACCGCGGGCCGCCUGGAGCGCCGCCGGGCCCGCGCGGCGGUGGCC GGCCGCGACGCCACGGGCCGCUUCACGGCCGGGCGGCCCCGGCGGGUCGAGCUGG ACGCCGACGCGGCCUCCGGCGCCUUCUACGCGCGCUACCGCGACGGGUACGUCAG CGGGGAGCCGUGGCCCGGGGCCGGCCCCCCGCCCCCGGGGCGCGUGCUGUACGGC GGGCUGGGCGACAGCCGCCCCGGCCUCUGGGGGGCGCCCGAGGCGGAGGAGGCGC GGGCCCGGUUCGAGGCCUCGGGCGCCCCGGCGCCCGUGUGGGCGCCCGAGCUGGG CGACGCGGCGCAGCAGUACGCCCUGAUCACGCGGCUGCUGUACACGCCGGACGCG GAGGCGAUGGGGUGGCUCCAGAACCCGCGCGUGGCGCCCGGGGACGUGGCGCUGG ACCAGGCCUGCUUCCGGAUCUCGGGCGCGGCGCGCAACAGCAGCUCCUUCAUCUC CGGCAGCGUGGCGCGGGCCGUGCCCCACCUGGGGUACGCCAUGGCGGCGGGCCGC UUCGGCUGGGGCCUGGCGCACGUGGCGGCCGCCGUGGCCAUGAGCCGCCGCUACG ACCGCGCGCAGAAGGGCUUCCUGCUGACCAGCCUGCGCCGCGCCUACGCGCCCCU GCUGGCGCGCGAGAACGCGGCGCUGACCGGGGCGCGAACCCCCGACGACGGCGGC GACGCCAACCGCCACGACGGCGACGACGCCCGCGGGAAGCCCGCCGCCGCCGCCGC CCCGUUGCCGUCGGCGGCGGCGUCGCCGGCCGACGAGCGCGCGGUGCCCGCCGGC UACGGCGCCGCGGGGGUGCUCGCCGCCCUGGGGCGCCUGAGCGCCGCGCCCGCCU CCGCGCCGGCCGGGGCCGACGACGACGACGACGACGACGGCGCCGGCGGUGGUGG CGGCGGCCGGCGCGCGGAGGCGGGCCGCGUGGCCGUGGAGUGCCUGGCCGCCUGC CGCGGGAUCCUGGAGGCGCUGGCGGAGGGCUUCGACGGCGACCUGGCGGCCGUGC CGGGGCUGGCCGGAGCCCGGCCCGCCGCGCCCCCGCGCCCGGGGCCCGCGGGCGCG GCCGCCCCGCCGCACGCCGACGCGCCCCGCCUGCGCGCCUGGCUGCGCGAGCUGCG GUUCGUGCGCGACGCGCUGGUGCUGAUGCGCCUGCGCGGGGACCUGCGCGUGGCC GGCGGCAGCGAGGCCGCCGUGGCCGCCGUGCGCGCCGUGAGCCUGGUCGCCGGGG CCCUGGGCCCGGCGCUGCCGCGGAGCCCGCGCCUGCUGAGCUCCGCCGCCGCCGCC GCCGCGGACCUGCUCUUCCAGAACCAGAGCCUGCGCCCCCUGCUGGCCGACACCG UCGCCGCGGCCGACUCGCUCGCCGCGCCCGCCUCCGCGCCGCGGGAGGCCGCGGAC GCCCCCCGCCCCGCGGCCGCCCCUCCCGCGGGGGCCGCGCCCCCCGCCCCGCCGAC GCCGCCGCCGCGGCCGCCGCGCCCCGCGGCGCUGACCCGCCGGCCCGCCGAGGGCC

Strain	Nucleic Acid Sequence
	CCGACCCGCAGGGCGGCUGGCGCCGCCAGCCGCCGGGGCCCAGCCACACGCCGGCG CCCUCGGCCGCCGCCCUGGAGGCCUACUGCGCCCCGCGGGCCGUGGCCGAGCUCAC GGACCACCCGCUCUUCCCCGCGCCGUGGCGCCCGGCCCUCAUGUUCGACCCGCGCG CGCUGGCCUCGCUGGCCGCGCGCUGCGCCGCCCCGCCCCCCCGGCGGCGCGCCCGCC GCCUUCGGCCCGCUGCGCGCCUCGGGCCCGCUGCGCCGCGCGGCGGCCUGGAUGC GCCAGGUGCCCGACCCGGAGGACGUGCGCGUGGUGAUCCUCUACUCGCCGCUGCC GGGCGAGGACCUGGCCGCGGGCCGCGCCGGGGGCGGGCCCCCCCCGGAGUGGUCC GCCGAGCGCGGCGGGCUGUCCUGCCUGCUGGCGGCCCUGGGCAACCGGCUCUGCG GGCCCGCCACGGCCGCCUGGGCGGGCAACUGGACCGGCGCCCCCGACGUCUCGGC GCUGGGCGCGCAGGGCGUGCUGCUGCUGUCCACGCGGGACCUGGCCUUCGCCGGC GCCGUGGAGUUCCUGGGGCUGCUGGCCGGCGCCUGCGACCGCCGCCUCAUCGUCG UCAACGCCGUGCGCGCCGCGGCCUGGCCCGCCGCUGCCCCCGUGGUCUCGCGGCAG CACGCCUACCUGGCCUGCGAGGUGCUGCCCGCCGUGCAGUGCGCCGUGCGCUGGC CGGCGGCGCGGGACCUGCGCCGCACCGUGCUGGCCUCCGGCCGCGUGUUCGGGCC GGGGGUCUUCGCGCGCGUGGAGGCCGCGCACGCGCGCCUGUACCCCGACGCGCCG CCGCUGCGCCUCUGCCGCGGGGCCAACGUGCGGUACCGCGUGCGCACGCGCUUCG GCCCCGACACGCUGGUGCCCAUGUCCCCGCGCGAGUACCGCCGCGCCGUGCUCCCG GCGCUGGACGGCCGGGCCGCCGCCUCGGGCGCGGGCGACGCCAUGGCGCCCGGCG CGCCGGACUUCUGCGAGGACGAGGCGCACUCGCACCGCGCCUGCGCGCGCUGGGG CCUGGGCGCGCCGCUGCGGCCCGUCUACGUGGCGCUGGGGCGCGACGCCGUGCGC GGCGGCCCGGCGGAGCUGCGCGGGCCGCGGCGGGAGUUCUGCGCGCGGGCGCUGC UCGAGCCCGACGGCGACGCGCCCCCGCUGGUGCUGCGCGACGACGCGGACGCGGG CCCGCCCCCGCAGAUACGCUGGGCGUCGGCCGCGGGCCGCGCGGGGACGGUGCUG GCCGCGGCGGGCGGCGGCGUGGAGGUGGUGGGGACCGCCGCGGGGCUGGCCACGC CGCCGAGGCGCGAGCCCGUGGACAUGGACGCGGAGCUGGAGGACGACGACGACGG ACUGUUUGGGGAGUGA (SEQ ID NO: 112)
MRK_HSV-2 gB, SQ-032178, CX-000747	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGAGAGGUGGUGGCUU AGUUUGCGCGCUGGUUGUCGGGGCGCUCGUAGCCGCCGUGGCGUCGGCCGCCCCU GCGGCUCCUCGCGCUAGCGGAGGCGUAGCCGCAACAGUUGCGGCGAACGGGGGUC CAGCCUCUCAGCCUCCUCCCGUCCCGAGCCCUGCGACCACCAAGGCUAGAAAGCG GAAGACCAAGAAACCGCCCAAGCGCCCCGAGGCCACCCCGCCCCCCGAUGCCAACG CGACUGUCGCCGCUGGCCAUGCGACGCUUCGCGCUCAUCUGAGGGAGAUCAAGGU UGAAAAUGCUGAUGCCCAAUUUUACGUGUGCCCGCCCCCGACGGGCGCCACGGUU GUGCAGUUUGAACAGCCGCGGCGCUGUCCGACGCGGCCAGAAGGCCAGAACUAUA CGGAGGGCAUAGCGGUGGUCUUUAAGGAAAACAUCGCCCCGUACAAAUUUAAGGC CACAAUGUACUACAAAGACGUGACAGUUUCGCAAGUGUGGUUUGGCCACAGAUAC UCGCAGUUUAUGGGAAUCUUCGAAGAUAGAGCCCCUGUUCCCUUCGAGGAAGUCA UCGACAAGAUUAAUGCCAAAGGGGUAUGCCGUUCCACGGCCAAAUACGUGCGCAA CAAUAUGGAGACCACCGCCUUUCACCGGGAUGAUCACGAGACCGACAUGGAGCUU AAGCCGGCGAAGGUCGCCACGCGUACCUCCCGGGGUUGGCACACCACAGAUCUUA AGUACAAUCCCUCGCGAGUUGAAGCAUUCCAUCGGUAUGGAACUACCGUUAACUG CAUCGUUGAGGAGGUGGAUGCGCGGUCGGUGUACCCUUACGAUGAGUUUGUGUU AGCGACCGGCGAUUUUGUGUACAUGUCCCCGUUUUACGGCUACCGGGAGGGGUCG CACACCGAACAUACCUCGUACGCCGCUGACAGGUUCAAGCAGGUCGAUGGCUUUU ACGCGCGCGAUCUCACCACGAAGGCCCGGGCCACGUCACCGACGACCAGGAACUU GCUCACGACCCCCAAGUUCACCGUCGCUUGGGAUUGGGUCCCAAAGCGUCCGGCG GUCUGCACGAUGACCAAAUGGCAGGAGGUGGACGAAAUGCUCCGCGCAGAAUACG GCGGCUCCUUCCGCUUCUCGUCCGACGCCAUCUCGACAACCUUCACCACCAAUCU GACCCAGUACAGUCUGUCGCGCGUUGAUUUAGGAGACUGCAUUGGCCGGGAUGCC CGGGAGGCCAUCGACAGAAUGUUUGCGCGUAAGUACAAUGCCACACAUAUUAAGG UGGGCCAGCCGCAAUACUACCUUGCCACGGGCGGCUUUCUCAUCGCGUACCAGCC CCUUCUCUCAAAUACGCUCGCUGAACUGUACGUGCGGGAGUAUAUGAGGGAACAG GACCGCAAGCCCCGCAAUGCCACGCCUGCGCCACUACGAGAGGCGCCUUCAGCUA AUGCGUCGGUGGAACGUAUCAAGACCACCUCCUCAAUAGAGUUCGCCCGGCUGCA AUUUACGUACAACCACAUCCAGCGCCACGUGAACGACAUGCUGGGCCGCAUCGCU GUCGCCUGGUGCGAGCUGCAGAAUCACGAGCUGACUCUUUGGAACGAGGCCCGAA AACUCAACCCCAACGCGAUCGCCUCCGCAACAGUCGGUAGACGGGUGAGCGCUCG CAUGCUAGGAGAUGUCAUGGCUGUGUCCACCUGCGUGCCCGUCGCUCCGGACAAC

Strain	Nucleic Acid Sequence
	GUGAUUGUGCAGAAUUCGAUGCGGGUCUCAUCGCGGCCGGGCACCUGCUACAGCA GGCCCCUCGUCAGCUUCCGGUACGAAGACCAGGGCCCGCUGAUUGAAGGGCAACU GGGAGAGAACAAUGAGCUGCGCCUCACCCGCGACGCGCUCGAACCCUGCACCGUC GGACAUCGGAGAUAUUUCAUCUUCGGAGGGGGCUACGUGUACUUCGAAGAGUAU GCCUACUCUCACCAGCUGAGUAGAGCCGACGUCACUACCGUCAGCACCUUUAUUG ACCUGAAUAUCACCAUGCUGGAGGACCACGAGUUUGUGCCCCUGGAAGUUUACAC UCGCCACGAAAUCAAAGACUCCGGCCUGUUGGAUUACACGGAGGUUCAGAGGCGG AACCAGCUGCAUGACCUGCGCUUUGCCGACAUCGACACCGUCAUCCGCGCCGAUG CCAACGCUGCCAUGUUCGCGGGGCUGUGCGCGUUCUUCGAGGGGAUGGGUGACUU GGGGCGCGCCGUCGGCAAGGUCGUCAUGGGAGUAGUGGGGGGCGUUGUGAGUGC CGUCAGCGGCGUGUCCUCCUUCAUGUCCAAUCCAUUCGGAGCGCUUGCUGUGGGG CUGCUGGUCCUGGCCGGGCUGGUAGCCGCCUUCUUCGCCUUUCGAUAUGUUCUGC AACUGCAACGCAAUCCCAUGAAAGCUCUAUAUCCGCUCACCACCAAGGAGCUAAA GACGUCAGAUCCAGGAGGCGUGGGCGGGGAAGGGGAAGAGGGCGCGGAGGGCGG AGGGUUUGACGAAGCCAAAUUGGCCGAGGCUCGUGAAAUGAUCCGAUAUAUGGC ACUAGUGUCGGCGAUGGAAAGGACCGAACAUAAGGCCCGAAAGAAGGGCACGUCG GCGCUGCUCUCAUCCAAGGUCACCAACAUGGUACUGCGCAAGCGCAACAAAGCCA GGUACUCUCCGCUCCAUAACGAGGACGAGGCGGGAGAUGAGGAUGAGCUCUAAUG AUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAG CCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGU GGGCGGC (SEQ ID NO: 113)
MRK_HSV-2 gC, SQ-032179, CX-000670	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGCCCUUGGACGGGU AGGCCUAGCCGUGGGCCUGUGGGGCCUACUGUGGGUGGGUGUGGUCGUGGUGCU GGCCAAUGCCUCCCCCGGACGCACGAUAACGGUGGGCCCGCGAGGCAACGCGAGC AAUGCUGCCCCCUCCGCGUCCCCGCGGAACGCAUCCGCCCCCCGAACCACACCCAC GCCCCCACAACCCCGCAAAGCGACGAAAUCCAAGGCCUCCACCGCCAAACCGGCUC CGCCCCCCAAGACCGGACCCCCGAAGACAUCCUCGGAGCCCGUGCGAUGCAACCGC CACGACCCGCUGGCCCGGUACGGCUCGCGGGUGCAAAUCCGAUGCCGGUUUCCCA ACUCCACGAGGACUGAGUCCCGUCUCCAGAUCUGGCGUUAUGCCACGGCGACGGA CGCCGAAAUCGGAACAGCGCCUAGCUUAGAAGAGGUGAUGGUGAACGUGUCGGCC CCGCCCGGGGGCCAACUGGUGUAUGACAGUGCCCCCAACCGAACGGACCCGCAUG UAAUCUGGGCGGAGGGCGCCGGCCCGGGCGCCAGCCCGCGCCUGUACUCGGUUGU CGGCCCGCUGGGUCGGCAGCGGCUCAUCAUCGAAGAGUUAACCCUGGAGACACAG GGCAUGUACUAUUGGGUGUGGGGCCGGACGGACCGCCCGUCCGCCUACGGGACCU GGGUCCGCGUUCGAGUAUUUCGCCCUCCGUCGCUGACCAUCCACCCCCACGCGGU GCUGGAGGGCCAGCCGUUUAAGGCGACGUGCACGGCCGCAACCUACUACCCGGGC AACCGCGCGGAGUUCGUCUGGUUUGAGGACGGUCGCCGCGUAUUCGAUCCGGCAC AGAUACACACGCAGACGCAGGAGAACCCCGACGGCUUUUCCACCGUCUCCACCGU GACCUCCGCGGCCGUCGGCGGGCAGGGCCCCCCUCGCACCUUCACCUGCCAGCUGA CGUGGCACCGCGACUCCGUGUCGUUCUCUCGGCGCAACGCCAGCGGCACGGCCUC GGUUCUGCCGCGGCCGACCAUUACCAUGGAGUUUACAGGCGACCAUGCGGUCUGC ACGGCCGGCUGUGUGCCCGAGGGGGUCACGUUUGCUUGGUUCCUGGGGGAUGACU CCUCGCCGGCGGAAAAGGUGGCCGUCGCGUCCCAGACAUCGUGCGGGCGCCCCGG CACCGCCACGAUCCGCUCCACCCUGCCGGUCUCGUACGAGCAGACCGAGUACAUC UGUAGACUGGCGGGAUACCCGGACGGAAUUCCGGUCCUAGAGCACCACGGAAGCC ACCAGCCCCCGCCGCGGGACCCAACCGAGCGGCAGGUGAUCCGGGCGGUGGAGGG GGCGGGGAUCGGAGUGGCUGUCCUUGUCGCGGUGGUUCUGGCCGGGACCGCGGUA GUGUACCUGACCCAUGCCUCCUCGGUACGCUAUCGUCGGCUGCGGUAAUGAUAAU AGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCU CCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGGCG GC (SEQ ID NO: 114)
MRK_HSV-2 gD, SQ-032180, CX-001301	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGGGCGUUUGACCUC CGGCGUCGGGACGGCGGCCCUGCUAGUUGUCGCGGUGGGACUCCGCGUCGUCUGC GCCAAAUACGCCUUAGCAGACCCCUCGCUUAAGAUGGCCGAUCCCAAUCGAUUUC GCGGGAAGAACCUUCCGGUUUUGGACCAGCUGACCGACCCCCCCGGGGUGAAGCG UGUUUACCACAUUCAGCCGAGCCUGGAGGACCCGUUCCAGCCCCCCAGCAUCCCG AUCACUGUGUACUACGCAGUGCUGGAACGUGCCUGCCGCAGCGUGCUCCUACAUG

Strain	Nucleic Acid Sequence
	CCCCAUCGGAGGCCCCCCAGAUCGUGCGCGGGGCUUCGGACGAGGCCCGAAAGCA CACGUACAACCUGACCAUCGCCUGGUAUCGCAUGGGAGACAAUUGCGCUAUCCCC AUCACGGUUAUGGAAUACACCGAGUGCCCCUACAACAAGUCGUUGGGGGUCUGCC CCAUCCGAACGCAGCCCCGCUGGAGCUACUAUGACAGCUUUAGCGCCGUCAGCGA GGAUAACCUGGGAUUCCUGAUGCACGCCCCCGCCUUCGAGACCGCGGGUACGUAC CUGCGGCUAGUGAAGAUAAACGACUGGACGGAGAUCACACAAUUUAUCCUGGAGC ACCGGGCCCGCGCCUCCUGCAAGUACGCUCUCCCCCCUGCGCAUCCCCCCGGCAGCG UGCCUCACCUCGAAGGCCUACCAACAGGGCGUGACGGUCGACAGCAUCGGGAUGC UACCCCGCUUUAUCCCCGAAAACCAGCGCACCGUCGCCCUAUACAGCUUAAAAAU CGCCGGGUGGCACGGCCCCAAGCCCCCGUACACCAGCACCCUGCUGCCGCCGGAGC UGUCCGACACCACCAACGCCACGCAACCCGAACUCGUUCCGGAAGACCCCGAGGA CUCGGCCCUCUUAGAGGAUCCCGCCGGGACGGUGUCUUCGCAGAUCCCCCCAAAC UGGCACAUCCCGUCGAUCCAGGACGUCGCACCGCACCACGCCCCCGCCGCCCCCAG CAACCCGGGCCUGAUCAUCGGCGCGCUGGCCGGCAGUACCCUGGCGGUGCUGGUC AUCGGCGGUAUUGCGUUUUGGGUACGCCGCCGCGCUCAGAUGGCCCCCAAGCGCC UACGUCUCCCCCCACAUCCGGGAUGACGACGCGCCCCCCUCGCACCAGCCAUUGUU UUACUAGUGAUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCC UCCCCCCAGCCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAA GUCUGAGUGGGCGGC (SEQ ID NO: 115)
MRK_HSV-2 gE, SQ-032181, CX-001391	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGCUAGGGGGGCCGG GUUGGUUUUUUUUGUUGGAGUUUGGGUCGUAAGCUGCCUCGCGGCAGCGCCCAG AACGUCCUGGAAACGCGUAACCUCGGGCGAAGACGUGGUGUUACUCCCCGCGCCG GCGGGGCCGGAAGAACGCACUCGGGCCCACAAACUACUGUGGGCAGCGGAACCGC UGGAUGCCUGCGGUCCCCUGAGGCCGUCAUGGGUGGCACUGUGGCCCCCCCGACG AGUGCUUGAGACGGUUGUCGAUGCGGCGUGCAUGCGCGCCCCGGAACCGCUCGCU AUCGCAUACAGUCCCCCGUUCCCUGCGGGCGACGAGGGACUUUAUUCGGAGUUGG CGUGGCGCGAUCGCGUAGCCGUGGUCAACGAGAGUUUAGUUAUCUACGGGGCCCU GGAGACGGACAGUGGUCUGUACACCCUGUCAGUGGUGGGCCUAUCCGACGAGGCC CGCCAAGUGGCGUCCGUGGUUCUCGUCGUCGAGCCCGCCCCUGUGCCUACCCCGA CCCCCGAUGACUACGACGAGGAGGAUGACGCGGGCGUGAGCGAACGCACGCCCGU CAGCGUUCCCCCCCCCAACACCCCCCCGACGUCCCCCCGUCGCCCCCCCGACGCACC CUCGUGUUAUCCCUGAGGUGAGCCACGUGCGGGGGGUGACGGUCCACAUGGAAAC CCCGGAGGCCAUUCUGUUUGCGCCAGGGGAGACGUUUGGGACGAACGUCUCCAUC CACGCAAUUGCCCACGACGACGGUCCGUACGCCAUGGACGUCGUCUGGAUGCGAU UUGAUGUCCCGUCCUCGUGCGCCGAGAUGCGGAUCUAUGAAGCAUGUCUGUAUCA CCCGCAGCUGCCUGAGUGUCUGUCUCCGGCCGAUGCGCCGUGCGCCGUAAGUUCG UGGGCGUACCGCCUGGCGGUCCGCAGCUACGCCGGCUGCUCCAGGACUACGCCCC CACCUCGAUGUUUUGCUGAAGCUCGCAUGGAACCGGUCCCCGGGUUGGCGUGGCU CGCAUCAACUGUUAAUCUGGAAUUCCAGCAUGCCUCUCCCCAACACGCCGGCCUC UAUCUGUGUGUGGUGUAUGUGGACGACCAUAUCCAUGCCUGGGGCCACAUGACCA UCUCCACAGCGGCCCAGUACCGGAAUGCGGUGGUGGAACAGCAUCUCCCCCAGCG CCAGCCCGAGCCCGUAGAACCCACCCGACCGCAUGUGAGAGCCCCCCCUCCCGCAC CCUCCGCGAGAGGCCCGUUACGCUUAGGUGCGGUCCUGGGGGCGGCCCUGUUGCU CGCGGCCCUCGGGCUAUCCGCCUGGGCGUGCAUGACCUGCUGGCGCAGGCGCAGU UGGCGGGCGGUUAAAAGUCGGGCCUCGGCGACCGGCCCCACUUACAUUCGAGUAG CGGAUAGCGAGCUGUACGCGGACUGGAGUUCGGACUCAGAGGGCGAGCGCGACGG UUCCCUGUGGCAGGACCCUCCGGAGAGACCCGACUCACCGUCCACAAAUGGAUCC GGCUUUGAGAUCUUAUCCCCAACGGCGCCCUCUGUAUACCCCCAUAGCGAAGGGC GUAAAUCGCGCCGCCCGCUCACCACCUUUGGUUCAGGAAGCCCGGGACGUCGUCA CUCCCAGGCGUCCUAUUCUUCCGUCUUAUGGUAAUGAUAAUAGGCUGGAGCCUCG GUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCUCCUCCCCUUCCUGCA CCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGGCGGC (SEQ ID NO: 116)
MRK_HSV-2 gI, SQ-032182, CX-000645	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGCCCGGCCGCUCGCUG CAGGGCCUGGCGAUCCUGGGCCUGUGGGUCUGCGCCACCGGCCUGGUCGUCCGCG GCCCCACGGUCAGUCUGGUCUCAGACUCACUCGUGGAUGCCGGGGCCGUGGGGCC CCAGGGCUUCGUGGAAGAGGACCUGCGUGUUUUCGGGGAGCUUCAUUUUGUGGG GGCCCAGGUCCCCCACACAAACUACUACGACGGCAUCAUCGAGCUGUUUCACUAC

Strain	Nucleic Acid Sequence
	CCCCUGGGGAACCACUGCCCCCGCGUUGUACACGUGGUCACACUGACCGCAUGCC CCCGCCGCCCCCGCCGUGGCGUUCACCUUGUGUCGCUCGACGCACCACGCCCACAGC CCCGCCUAUCCGACCCUGGAGCUGGGUCUGGCGCGGCAGCCGCUUCUGCGGGUUC GAACGGCAACGCGCGACUAUGCCGGUCUGUAUGUCCUGCGCGUAUGGGUCGGCAG CGCGACGAACGCCAGCCUGUUUGUUUUGGGGGUGGCGCUCUCUGCCAACGGGACG UUUGUGUAUAACGGCUCGGACUACGGCUCCUGCGAUCCGGCGCAGCUUCCCUUUU CGGCCCCGCGCCUGGGACCCUCGAGCGUAUACACCCCCGGAGCCUCCCGGCCCACC CCUCCACGGACAACGACAUCACCGUCCUCCCCACGAGACCCGACCCCCGCCCCCGG GGACACAGGGACGCCUGCUCCCGCGAGCGGCGAGAGAGCCCCGCCCAAUUCCACG CGAUCGGCCAGCGAAUCGAGACACAGGCUAACCGUAGCCCAGGUAAUCCAGAUCG CCAUACCGGCGUCCAUCAUCGCCUUUGUGUUUCUGGGCAGCUGUAUCUGCUUCAU CCAUAGAUGCCAGCGCCGAUACAGGCGCCCCCGCGGCCAGAUUUACAACCCCGGG GGCGUUUCCUGCGCGGUCAACGAGGCGGCCAUGGCCCGCCUCGGAGCCGAGCUGC GAUCCCACCCAAACACCCCCCCCAAACCCCGACGCCGUUCGUCGUCGUCCACGACC AUGCCUUCCCUAACGUCGAUAGCUGAGGAAUCGGAGCCAGGUCCAGUCGUGCUGC UGUCCGUCAGUCCUCGGCCCCGCAGUGGCCCGACGGCCCCCCAAGAGGUCUAGUG AUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAG CCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGU GGGCGGC (SEQ ID NO: 117)
MRK_HSV-2 SgB, SQ-032210, CX-000655	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGCGCGGGGGGGGCUU AGUUUGCGCGCUGGUCGUGGGGGCGCUCGUAGCCGCGGUCGCGUCGGCGGCUCCG GCUGCCCCACGCGCUUCAGGUGGUGUCGCUGCGACCGUUGCGGCGAAUGGUGGUC CCGCCAGCCAACCGCCUCCCGUCCCGAGCCCCGCGACCACUAAGGCCCGGAAGCGG AAGACCAAGAAGCCACCCAAGCGGCCCGAGGCGACUCCGCCCCCAGACGCCAACG CGACCGUCGCCGCCGGCCACGCCACUCUGCGUGCGCACCUGCGGGAAAUCAAGGU CGAGAACGCGGACGCCCAGUUUUACGUGUGCCCGCCGCCGACUGGCGCCACGGUG GUGCAGUUUGAGCAACCUAGGCGCUGCCCGACGCGACCAGAGGGGCAGAACUACA CCGAGGGCAUAGCGGUGGUCUUUAAGGAAAACAUCGCCCCGUACAAAUUCAAGGC CACCAUGUACUACAAAGACGUGACCGUGUCGCAGGUGUGGUUCGGCCACCGCUAC UCCCAGUUUAUGGGGAUAUUCGAGGACCGCGCCCCCGUUCCCUUCGAAGAGGUGA UUGACAAAAUUAACGCCAAGGGGGUCUGCCGCAGUACGGCGAAGUACGUCCGGAA CAACAUGGAGACCACUGCCUUCCACCGGGACGACCACGAAACAGACAUGGAGCUC AAACCGGCGAAAGUCGCCACGCGCACGAGCCGGGGGUGGCACACCACCGACCUCA AAUACAAUCCUUCGCGGGUGGAAGCAUUCCAUCGGUAUGGCACGACCGUCAACUG UAUCGUAGAGGAGGUGGAUGCGCGGUCGGUGUACCCCUACGAUGAGUUCGUGCU GGCAACGGGCGAUUUUGUGUACAUGUCCCCUUUUUACGGCUACCGGGAAGGUAGU CACACCGAGCACACCAGUUACGCCGCCGACCGCUUUAAGCAAGUGGACGGCUUCU ACGCGCGCGACCUCACCACAAAGGCCCGGGCCACGUCGCCGACGACCCGCAAUUU GCUGACGACCCCCAAGUUUACCGUGGCCUGGGACUGGGUGCCUAAGCGACCGGCG GUCUGUACCAUGACAAAGUGGCAGGAGGUGGACGAAAUGCUCCGCGCUGAAUACG GUGGCUCUUUCCGCUUCUCUUCCGACGCCAUCUCCACCACGUUCACCACCAACCU GACCCAAUACUCGCUCUCGAGAGUCGAUCUGGGAGACUGCAUUGGCCGGGAUGCC CGCGAGGCAAUUGACCGCAUGUUCGCGCGCAAGUACAACGCUACGCACAUAAAGG UUGGCCAACCCCAGUACUACCUAGCCACGGGGGGCUUCCUCAUCGCUUAUCAACC CCUCCUCAGCAACACGCUCGCCGAGCUGUACGUGCGGGAAUAUAUGCGGGAACAG GACCGCAAACCCCGAAACGCCACGCCCGCGCCGCUGCGGGAAGCACCGAGCGCCA ACGCGUCCGUGGAGCGCAUCAAGACGACAUCCUCGAUUGAGUUUGCUCGUCUGCA GUUUACGUAUAACCACAUACAGCGCCAUGUAAACGACAUGCUCGGGCGCAUCGCC GUCGCGUGGUGCGAGCUCCAAAAUCACGAGCUCACUCUGUGGAACGAGGCACGCA AGCUCAAUCCCAACGCCAUCGCAUCCGCCACCGUAGGCCGGCGGGUGAGCGCUCG CAUGCUCGGGGAUGUCAUGGCCGUCUCCACGUGCGUGCCCGUCGCCCCGGACAAC GUGAUCGUGCAAAAUAGCAUGCGCGUUUCUUCGCGGCCGGGGACGUGCUACAGCC GCCCGCUGGUUAGCUUUCGGUACGAAGACCAAGGCCCGCUGAUUGAGGGGCAGCU GGGUGAGAACAACGAGCUGCGCCUCACCCGCGAUGCGUUAGAGCCGUGUACCGUC GGCCACCGGCGCUACUUCAUCUUCGGAGGGGGAUACGUAUACUUCGAAGAAUAUG CGUACUCUCACCAAUUGAGUCGCGCCGAUGUCACCACUGUUAGCACCUUCAUCGA CCUGAACAUCACCAUGCUGGAGGACCACGAGUUCGUGCCCCUGGAGGUCUACACA CGCCACGAGAUCAAGGAUUCCGGCCUACUGGACUACACCGAAGUCCAGAGACGAA

Strain	Nucleic Acid Sequence
	AUCAGCUGCACGAUCUCCGCUUUGCUGACAUCGAUACUGUUAUCCGCGCCGACGC CAACGCCGCCAUGUUCGCAGGUCUGUGUGCGUUUUUCGAGGGUAUGGGUGACUUA GGGCGCGCGGUGGGCAAGGUCGUCAUGGGGGUAGUCGGGGGCGUGGUGUCGGCC GUCUCGGGCGUCUCCUCCUUUAUGUCUAACCCCUGAUAAUAGGCUGGAGCCUCGG UGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCUCCUCCCCUUCCUGCAC CCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGGCGGC (SEQ ID NO: 118)
MRK_HSV-2 SgC, SQ-032835, CX-000616	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGCACUGGGAAGAGU GGGAUUGGCCGUCGGACUGUGGGGACUGCUGUGGGUGGGAGUCGUCGUCGUCCU GGCUAACGCCUCACCCGGUCGGACUAUCACUGUGGGACCCAGGGGGAACGCCUCU AACGCCGCGCCCUCAGCUAGCCCCAGGAAUGCCAGCGCUCCCAGGACCACCCCGAC UCCUCCGCAACCCCGCAAGGCGACCAAGUCCAAGGCGUCCACUGCCAAGCCAGCG CCUCCGCCUAAGACUGGCCCCCCUAAGACCUCCAGCGAACCUGUGCGGUGCAACC GGCACGACCCUCUGGCACGCUACGGAUCGCGGGUCCAAAUCCGGUGUCGGUUCCC GAACAGCACUCGGACCGAAUCGCGGCUCCAGAUUUGGAGAUACGCAACUGCCACU GAUGCCGAGAUCGGCACUGCCCCAAGCCUUGAGGAGGUCAUGGUCAACGUGUCAG CUCCUCCUGGAGGCCAGCUGGUGUACGACUCCGCUCCGAACCGAACCGACCCGCA CGUCAUCUGGGCCGAAGGAGCCGGUCCUGGUGCAUCGCCGAGGUUGUACUCGGUA GUGGGUCCCCUGGGGAGACAGCGGCUGAUCAUCGAAGAACUGACUCUGGAGACUC AGGGCAUGUACUAUUGGGUGUGGGGCAGAACCGAUAGACCAUCCGCAUACGGAAC CUGGGUGCGCGUGAGAGUGUUCAGACCCCCGUCCUUGACAAUCCACCCGCAUGCG GUGCUCGAAGGGCAGCCCUUCAAGGCCACUUGCACUGCGGCCACUUACUACCCUG GAAACCGGGCCGAAUUCGUGUGGUUCGAGGAUGGACGGAGGGUGUUCGACCCGGC GCAGAUUCAUACGCAGACUCAGGAAAACCCGGACGGCUUCUCCACCGUGUCCACU GUGACUUCGGCCGCUGUGGGAGGACAAGGACCGCCACGCACCUUCACCUGUCAGC UGACCUGGCACCGCGACAGCGUGUCCUUUAGCCGGCGGAACGCAUCAGGCACUGC CUCCGUGUUGCCUCGCCCAACCAUUACCAUGGAGUUCACCGGAGAUCACGCCGUG UGCACUGCUGGCUGCGUCCCCGAAGGCGUGACCUUCGCCUGGUUUCUCGGGGACG ACUCAUCCCCGGCGGAAAAGGUGGCCGUGGCCUCUCAGACCAGCUGCGGUAGACC GGGAACCGCCACCAUCCGCUCCACUCUGCCGGUGUCGUACGAGCAGACCGAGUAC AUUUGUCGCCUGGCCGGAUACCCGGACGGUAUCCCAGUGCUCGAACACCACGGCA GCCAUCAGCCUCCGCCGAGAGAUCCUACCGAGCGCCAGGUCAUCCGGGCCGUGGA AGGAUGAUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCC CCCCAGCCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUC UGAGUGGGCGGC (SEQ ID NO: 119)
MRK_HSV-2 SgE, SQ-032211, CX-003794	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGCUCGCGGGGCCGG GUUGGUGUUUUUUGUUGGAGUUUGGGUCGUAUCGUGCCUGGCGGCAGCACCCAG AACGUCCUGGAAACGGGUUACCUCGGGCGAGGACGUGGUGUUGCUUCCGGCGCCC GCGGGGCCGGAGGAACGCACACGGGCCCACAAACUACUGUGGGCCGCGGAACCCC UGGAUGCCUGCGGUCCCCUGAGGCCGUCGUGGGUGGCGCUGUGGCCCCCGCGACG GGUGCUCGAAACGGUCGUGGAUGCGGCGUGCAUGCGCGCCCCGGAACCGCUCGCC AUAGCAUACAGUCCCCCGUUCCCCGCGGGCGACGAGGGACUGUAUUCGGAGUUGG CGUGGCGCGAUCGCGUAGCCGUGGUCAACGAGAGUCUGGUCAUCUACGGGGCCCU GGAGACGGACAGCGGUCUGUACACCCUGUCCGUGGUCGGCCUAAGCGACGAGGCG CGCCAAGUGGCGUCGGUGGUUCUGGUCGUGGAGCCCGCCCCUGUGCCGACCCCGA CCCCCGACGACUACGACGAAGAAGACGACGCGGGCGUGAGCGAACGCACGCCGGU CAGCGUACCCCCCCCGACCCCACCCCGUCGUCCCCCCGUCGCCCCCCCUACGCACC CUCGUGUUAUCCCCGAGGUGUCCCACGUGCGCGGGGUAACGGUCCAUAUGGAGAC CCCGGAGGCCAUUCUGUUUGCCCCCGGAGAGACGUUUGGGACGAACGUCUCCAUC CACGCCAUUGCCCAUGACGACGGUCCGUACGCCAUGGACGUCGUCUGGAUGCGGU UUGACGUGCCGUCCUCGUGCGCCGAGAUGCGGAUCUACGAAGCUUGUCUGUAUCA CCCGCAGCUUCCAGAAUGUCUAUCUCCGGCCGACGCGCCGUGCGCUGUAAGUUCC UGGGCGUACCGCCUGGCGGUCCGCAGCUACGCCGGCUGUUCCAGGACUACGCCCC CGCCGCGAUGUUUUGCCGAGGCUCGCAUGGAACCGGUCCCGGGGUUGGCGUGGUU AGCCUCCACCGUCAACCUGGAAUUCCAGCACGCCUCCCCUCAGCACGCCGGCCUUU ACCUGUGCGUGGUGUACGUGGACGAUCAUAUCCACGCCUGGGGCCACAUGACCAU CUCUACCGCGGCGCAGUACCGGAACGCGGUGGUGGAACAGCACUUGCCCCAGCGC CAGCCUGAACCCGUCGAGCCCACCCGCCCGCACGUAAGAGCACCCCCUCCCGCGCC

Strain	Nucleic Acid Sequence
	UUCCGCGCGCGGCCCGCUGCGCUGAUAAUAGGCUGGAGCCUCGGUGGCCAUGCUU CUUGCCCCUUGGGCCUCCCCCCAGCCCCUCCUCCCCUUCCUGCACCCGUACCCCCG UGGUCUUUGAAUAAAGUCUGAGUGGGCGGC (SEQ ID NO: 120)
MRK_HSV-2 SgI, SQ-032323, CX-002683	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGCCCGGCCGCUCGCUG CAGGGCCUGGCGAUCCUGGGCCUGUGGGUCUGCGCCACCGGCCUGGUCGUCCGCG GCCCCACGGUCAGUCUGGUCUCAGACUCACUCGUGGAUGCCGGGGCCGUGGGGCC CCAGGGCUUCGUGGAAGAGGACCUGCGUGUUUUCGGGGAGCUUCAUUUUGUGGG GGCCCAGGUCCCCCACACAAACUACUACGACGGCAUCAUCGAGCUGUUUCACUAC CCCCUGGGGAACCACUGCCCCCGCGUUGUACACGUGGUCACACUGACCGCAUGCC CCCGCCGCCCCGCCGUGGCGUUCACCUUGUGUCGCUCGACGCACCACGCCCACAGC CCCGCCUAUCCGACCCUGGAGCUGGGUCUGGCGCGGCAGCCGCUUCUGCGGGUUC GAACGGCAACGCGCGACUAUGCCGGUCUGUAUGUCCUGCGCGUAUGGGUCGGCAG CGCGACGAACGCCAGCCUGUUUGUUUUGGGGGUGGCGCUCUCUGCCAACGGGACG UUUGUGUAUAACGGCUCGGACUACGGCUCCUGCGAUCCGGCGCAGCUUCCCUUUU CGGCCCCGCGCCUGGGACCCUCGAGCGUAUACACCCCCGGAGCCUCCCGGCCCACC CCUCCACGGACAACGACAUCCCCGUCCUCCCCCUAGAGACCCGACCCCCGCCCCCGG GGACACAGGAACGCCUGCGCCCGCGAGCGGCGAGAGAGCCCCGCCCAAUUCCACG CGAUCGGCCAGCGAAUCGAGACACAGGCUAACCGUAGCCCAGGUAAUCCAGUGAU AAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCC CCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGG GCGGC (SEQ ID NO: 121)
MRK_HSV-2 SgD, SQ-032172, CX-004714	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGGGCGUUUGACCUC CGGCGUCGGGACGGCGGCCCUGCUAGUUGUCGCGGUGGGACUCCGCGUCGUCUGC GCCAAAUACGCCUUAGCAGACCCCUCGCUUAAGAUGGCCGAUCCCAAUCGAUUUC GCGGGAAGAACCUUCCGGUUUUGGACCAGCUGACCGACCCCCCCCGGGGUGAAGCG UGUUUACCACAUUCAGCCGAGCCUGGAGGACCCGUUCCAGCCCCCCAGCAUCCCG AUCACUGUGUACUACGCAGUGCUGGAACGUGCCUGCCGCAGCGUGCUCCUACAUG CCCCAUCGGAGGCCCCCCAGAUCGUGCGCGGGGCUUCGGACGAGGCCCGAAAGCA CACGUACAACCUGACCAUCGCCUGGUAUCGCAUGGGAGACAAUUGCGCUAUCCCC AUCACGGUUAUGGAAUACACCGAGUGCCCCUACAACAAGUCGUUGGGGGUCUGCC CCAUCCGAACGCAGCCCCGCUGGAGCUACUAUGACAGCUUUAGCGCCGUCAGCGA GGAUAACCUGGGAUUCCUGAUGCACGCCCCCGCCUUCGAGACCGCGGGUACGUAC CUGCGGCUAGUGAAGAUAAACGACUGGACGGAGAUCACACAAUUUAUCCUGGAGC ACCGGGCCCGCGCCUCCUGCAAGUACGCUCUCCCCCCUGCGCAUCCCCCCGGCAGCG UGCCUCACCUCGAAGGCCUACCAACAGGGCGUGACGGUCGACAGCAUCGGGAUGC UACCCCGCUUUAUCCCCGAAAACCAGCGCACCGUCGCCCUAUACAGCUUAAAAAU CGCCGGGUGGCACGGCCCCAAGCCCCCGUACACCAGCACCCUGCUGCCGCCGGAGC UGUCCGACACCACCAACGCCACGCAACCCGAACUCGUUCCGGAAGACCCCGAGGA CUCGGCCCUCUUAGAGGAUCCCGCCGGGACGGUGUCUUCGCAGAUCCCCCCAAAC UGGCACAUCCCGUCGAUCCAGGACGUCGCGCCGCACCACGCCCCCGCCGCCCCCAG CAACCCGUGAUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCC UCCCCCCAGCCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAA GUCUGAGUGGGCGGC (SEQ ID NO: 122)
MRK_HSV-2 ICP-0, SQ-032521, CX-004422	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGAACCGCGGCCUGG UACUUCAUCCCGCGCCGAUCCUGGACCGGAACGGCCACCUCGCCAGACCCCUGGA ACGCAGCCUGCAGCCCCUCACGCCUGGGGGAUGCUGAAUGAUAUGCAGUGGCUGG CCUCAAGCGACUCCGAGGAAGAGACAGAGGUCGGCAUCUCCGACGAUGAUCUCCA UCGGGAUUCUACUUCGGAAGCGGGCUCCACCGACACAGAGAUGUUCGAGGCCGGC CUGAUGGAUGCUGCGACCCCUCCCGCAAGACCGCCUGCCGAACGCCAAGGCUCGC CGACCCCUGCUGACGCCCAGGGUUCGUGCGGUGGAGGCCCUGUGGGGGAGGAGGA AGCUGAAGCCGGAGGCGGUGGAGAUGUCAACACCCCGGUGGCCUACCUGAUCGUG GGCGUGACUGCCAGCGGAUCCUUCUCGACCAUCCCCAUUGUCAACGAUCCCCGCA CUCGGGUCGAAGCGGAGGCCGCAGUGCGGGCUGGAACUGCCGUGGACUUCAUUUG GACUGGCAAUCCCAGGACCGCUCCCCGGUCACUGUCCCUGGGAGGACACACCGUC CGCGCCCUGUCACCAACUCCCCCGUGGCCUGGAACCGAUGACGAGGACGACGACC UGGCCGAUGUGGACUACGUGCCCCCUGCCCCAAGACGGGCUCCACGGAGAGGAGG

Strain	Nucleic Acid Sequence
	CGGAGGCGCCGGUGCCACCAGGGGCACCAGCCAACCCGCUGCCACCCGGCCUGCUC CUCCUGGGGCCCCGAGAUCCUCCUCAUCCGGCGGGGCACCUCUGAGAGCAGGAGU GGGCUCAGGCUCCGGAGGAGGACCCGCCGUGGCAGCUGUGGUCCCGCGAGUGGCC UCCUUGCCUCCGGCCGCAGGAGGCGGCCGGGCCCAGGCCAGAAGGGUGGGGGAGG ACGCGGCAGCCGCCGAAGGGCGCACUCCUCCAGCGCGCCAACCAAGAGCAGCGCA AGAGCCUCCGAUCGUGAUCUCCGAUAGCCCCCCACCGUCACCUCGCAGACCAGCC GGACCCGGGCCUCUGUCGUUCGUGAGCUCCAGCUCGGCCCAGGUGUCGAGCGGAC CUGGCGGUGGUGGACUCCCUCAGAGCAGCGGCAGAGCUGCCAGACCUCGCGCCGC CGUGGCCCCGAGGGUCAGGUCGCCGCCGAGAGCAGCUGCCGCCCCAGUGGUGUCC GCCUCAGCCGACGCCGCCGGUCCCGCGCCUCCUGCUGUGCCAGUGGACGCCCAUA GAGCGCCGCGGAGCAGAAUGACUCAGGCACAGACUGACACCCAGGCCCAGUCGCU CGGUAGGGCUGGAGCCACCGACGCCAGAGGAUCGGGCGGACCCGGAGCCGAAGGA GGGUCCGGUCCCGCCGCUUCCUCCUCCGCGUCCUCAUCAGCCGCUCCGCGCUCACC GCUCGCACCCCAGGGUGUCGGAGCAAAGCGAGCAGCUCCUCGCCGGGCCCCUGAC UCCGACUCAGGAGAUCGGGGCCACGGACCACUCGCGCCUGCCAGCGCUGGAGCGG CUCCUCCAUCGGCUUCCCCAUCCUCGCAAGCAGCCGUGGCCGCCGCAUCCUCAAGC UCGGCGUCCUCUAGCUCAGCGAGCUCCUCCAGCGCCUCGUCCUCGUCCGCCUCCAG CAGCUCAGCCUCCUCGUCCUCGGCCUCCUCAUCGUCCGCCUCCUCCUCCGCUGGAG GUGCCGGAGGAUCGGUCGCAUCCGCUUCCGGCGCAGGGGAGCGCCGAGAAACGUC CCUGGGUCCGCGGGCAGCUGCUCCGAGGGGUCCUCGCAAGUGCGCGCGGAAAACU CGGCACGCGGAGGGAGGACCGGAACCUGGCGCGAGAGAUCCUGCGCCUGGACUGA CCCGGUACCUCCCCCAUUGCCGGGGUGUCCAGCGUGGUGGCACUUGCCCCGUACGU CAACAAGACCGUGACCGGGGACUGUCUCCCCGUGCUCGACAUGGAGACUGGACAC AUUGGCGCGUAUGUGGUCCUGGUGGAUCAGACCGGUAAUGUGGCCGACCUUUUG AGAGCAGCGGCCCCAGCAUGGUCCCGCAGAACCCUGCUGCCUGAGCACGCCAGGA AUUGCGUGCGGCCGCCGGACUACCCGACUCCGCCCGCCAGCGAAUGGAACUCACU GUGGAUGACUCCCGUGGGCAACAUGCUGUUCGAUCAGGGGACCCUGGUCGGAGCC CUGGAUUUUCACGGCCUGCGCUCCAGACAUCCGUGGUCUAGGGAACAGGGUGCUC CUGCUCCCGCGGGUGAUGCCCCUGCUGGCCACGGCGAAUAGUGAUAAUAGGCUGG AGCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCUCCUCCCCU UCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGGCGGC (SEQ ID NO: 123)
MRK_HSV-2 ICP-4, SQ-032440, CX-002146	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGGAAAUAAG AGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGUCGGCCGAGCAGCG CAAGAAGAAGAAAACGACCACCACUACCCAGGGCAGAGGAGCCGAAGUCGCCAUG GCCGAUGAAGAUGGCGGGAGGCUGCGGGCCGCCGCUGAAACCACCGGAGGACCGG GAUCCCCUGACCCUGCGGACGGCCCACCUCCCACACCGAACCCGGACAGACGGCCU GCUGCAAGGCCCGGUUUCGGAUGGCACGGGGGACCCGAAGAGAACGAGGACGAAG CCGAUGACGCCGCGGCGGAUGCAGACGCCGACGAGGCGGCUCCCGCUUCGGGAGA AGCGGUGGACGAACCGGCCGCCGAUGGAGUGGUCAGCCCCCGCCAGCUCGCGCUG CUCGCGUCCAUGGUGGAUGAAGCCGUGAGAACUAUCCCCUCACCUCCGCCGGAAC GGGAUGGAGCUCAAGAGGAAGCCGCCAGAAGCCCGUCCCCUCCGAGAACUCCAUC CAUGCGGGCCGACUACGGCGAAGAGAAUGACGACGAUGAUGACGACGAUGAUGAC GAUGACCGCGAUGCCGGACGGUGGGUCCGCGGACCUGAGACUACCUCCGCCGUGC GCGGAGCCUACCCUGAUCCGAUGGCCUCACUUAGCCCCCGGCCACCCGCCCCCCGC CGCCACCACCACCAUCAUCACCACCGCAGAAGAAGGGCUCCCAGGCGCAGAUCAG CAGCUUCCGACAGCUCGAAGUCCGGCUCCUCGUCCUCCGCCAGCAGCGCAUCCUC GUCAGCGUCCUCAUCGUCCAGCGCCUCGGCGAGCUCCUCCGACGAUGACGACGAC GACGAUGCCGCCAGAGCUCCGGCAUCAGCCGCGGACCAUGCCGCCGGAGGAACCC UCGGUGCCGACGACGAGGAGGCCGGCGUGCCUGCCCGCGCUCCGGGAGCUGCUCC UAGGCCUUCACCACCCCGGGCGGAGCCAGCCCCUGCCAGAACGCCAGCAGCCACCG CUGGGCGAUUGGAGAGGCGGAGAGCCCGGGCCGCCGUGGCCGGUCGGGAUGCCAC CGGCCGCUUCACUGCCGGACGCCCUCGGCGCGUCGAACUGGACGCAGACGCCGCC UCGGGCGCGUUCUACGCCCGCUAUCGGGACGGUUAUGUGUCCGGCGAGCCUUGGC CUGGUGCCGGUCCUCCUCCGCCUGGGAGAGUGCUCUACGGGGGUCUGGGUGAUUC UCGGCCAGGGUUGUGGGGAGCCCCCGAGGCGGAGGAAGCCAGAGCCCGCUUCGAA GCAUCCGGAGCACCGGCCCCUGUGUGGGCGCCGGAACUGGGCGACGCCGCCCAAC AAUACGCCCUGAUCACACGCCUGCUCUACACUCCGGACGCCGAAGCCAUGGGCUG GCUGCAGAACCCGAGAGUGGCCCCGGGUGAUGUGGCCCUGGACCAGGCAUGCUUC

Strain	Nucleic Acid Sequence
	AGGAUUAGCGGAGCCGCGAGAAACUCGAGCAGCUUUAUCUCAGGAUCUGUGGCCC GAGCCGUGCCGCACCUGGGCUACGCGAUGGCCGCCGGACGCUUCGGAUGGGGGCU GGCCCAUGUCGCUGCCGCGGUGGCGAUGUCCCGGCGGUACGACCGGGCUCAGAAG GGUUUCCUCCUCACCAGCCUCCGGAGGGCAUACGCCCCGUUGCUGGCUCGGGAGA ACGCCGCUCUGACUGGCGCCCGCACUCCUGAUGACGGUGGCGACGCCAACCGCCA CGACGGCGACGAUGCACGGGGAAAGCCCGCGGCCGCCGCCGCCCCCCUUCCUAGC GCAGCCGCUUCGCCUGCCGACGAACGGGCUGUCCCUGCCGGAUACGGAGCCGCCG GUGUGCUGGCGGCCCUUGGGAGACUGUCAGCCGCGCCUGCUUCAGCGCCGGCCGG AGCCGACGAUGACGACGACGACGAUGGAGCCGGAGGAGGGGGCGGCGGUCGGAGA GCAGAAGCCGGCAGGGUGGCAGUCGAAUGCCUUGCUGCCUGUCGCGGGAUCCUCG AGGCGUUGGCCGAAGGCUUCGACGGCGACCUGGCGGCAGUGCCUGGCCUGGCCGG CGCCCGCCCCGCUGCCCCUCCACGGCCCGGUCCGGCCGGGGCCGCAGCCCCUCCGC AUGCUGACGCGCCUCGCCUCAGAGCAUGGCUGAGAGAAUUGAGAUUUGUGCGGGA UGCGCUGGUCCUUAUGCGCCUGAGGGGGGAUCUGAGGGUGGCCGGAGGUUCCGAG GCGGCCGUGGCUGCUGUGCGGGCCGUGUCCCUGGUGGCCGGUGCGCUGGGUCCCG CUCUGCCGCGGUCCCCUAGAUUGCUUUCCUCAGCGGCCGCCGCCGCAGCCGAUCU GCUCUUUCAGAACCAAAGCCUCAGGCCGCUGCUGGCCGACACUGUCGCCGCUGCG GACUCCCUCGCUGCCCCAGCCUCGGCCCCAAGAGAGGCUGCCGAUGCCCCUCGCCC CGCCGCGGCCCCGCCUGCCGGAGCAGCGCCGCCUGCACCCCCUACUCCCCCCCCGC GACCGCCACGCCCAGCCGCUCUUACCAGAAGGCCAGCUGAGGGUCCUGACCCGCA GGGCGGCUGGCGCAGACAGCCCCCGGGACCUUCCCACACUCCCGCCCCAUCUGCGG CUGCCCUUGAAGCAUACUGUGCCCCGAGAGCUGUGGCGGAGCUGACCGACCACCC UCUGUUCCCUGCACCUUGGCGGCCUGCCCUGAUGUUUGACCCGAGAGCGUUGGCC UCCCUGGCGGCCAGAUGUGCGGCCCCGCCUCCCGGAGGAGCCCCAGCUGCAUUCG GACCUCUGCGGGCAUCCGGACCACUGCGGCGCGCUGCUGCAUGGAUGCGGCAAGU GCCGGACCCUGAGGACGUUCGCGUGGUCAUUCUUUACUCCCCCCUGCCGGGAGAA GAUCUCGCCGCCGGCCGCGCGGGAGGAGGCCCUCCACCCGAGUGGUCCGCUGAAC GGGGAGGCCUGUCCUGCCUGCUGGCUGCCCUGGGAAACCGCCUGUGCGGACCAGC UACUGCCGCCUGGGCUGGAAACUGGACCGGCGCACCCGAUGUGUCAGCCCUCGGA GCGCAGGGAGUGCUGCUGCUGUCAACUCGCGACCUGGCAUUCGCCGGAGCUGUGG AGUUCCUGGGUCUGCUUGCCGGCGCGUGCGACCGGAGAUUGAUCGUCGUGAACGC UGUCAGAGCGGCCGCUUGGCCUGCCGCUGCUCCGGUGGUCAGCCGGCAGCACGCA UAUCUGGCCUGCGAGGUGCUGCCCGCCGUGCAGUGUGCCGUGCGGUGGCCAGCGG CCAGAGACUUGCGACGGACCGUGCUGGCCUCCGGUAGGGUCUUUGGCCCCGGAGU GUUCGCCCGCGUGGAGGCCGCCCAUGCCAGACUGUACCCCGACGCACCGCCCCUG AGACUGUGCCGGGGAGCCAACGUGCGGUACAGAGUCCGCACCCGCUUCGGACCCG AUACUCUGGUGCCAAUGUCACCGCGGGAAUAUAGGAGAGCCGUGCUCCCGGCACU GGACGGCAGAGCCGCCGCAUCCGGUGCUGGGGACGCGAUGGCACCCGGAGCCCCC GACUUUUGCGAGGAUGAAGCCCACAGCCAUCGGGCCUGUGCCAGAUGGGGCCUGG GUGCCCCUCUUCGCCCCGUGUACGUGGCCCUGGGGAGAGAUGCCGUCCGCGGUGG ACCAGCCGAGCUGAGAGGCCCACGCCGGGAAUUUUGCGCUCGGGCCCUGCUCGAG CCCGAUGGAGAUGCGCCUCCCCUUGUGCUGCGCGACGACGCUGACGCCGGCCCAC CUCCGCAAAUCCGGUGGGCCAGCGCCGCCGGUCGAGCAGGAACGGUGUUGGCAGC AGCCGGAGGAGGAGUCGAAGUGGUCGGAACCGCGGCUGGACUGGCAACCCCGCCA AGGCGCGAACCUGUGGAUAUGGACGCCGAGCUGGAGGAUGACGACGAUGGCCUUU UCGGCGAGUGA<u>UGAUAAUAGGCUGGAGCCUCGGUGGCCAUGCUUCUUGCCCCUUG GGCCUCCCCCCAGCCCCUCCUCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAA UAAAGUCUGAGUGGGCGGC</u> (SEQ ID NO: 124)

The first underlined sequence is representive of the 5' UTR, which may be included in or omitted from any of the constructs listed in Table 1.

The second underlined sequence is representive of the 3' UTR, which may be included in or omitted from any of the constructs listed in Table 1.

Table 2: HSV Amino Acid Sequences

Strain	Amino Acid Sequence
gi|138220|sp|P06475.1|GC_HHV23 RecName: Full=Envelope glycoprotein C; Flags: Precursor	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTESRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTERQVIRAVEGAGIGVAVLVAVVLAGTAVVYLT HASSVRYRRLR (SEQ ID NO: 24)
gi|2842677|sp|Q89730.1|GC_HHV2H RecName: Full=Envelope glycoprotein C; Flags: Precursor	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTEFRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTERQVIRAVEGAGIGVAVLVAVVLAGTAVVYLT HASSVRYRRLR (SEQ ID NO: 25)
gi|138219|sp|P03173.1|GC_HHV2G RecName: Full=Envelope glycoprotein C; AltName: Full=Glycoprotein F; Flags: Precursor	MALGRVGLTVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSVPR NRSAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLAR YGSRVQIRCRFPNSTRTESRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPGG QLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQGM YYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATYY PGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPRT FTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGVT FAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYPD GIPVLEHHGSHQPPPRDPTERQVIRAVEGAGIGVAVLVAVVLAGTAVVYLTH ASSVRYRRLR (SEQ ID NO: 26)
gi|156072158|gb|ABU45430.1| glycoprotein C [Human herpesvirus 2]	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTESRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSPPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTERQVIRAVEGAGIGVAVLVAVVLAGTAVVYLT HASSVRYRRLR (SEQ ID NO: 27)
gi|156072221|gb|ABU45459.1| glycoprotein C [Human herpesvirus 2]	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTESRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRPIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTERQVIRAVEGAGIGVAVLVAVVLAGTAVVYLT HASSVRYRRLR (SEQ ID NO: 28)

Strain	Amino Acid Sequence
gi|807203116|gb|AKC59499.1| envelope glycoprotein C [Human herpesvirus 2]	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASPAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTEFRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTERQVIRAVEGAGIGVAVLVAVVLAGTAVVYLT HASSVRYRRLR (SEQ ID NO: 29)
gi|522172|gb|AAB60549.1| glycoprotein C [Human herpesvirus 2]	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTEFRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP HGIPVLEHHGSHQPPPRDPTERQVIRAVEGAGIGVAVLVAVVLAGTAVVYLT HASSVRYRRLR (SEQ ID NO: 30)
gi|392937653|gb|AFM93864.1| virion glycoprotein C [Human herpesvirus 2 strain 186]	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTEFRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTKRQVIRAVEGAGIGVAVLVAVVLAGTAVVYLT HASSVRYRRLR (SEQ ID NO: 31)
gi|330271|gb|AAA45842.1| glycoprotein-D [Human herpesvirus 2]	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFTPENQRTVALYSLKI AGWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPP NWHIPSIQDVAPHHAPAAPANPGLIIGALAGSTLAALVIGGIAFWVRRRRSVA PKRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 32)
gi|56698864|gb|AAW23130.1| glycoprotein-D [Human herpesvirus 2]	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAALVIGGIAFWVRRRAQMAP KRPRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 33)
gi|405168231|gb|AFS18221.1| virion glycoprotein D [Human herpesvirus 2]	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDTLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAVLVIGGIAFWVRRRAQMAP KRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 34)

Strain	Amino Acid Sequence
gi|674748224|gb|AIL27730.1| glycoprotein D [Human herpesvirus 2]	MGRLTSGVGTAALLVVAVGLRVVYAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYMRLVKINDWTEITQFILEHR ARASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKI AGWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPP NWHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAALVIGGIAFWVRRRAQMA PKRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 35)
gi|674748211|gb|AIL27728.1| glycoprotein D [Human herpesvirus 2]	MGRLTSGVGTAALLVVAVGLRVVYAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAALVIGGIAFWVRRRAQMAP KRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 36)
gi|154744645|gb|ABS84899.1| glycoprotein D [Human herpesvirus 2]	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAASEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAVLVIGGIAFWVRRRAQMAP KRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 37)
gi|156072225|gb|ABU45461.1| glycoprotein D [Human herpesvirus 2]	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DRLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAVLVIGGIAFWVRRRAQMAP KRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 38)
gi|82013827|sp|Q69467.1|GD_HHV2H| glycoprotein D	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAVLVIGGIAFWVRRRAQMAP KRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 39)
gi|522178|gb|AAB60554.1 glycoprotein D [Human herpesvirus 2]|	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAALVIGGIAFWVRRRAQMAP KRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 40)
gi|674748163|gb|AIL27723.1| glycoprotein D [Human herpesvirus 2]	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAALVIGGIAFWVRRRAQMAP KRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 41)

Strain	Amino Acid Sequence
HSV-2 gB; accession number HM011304 (isolate 00-10045)	MRGGGLVCALVVGALVAAVASAAPAAPRASGGVAATVAANGGPASQPPPV PSPATTKARKRKTKKPPKRPEATPPPDANATVAAGHATLRAHLREIKVENAD AQFYVCPPPTGATVVQFEQPRRCPTRPEGQNYTEGIAVVFKENIAPYKFKATM YYKDVTVSQVWFGHRYSQFMGIFEDRAPVPFEEVIDKINAKGVCRSTAKYVR NNMETTAFHRDDHETDMELKPAKVATRTSRGWHTTDLKYNPSRVEAFHRY GTTVNCIVEEVDARSVYPYDEFVLATGDFVYMSPFYGYREGSHTEHTSYAAD RFKQVDGFYARDLTTKARATSPTTRNLLTTPKFTVAWDWVPKRPAVCTMTK WQEVDEMLRAEYGGSFRFSSDAISTTFTTNLTQYSLSRVDLGDCIGRDAREAI DRMFARKYNATHIKVGQPQYYLATGGFLIAYQPLLSNTLAELYVREYMREQ DRKPRNATPAPLREAPSANASVERIKTTSSIEFARLQFTYNHIQRHVNDMLGRI AVAWCELQNHELTLWNEARKLNPNAIASATVGRRVSARMLGDVMAVSTCV PVAPDNVIVQNSMRVSSRPGTCYSRPLVSFRYEDQGPLIEGQLGENNELRLTR DALEPCTVGHRRYFIFGGGYVYFEEYAYSHQLSRADVTTVSTFIDLNITMLED HEFVPLEVYTRHEIKDSGLLDYTEVQRRNQLHDLRFADIDTVIRADANAAMF AGLCAFFEGMGDLGRAVGKVVMGVVGGVVSAVSGVSSFMSNPFGALAVGL LVLAGLVAAFFAFRYVLQLQRNPMKALYPLTTKELKTSDPGGVGGEGEEGA EGGGFDEAKLAEAREMIRYMALVSAMERTEHKARKKGTSALLSSKVTNMVL RKRNKARYSPLHNEDEAGDEDEL (SEQ ID NO: 42)
HSV-2 gC; accession number KP192856 (strain 333)	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTESRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTERQVIRAVEGAGIGVAVLVAVVLAGTAVVYLT HASSVRYRRLR (SEQ ID NO: 43)
HSV-2 gD; accession number JN561323 (strain HG52)	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAVLVIGGIAFWVRRRAQMAP KRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 44)
HSV-2 gE; accession number EU018094 (strain 333)	MARGAGLVFFVGVWVVSCLAAAPRTSWKRVTSGEDVVLLPAPAGPEERTRA HKLLWAAEPLDACGPLRPSWVALWPPRRVLETVVDAACMRAPEPLAIAYSP PFPAGDEGLYSELAWRDRVAVVNESLVIYGALETDSGLYTLSVVGLSDEARQ VASVVLVVEPAPVPTPTPDDYDEEDDAGVSERTPVSVPPPTPPRRPPVAPPTH PRVIPEVSHVRGVTVHMETPEAILFAPGETFGTNVSIHAIAHDDGPYAMDVV WMRFDVPSSCAEMRIYEACLYHPQLPECLSPADAPCAVSSWAYRLAVRSYA GCSRTTPPPRCFAEARMEPVPGLAWLASTVNLEFQHASPQHAGLYLCVVYVD DHIHAWGHMTISTAAQYRNAVVEQHLPQRQPEPVEPTRPHVRAPPPAPSARG PLRLGAVLGAALLLAALGLSAWACMTCWRRRSWRAVKSRASATGPTYIRVA DSELYADWSSDSEGERDGSLWQDPPERPDSPSTNGSGFEILSPTAPSVYPHSE GRKSRRPLTTFGSGSPGRRHSQASYSSVLW* (SEQ ID NO: 45)
HSV-2 gI; accession number KP192856 (strain 333)	MPGRSLQGLAILGLWVCATGLVVRGPTVSLVSDSLVDAGAVGPQGFVEEDL RVFGELHFVGAQVPHTNYYDGIIELFHYPLGNHCPRVVHVVTLTACPRRPAV AFTLCRSTHHAHSPAYPTLELGLARQPLLRVRTATRDYAGLYVLRVWVGSAT NASLFVLGVALSANGTFVYNGSDYGSCDPAQLPFSAPRLGPSSVYTPGASRPT PPRTTTSPSSPRDPTPAPGDTGTPAPASGERAPPNSTRSASESRHRLTVAQVIQI AIPASIIAFVFLGSCICFIHRCQRRYRRPRGQIYNPGGVSCAVNEAAMARLGAE LRSHPNTPPKPRRRSSSSTTMPSLTSIAEESEPGPVVLLSVSPRPRSGPTAPQEV (SEQ ID NO: 46)

Strain	Amino Acid Sequence
HSV-2 ICP-0; Based on strain HG52(inactivated by deletion of the nuclear localization signal and zinc-binding ring finger)	MEPRPGTSSRADPGPERPPRQTPGTQPAAPHAWGMLNDMQWLASSDSEEET EVGISDDDLHRDSTSEAGSTDTEMFEAGLMDAATPPARPPAERQGSPTPADA QGSCGGGPVGEEEAEAGGGGDVNTPVAYLIVGVTASGSFSTIPIVNDPRTRVE AEAAVRAGTAVDFIWTGNPRTAPRSLSLGGHTVRALSPTPPWPGTDDEDDDL ADVDYVPPAPRRAPRRGGGGAGATRGTSQPAATRPAPPGAPRSSSSGGAPLR AGVGSGSGGGPAVAAVVPRVASLPPAAGGGRAQARRVGEDAAAAEGRTPP ARQPRAAQEPPIVISDSPPPSPRRPAGPGPLSFVSSSSAQVSSGPGGGGLPQSSG RAARPRAAVAPRVRSPPRAAAAPVVSASADAAGPAPPAVPVDAHRAPRSRM TQAQTDTQAQSLGRAGATDARGSGGPGAEGGSGPAASSSASSSAAPRSPLAP QGVGAKRAAPRRAPDSDSGDRGHGPLAPASAGAAPPSASPSSQAAVAAASSS SASSSSASSSSASSSSASSSSASSSSASSSSASSSAGGAGGSVASASGAGERRET SLGPRAAAPRGPRKCARKTRHAEGGPEPGARDPAPGLTRYLPIAGVSSVVAL APYVNKTVTGDCLPVLDMETGHIGAYVVLVDQTGNVADLLRAAAPAWSRR TLLPEHARNCVRPPDYPTPPASEWNSLWMTPVGNMLFDQGTLVGALDFHGL RSRHPWSREQGAPAPAGDAPAGHGE (SEQ ID NO: 47)
HSV-2 SgB; (based on accession number HM011304; isolate 00-10045; truncated to remove transmembrane region)	MRGGGLVCALVVGALVAAVASAAPAAPRASGGVAATVAANGGPASQPPPV PSPATTKARKRKTKKPPKRPEATPPPDANATVAAGHATLRAHLREIKVENAD AQFYVCPPPTGATVVQFEQPRRCPTRPEGQNYTEGIAVVFKENIAPYKFKATM YYKDVTVSQVWFGHRYSQFMGIFEDRAPVPFEEVIDKINAKGVCRSTAKYVR NNMETTAFHRDDHETDMELKPAKVATRTSRGWHTTDLKYNPSRVEAFHRY GTTVNCIVEEVDARSVYPYDEFVLATGDFVYMSPFYGYREGSHTEHTSYAAD RFKQVDGFYARDLTTKARATSPTTRNLLTTPKFTVAWDWVPKRPAVCTMTK WQEVDEMLRAEYGGSFRFSSDAISTTFTTNLTQYSLSRVDLGDCIGRDAREAI DRMFARKYNATHIKVGQPQYYLATGGFLIAYQPLLSNTLAELYVREYMREQ DRKPRNATPAPLREAPSANASVERIKTTSSIEFARLQFTYNHIQRHVNDMLGRI AVAWCELQNHELTLWNEARKLNPNAIASATVGRRVSARMLGDVMAVSTCV PVAPDNVIVQNSMRVSSRPGTCYSRPLVSFRYEDQGPLIEGQLGENNELRLTR DALEPCTVGHRRYFIFGGGYVYFEEYAYSHQLSRADVTTVSTFIDLNITMLED HEFVPLEVYTRHEIKDSGLLDYTEVQRRNQLHDLRFADIDTVIRADANAAMF AGLCAFFEGMGDLGRAVGKVVMGVVGGVVSAVSGVSSFMSNP (SEQ ID NO: 48)
HSV-2 SgC; (based on accession number KP192856; strain 333; truncated to remove transmembrane region	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTESRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTERQVIRAVEG (SEQ ID NO: 49)
HSV-2 SgD (based on accession number JN561323; strain HG52; truncated to remove transmembrane region)	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNP (SEQ ID NO: 50)
HSV-2 SgE; (based on accession number EU018094; strain 333; truncated to remove transmembrane region)	MARGAGLVFFVGVWVVSCLAAAPRTSWKRVTSGEDVVLLPAPAGPEERTRA HKLLWAAEPLDACGPLRPSWVALWPPRRVLETVVDAACMRAPEPLAIAYSP PFPAGDEGLYSELAWRDRVAVVNESLVIYGALETDSGLYTLSVVGLSDEARQ VASVVLVVEPAPVPTPTPDDYDEEDDAGVSERTPVSVPPPTPPRRPPVAPPTH PRVIPEVSHVRGVTVHMETPEAILFAPGETFGTNVSIHAIAHDDGPYAMDVV WMRFDVPSSCAEMRIYEACLYHPQLPECLSPADAPCAVSSWAYRLAVRSYA GCSRTTPPPRCFAEARMEPVPGLAWLASTVNLEFQHASPQHAGLYLCVVYVD DHIHAWGHMTISTAAQYRNAVVEQHLPQRQPEPVEPTRPHVRAPPPAPSARG PLR (SEQ ID NO: 51)

Strain	Amino Acid Sequence
HSV-2 SgI; based on accession number KP192856; strain 333; truncated to remove transmembrane region)	MPGRSLQGLAILGLWVCATGLVVRGPTVSLVSDSLVDAGAVGPQGFVEEDL RVFGELHFVGAQVPHTNYYDGIIELFHYPLGNHCPRVVHVVTLTACPRRPAV AFTLCRSTHHAHSPAYPTLELGLARQPLLRVRTATRDYAGLYVLRVWVGSAT NASLFVLGVALSANGTFVYNGSDYGSCDPAQLPFSAPRLGPSSVYTPGASRPT PPRTTTSPSSPRDPTPAPGDTGTPAPASGERAPPNSTRSASESRHRLTVAQVIQ (SEQ ID NO: 52)
HSV-2 ICP-4; Based on strain HG52; (inactivated by deletion of nuclear localization signal and alanine substitution for key residues in the transactivation region)	MSAEQRKKKKTTTTTQGRGAEVAMADEDGGRLRAAAETTGGPGSPDPADG PPPTPNPDRRPAARPGFGWHGGPEENEDEADDAAADADADEAAPASGEAVD EPAADGVVSPRQLALLASMVDEAVRTIPSPPPERDGAQEEAARSPSPPRTPSM RADYGEENDDDDDDDDDDDRDAGRWVRGPETTSAVRGAYPDPMASLSPRP PAPRRHHHHHHHRRRRAPRRRSAASDSSKSGSSSSASSASSSSSSSASASSS DDDDDDDAARAPASAADHAAGGTLGADDEEAGVPARAPGAAPRPSPPRAEP APARTPAATAGRLERRRARAAVAGRDATGRFTAGRPRRVELDADAASGAFY ARYRDGYVSGEPWPGAGPPPPGRVLYGGLGDSRPGLWGAPEAEEARARFEA SGAPAPVWAPELGDAAQQYALITRLLYTPDAEAMGWLQNPRVAPGDVALD QACFRISGAARNSSSFISGSVARAVPHLGYAMAAGRFGWGLAHVAAAVAMS RRYDRAQKGFLLTSLRRAYAPLLARENAALTGARTPDDGGDANRHDGDDAR GKPAAAAAPLPSAAASPADERAVPAGYGAAGVLAALGRLSAAPASAPAGAD DDDDDDGAGGGGGGRRAEAGRVAVECLAACRGILEALAEGFDGDLAAVPG LAGARPAAPPRPGPAGAAAPPHADAPRLRAWLRELRFVRDALVLMRLRGDL RVAGGSEAAVAAVRAVSLVAGALGPALPRSPRLLSSAAAAAADLLFQNQSL RPLLADTVAAADSLAAPASAPREAADAPRPAAAPPAGAAPPAPPTPPPRPPRP AALTRRPAEGPDPQGGWRRQPPGPSHTPAPSAAALEAYCAPRAVAELTDHPL FPAPWRPALMFDPRALASLAARCAAPPPGGAPAAFGPLRASGPLRRAAAWM RQVPDPEDVRVVILYSPLPGEDLAAGRAGGGPPPEWSAERGGLSCLLAALGN RLCGPATAAWAGNWTGAPDVSALGAQGVLLLSTRDLAFAGAVEFLGLLAG ACDRRLIVVNAVRAAAWPAAAPVVSRQHAYLACEVLPAVQCAVRWPAARD LRRTVLASGRVFGPGVFARVEAAHARLYPDAPPLRLCRGANVRYRVRTRFGP DTLVPMSPREYRRAVLPALDGRAAASGAGDAMAPGAPDFCEDEAHSHRACA RWGLGAPLRPVYVALGRDAVRGGPAELRGPRREFCARALLEPDGDAPPLVL RDDADAGPPPQIRWASAAGRAGTVLAAAGGGVEVVGTAAGLATPPRREPVD MDAELEDDDDGLFGE* (SEQ ID NO: 53)
MRK_HSV-2 gB, SQ-032178	MRGGGLVCALVVGALVAAVASAAPAAPRASGGVAATVAANGGPASQPPPV PSPATTKARKRKTKKPPKRPEATPPPDANATVAAGHATLRAHLREIKVENAD AQFYVCPPPTGATVVQFEQPRRCPTRPEGQNYTEGIAVVFKENIAPYKFKATM YYKDVTVSQVWFGHRYSQFMGIFEDRAPVPFEEVIDKINAKGVCRSTAKYVR NNMETTAFHRDDHETDMELKPAKVATRTSRGWHTTDLKYNPSRVEAFHRY GTTVNCIVEEVDARSVYPYDEFVLATGDFVYMSPFYGYREGSHTEHTSYAAD RFKQVDGFYARDLTTKARATSPTTRNLLTTPKFTVAWDWVPKRPAVCTMTK WQEVDEMLRAEYGGSFRFSSDAISTTFTTNLTQYSLSRVDLGDCIGRDAREAI DRMFARKYNATHIKVGQPQYYLATGGFLIAYQPLLSNTLAELYVREYMREQ DRKPRNATPAPLREAPSANASVERIKTTSSIEFARLQFTYNHIQRHVNDMLGRI AVAWCELQNHELTLWNEARKLNPNAIASATVGRRVSARMLGDVMAVSTCV PVAPDNVIVQNSMRVSSRPGTCYSRPLVSFRYEDQGPLIEGQLGENNELRLTR DALEPCTVGHRRYFIFGGGYVYFEEYAYSHQLSRADVTTVSTFIDLNITMLED HEFVPLEVYTRHEIKDSGLLDYTEVQRRNQLHDLRFADIDTVIRADANAAMF AGLCAFFEGMGDLGRAVGKVVMGVVGGVVSAVSGVSSFMSNPFGALAVGL LVLAGLVAAFFAFRYVLQLQRNPMKALYPLTTKELKTSDPGGVGGEGEEGA EGGGFDEAKLAEAREMIRYMALVSAMERTEHKARKKGTSALLSSKVTNMVL RKRNKARYSPLHNEDEAGDEDEL (SEQ ID NO: 66)

Strain	Amino Acid Sequence
MRK_HSV-2 gC, SQ-032179	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTESRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTERQVIRAVEGAGIGVAVLVAVVLAGTAVVYLT HASSVRYRRLR (SEQ ID NO: 67)
MRK_HSV-2 gD, SQ-032180	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNPGLIIGALAGSTLAVLVIGGIAFWVRRRAQMAP KRLRLPHIRDDDAPPSHQPLFY (SEQ ID NO: 68)
MRK_HSV-2 gE, SQ-032181	MARGAGLVFFVGVWVVSCLAAAPRTSWKRVTSGEDVVLLPAPAGPEERTRA HKLLWAAEPLDACGPLRPSWVALWPPRRVLETVVDAACMRAPEPLAIAYSP PFPAGDEGLYSELAWRDRVAVVNESLVIYGALETDSGLYTLSVVGLSDEARQ VASVVLVVEPAPVPTPTPDDYDEEDDAGVSERTPVSVPPPTPPRRPPVAPPTH PRVIPEVSHVRGVTVHMETPEAILFAPGETFGTNVSIHAIAHDDGPYAMDVV WMRFDVPSSCAEMRIYEACLYHPQLPECLSPADAPCAVSSWAYRLAVRSYA GCSRTTPPPRCFAEARMEPVPGLAWLASTVNLEFQHASPQHAGLYLCVVYVD DHIHAWGHMTISTAAQYRNAVVEQHLPQRQPEPVEPTRPHVRAPPPAPSARG PLRLGAVLGAALLLAALGLSAWACMTCWRRRSWRAVKSRASATGPTYIRVA DSELYADWSSDSEGERDGSLWQDPPERPDSPSTNGSGFEILSPTAPSVYPHSE GRKSRRPLTTFGSGSPGRRHSQASYSSVLW (SEQ ID NO: 69)
MRK_HSV-2 gI, SQ-032182	MPGRSLQGLAILGLWVCATGLVVRGPTVSLVSDSLVDAGAVGPQGFVEEDL RVFGELHFVGAQVPHTNYYDGIIELFHYPLGNHCPRVVHVVTLTACPRRPAV AFTLCRSTHHAHSPAYPTLELGLARQPLLRVRTATRDYAGLYVLRVWVGSAT NASLFVLGVALSANGTFVYNGSDYGSCDPAQLPFSAPRLGPSSVYTPGASRPT PPRTTTSPSSPRDPTPAPGDTGTPAPASGERAPPNSTRSASESRHRLTVAQVIQI AIPASIIAFVFLGSCICFIHRCQRRYRRPRGQIYNPGGVSCAVNEAAMARLGAE LRSHPNTPPKPRRRSSSSTTMPSLTSIAEESEPGPVVLLSVSPRPRSGPTAPQEV (SEQ ID NO: 70)
MRK_HSV-2 SgB, SQ-032210	MRGGGLVCALVVGALVAAVASAAPAAPRASGGVAATVAANGGPASQPPPV PSPATTKARKRKTKKPPKRPEATPPPDANATVAAGHATLRAHLREIKVENAD AQFYVCPPPTGATVVQFEQPRRCPTRPEGQNYTEGIAVVFKENIAPYKFKATM YYKDVTVSQVWFGHRYSQFMGIFEDRAPVPFEEVIDKINAKGVCRSTAKYVR NNMETTAFHRDDHETDMELKPAKVATRTSRGWHTTDLKYNPSRVEAFHRY GTTVNCIVEEVDARSVYPYDEFVLATGDFVYMSPFYGYREGSHTEHTSYAAD RFKQVDGFYARDLTTKARATSPTTRNLLTTPKFTVAWDWVPKRPAVCTMTK WQEVDEMLRAEYGGSFRFSSDAISTTFTTNLTQYSLSRVDLGDCIGRDAREAI DRMFARKYNATHIKVGQPQYYLATGGFLIAYQPLLSNTLAELYVREYMREQ DRKPRNATPAPLREAPSANASVERIKTTSSIEFARLQFTYNHIQRHVNDMLGRI AVAWCELQNHELTLWNEARKLNPNAIASATVGRRVSARMLGDVMAVSTCV PVAPDNVIVQNSMRVSSRPGTCYSRPLVSFRYEDQGPLIEGQLGENNELRLTR DALEPCTVGHRRYFIFGGGYVYFEEYAYSHQLSRADVTTVSTFIDLNITMLED HEFVPLEVYTRHEIKDSGLLDYTEVQRRNQLHDLRFADIDTVIRADANAAMF AGLCAFFEGMGDLGRAVGKVVMGVVGGVVSAVSGVSSFMSNP (SEQ ID NO: 71)

Strain	Amino Acid Sequence
MRK_HSV-2 SgC, SQ-032835	MALGRVGLAVGLWGLLWVGVVVVLANASPGRTITVGPRGNASNAAPSASP RNASAPRTTPTPPQPRKATKSKASTAKPAPPPKTGPPKTSSEPVRCNRHDPLA RYGSRVQIRCRFPNSTRTESRLQIWRYATATDAEIGTAPSLEEVMVNVSAPPG GQLVYDSAPNRTDPHVIWAEGAGPGASPRLYSVVGPLGRQRLIIEELTLETQG MYYWVWGRTDRPSAYGTWVRVRVFRPPSLTIHPHAVLEGQPFKATCTAATY YPGNRAEFVWFEDGRRVFDPAQIHTQTQENPDGFSTVSTVTSAAVGGQGPPR TFTCQLTWHRDSVSFSRRNASGTASVLPRPTITMEFTGDHAVCTAGCVPEGV TFAWFLGDDSSPAEKVAVASQTSCGRPGTATIRSTLPVSYEQTEYICRLAGYP DGIPVLEHHGSHQPPPRDPTERQVIRAVEG (SEQ ID NO: 72)
MRK_HSV-2 SgE, SQ-032211	MARGAGLVFFVGVWVVSCLAAAPRTSWKRVTSGEDVVLLPAPAGPEERTRA HKLLWAAEPLDACGPLRPSWVALWPPRRVLETVVDAACMRAPEPLAIAYSP PFPAGDEGLYSELAWRDRVAVVNESLVIYGALETDSGLYTLSVVGLSDEARQ VASVVLVVEPAPVPTPTPDDYDEEDDAGVSERTPVSVPPPTPPRRPPVAPPTH PRVIPEVSHVRGVTVHMETPEAILFAPGETFGTNVSIHAIAHDDGPYAMDVV WMRFDVPSSCAEMRIYEACLYHPQLPECLSPADAPCAVSSWAYRLAVRSYA GCSRTTPPPRCFAEARMEPVPGLAWLASTVNLEFQHASPQHAGLYLCVVYVD DHIHAWGHMTISTAAQYRNAVVEQHLPQRQPEPVEPTRPHVRAPPPAPSARG PLR (SEQ ID NO: 73)
MRK_HSV-2 SgI, SQ-032323	MPGRSLQGLAILGLWVCATGLVVRGPTVSLVSDSLVDAGAVGPQGFVEEDL RVFGELHFVGAQVPHTNYYDGIIELFHYPLGNHCPRVVHVVTLTACPRRPAV AFTLCRSTHHAHSPAYPTLELGLARQPLLRVRTATRDYAGLYVLRVWVGSAT NASLFVLGVALSANGTFVYNGSDYGSCDPAQLPFSAPRLGPSSVYTPGASRPT PPRTTTSPSSPRDPTPAPGDTGTPAPASGERAPPNSTRSASESRHRLTVAQVIQ (SEQ ID NO: 74)
MRK_HSV-2 SgD, SQ-032172	MGRLTSGVGTAALLVVAVGLRVVCAKYALADPSLKMADPNRFRGKNLPVL DQLTDPPGVKRVYHIQPSLEDPFQPPSIPITVYYAVLERACRSVLLHAPSEAPQI VRGASDEARKHTYNLTIAWYRMGDNCAIPITVMEYTECPYNKSLGVCPIRTQ PRWSYYDSFSAVSEDNLGFLMHAPAFETAGTYLRLVKINDWTEITQFILEHRA RASCKYALPLRIPPAACLTSKAYQQGVTVDSIGMLPRFIPENQRTVALYSLKIA GWHGPKPPYTSTLLPPELSDTTNATQPELVPEDPEDSALLEDPAGTVSSQIPPN WHIPSIQDVAPHHAPAAPSNP (SEQ ID NO: 75)
MRK_HSV-2 ICP-0, SQ-032521	MEPRPGTSSRADPGPERPPRQTPGTQPAAPHAWGMLNDMQWLASSDSEEET EVGISDDDLHRDSTSEAGSTDTEMFEAGLMDAATPPARPPAERQGSPTPADA QGSCGGGPVGEEEAEAGGGGDVNTPVAYLIVGVTASGSFSTIPIVNDPRTRVE AEAAVRAGTAVDFIWTGNPRTAPRSLSLGGHTVRALSPTPPWPGTDDEDDDL ADVDYVPPAPRRAPRRGGGGAGATRGTSQPAATRPAPPGAPRSSSSGGAPLR AGVGSGSGGGPAVAAVVPRVASLPPAAGGGRAQARRVGEDAAAAEGRTPP ARQPRAAQEPPIVISDSPPPSPRRPAGPGPLSFVSSSSAQVSSGPGGGGLPQSSG RAARPRAAVAPRVRSPPRAAAAPVVSASADAAGPAPPAVPVDAHRAPRSRM TQAQTDTQAQSLGRAGATDARGSGGPGAEGGSGPAASSSASSSAAPRSPLAP QGVGAKRAAPRRAPDSDSGDRGHGPLAPASAGAAPPSASPSSQAAVAAASSS SASSSSASSSSASSSSASSSSASSSSASSSSASSSASSSAGGAGGSVASASGAGERRET SLGPRAAAPRGPRKCARKTRHAEGGPEPGARDPAPGLTRYLPIAGVSSVVAL APYVNKTVTGDCLPVLDMETGHIGAYVVLVDQTGNVADLLRAAAPAWSRR TLLPEHARNCVRPPDYPTPPASEWNSLWMTPVGNMLFDQGTLVGALDFHGL RSRHPWSREQGAPAPAGDAPAGHGE (SEQ ID NO: 76)

Strain	Amino Acid Sequence
MRK_HSV-2 ICP-4, SQ-032440	MSAEQRKKKKTTTTTQGRGAEVAMADEDGGRLRAAAETTGGPGSPDPADG PPPTPNPDRRPAARPGFGWHGGPEENEDEADDAAADADADEAAPASGEAVD EPAADGVVSPRQLALLASMVDEAVRTIPSPPPERDGAQEEAARSPSPPRTPSM RADYGEENDDDDDDDDDDDRDAGRWVRGPETTSAVRGAYPDPMASLSPRP PAPRRHHHHHHHRRRRAPRRRSAASDSSKSGSSSSASSASSSASSSSSASASSS DDDDDDDAARAPASAADHAAGGTLGADDEEAGVPARAPGAAPRPSPPRAEP APARTPAATAGRLERRRARAAVAGRDATGRFTAGRPRRVELDADAASGAFY ARYRDGYVSGEPWPGAGPPPPGRVLYGGLGDSRPGLWGAPEAEEARARFEA SGAPAPVWAPELGDAAQQYALITRLLYTPDAEAMGWLQNPRVAPGDVALD QACFRISGAARNSSSFISGSVARAVPHLGYAMAAGRFGWGLAHVAAAVAMS RRYDRAQKGFLLTSLRRAYAPLLARENAALTGARTPDDGGDANRHDGDDAR GKPAAAAAPLPSAAASPADERAVPAGYGAAGVLAALGRLSAAPASAPAGAD DDDDDDGAGGGGGGRRAEAGRVAVECLAACRGILEALAEGFDGDLAAVPG LAGARPAAPPRPGPAGAAAPPHADAPRLRAWLRELRFVRDALVLMRLRGDL RVAGGSEAAVAAVRAVSLVAGALGPALPRSPRLLSSAAAAAADLLFQNQSL RPLLADTVAAADSLAAPASAPREAADAPRPAAAPPAGAAPPAPPTPPPRPPRP AALTRRPAEGPDPQGGWRRQPPGPSHTPAPSAAALEAYCAPRAVAELTDHPL FPAPWRPALMFDPRALASLAARCAAPPPGGAPAAFGPLRASGPLRRAAAWM RQVPDPEDVRVVILYSPLPGEDLAAGRAGGGPPPEWSAERGGLSCLLAALGN RLCGPATAAWAGNWTGAPDVSALGAQGVLLLSTRDLAFAGAVEFLGLLAG ACDRRLIVVNAVRAAAWPAAAPVVSRQHAYLACEVLPAVQCAVRWPAARD LRRTVLASGRVFGPGVFARVEAAHARLYPDAPPLRLCRGANVRYRVRTRFGP DTLVPMSPREYRRAVLPALDGRAAASGAGDAMAPGAPDFCEDEAHSHRACA RWGLGAPLRPVYVALGRDAVRGGPAELRGPRREFCARALLEPDGDAPPLVL RDDADAGPPPQIRWASAAGRAGTVLAAAGGGVEVVGTAAGLATPPRREPVD MDAELEDDDDGLFGE (SEQ ID NO: 77)

Table 3: HSV strains/isolates, Envelope proteins/variants – *Homo sapiens*

Strain		NCBI Accession No.	Protein Accession No.
Human herpesvirus 2 strain CtSF	partial genome	KP334097.1	P06475.1 (SwissProt/EMBL)
Human herpesvirus 2 strain GSC-56	partial genome	KP334094.1	
Human herpesvirus 2 strain 333	partial genome	KP192856.1	
Herpes simplex virus type 2 glycoprotein C and 18K protein genes	complete cds	M10053.1	
Herpes simplex virus type 2 (strain 333) gene for glycoprotein C (gC-2) and 18K protein		X01996.1	
Human herpesvirus 2 MMA glycoprotein C (UL44) gene	complete cds	U12178.1	
Human herpesvirus 2 strain 333 glycoprotein C (UL44) gene	complete cds	EU018087.1	
Human herpesvirus 2 strain 1192	partial genome	KP334095.1	
Human herpesvirus 2 strain SD90e	complete genome	KF781518.1	
Human herpesvirus 2 strain 333 (variant A4) glycoprotein C (UL44) gene	complete cds	EU018090.1	

Strain		NCBI Accession No.	Protein Accession No.
Human herpesvirus 2 strain 333 (variant AC8) glycoprotein C (UL44) gene	complete cds	EU018089.1	
Human herpesvirus 2 glycoprotein C precursor (UL44) gene	complete cds	AF021341.1	Q89730.1 (SwissProt/EMBL) YP_009137196.1 (GenBank)
Human herpesvirus 2 WTW1A glycoprotein C (UL44) gene	complete cds	U12179.1	
Herpes simplex virus type 2 ul44 gene for glycoprotein C	isolate B4327UR	AJ297389.1	
Human herpesvirus 2 strain HG52	complete genome	JN561323.2	
Herpes simplex virus type 2 (strain HG52)	complete genome	Z86099.2	
Human herpesvirus 2 JDZ3 glycoprotein C (UL44) gene	complete cds	U12177.1	
Herpes simplex virus type 2 glycoprotein F gene		X01456.1	P03173.1 (SwissProt/EMBL)
Human herpesvirus 2 strain 333 (variant AC1) glycoprotein C (UL44) gene	complete cds	EU018088.1	ABU45430.1 GI:156072158
Human herpesvirus 2 strain 333 (variant A2) glycoprotein C (UL44) gene	complete cds	EU018122.1	ABU45459.1 GI:156072221
Human herpesvirus 2 strain COH 3818	partial genome	KP334096.1	AKC59499.1 GI:807203116
Human herpesvirus 2 strain CtSF-R	partial genome	KP334093.1	
Human herpesvirus 2 CAM4B glycoprotein C (UL44) gene	complete cds	U12176.1	AAB60549.1 GI:522172
Human herpesvirus 2 Strain 186 (Broad Institute)	partial genome	JX112656.1	AFM93864.1 GI:392937653
Human herpesvirus 2 isolate 10045 from USA glycoprotein C (UL44) gene	partial cds	AY827344.1	
Human herpesvirus 2 9788_00_802swab_1486 gC gene	partial cds	DQ236133.1	
Human herpesvirus 2 isolate 8484 from USA glycoprotein C (UL44) gene	partial cds	AY827357.1	

Strain		NCBI Accession No.	Protein Accession No.
Human herpesvirus 2 isolate 8028 from USA glycoprotein C (UL44) gene	partial cds	AY827351.1	
Human herpesvirus 2 isolate 8456 from USA glycoprotein C (UL44) gene	partial cds		
Human herpesvirus 2 strain 16293 glycoprotein D (US6) gene	complete cds	AY779754.1	Q69467.1 GI:82013827 (SwissProt/EMBL) YP_009137218.1 (BenBank)
Human herpesvirus 2 strain HG52	complete genome	JN561323.2	
Herpes simplex virus type 2 (strain HG52)	complete genome	Z86099.2	
Human herpesvirus 2 JDZ3 glycoprotein D (US6) gene	complete cds	U12181.1	
HSV-2 genomic HindIII l region of short unique component U(s) with genes US2 to US8		X04798.1	
Human herpesvirus 2 isolate pat5 glycoprotein D (US6) gene	complete cds	KF588422.1	
Human herpesvirus 2 isolate pat14 glycoprotein D (US6) gene	complete cds	KM068891.1	
Human herpesvirus 2 isolate pat13 glycoprotein D (US6) gene	complete cds	KM068890.1	
Human herpesvirus 2 strain 2899 glycoprotein D (US6) gene	complete cds	AY779751.1	
Human herpesvirus 2 strain CtSF	partial genome	KP334097.1	
Human herpesvirus 2 strain COH 3818	partial genome	KP334096.1	
Human herpesvirus 2 strain GSC-56	partial genome	KP334094.1	
Human herpesvirus 2 strain CtSF-R	partial genome	KP334093.1	
Human herpesvirus 2 strain 333	partial genome	KP192856.1	
Human herpesvirus 2 strain SD90e	complete genome	KF781518.1	
Human herpesvirus 2 isolate Pt13 virion glycoprotein D (US6) gene	complete cds	JQ956362.1	
Human herpesvirus 2 strain MS glycoprotein D gene	complete cds	EU445527.1	
Human herpesvirus 2 strain 333 glycoprotein D (US6) gene	complete cds	EU018091.1	
Human herpesvirus 2 isolate Pt21 virion glycoprotein D (US6) gene	complete cds	JQ956369.1	

Strain		NCBI Accession No.	Protein Accession No.
Human herpesvirus 2 isolate Pt05 virion glycoprotein D (US6) gene	complete cds	JQ956354.1	
Human herpesvirus 2 isolate Pt01 virion glycoprotein D (US6) gene	complete cds	JQ956351.1	
Human herpesvirus 2 strain 333 (variant AC2) glycoprotein D (US6) gene	complete cds	EU018092.1	
Human herpesvirus 2 isolate Iranian glycoprotein D (us6) gene	complete cds	AY517492.1	
Human herpesvirus 2 MMA glycoprotein D (US6) gene	complete cds	U12182.1	AAB60554.1 GI:522178
Human herpesvirus 2 glycoprotein D precursor (US6) gene	complete cds	AF021342.1	
Human herpesvirus 2 CAM4B glycoprotein D (US6) gene	complete cds	U12180.1	
Herpes simplex virus type 2 (HSV-2) glycoprotein D (gD-2) gene and flanks	K01408.1		
Human herpesvirus 2 isolate Pt11 virion glycoprotein D (US6) gene	complete cds	JQ956360.1	
Human herpesvirus 2 strain 1192	partial genome	KP334095.1	
Human herpesvirus 2 isolate pat6 glycoprotein D (US6) gene	complete cds	KF588423.1	
Human herpesvirus 2 isolate Pt25 virion glycoprotein D (US6) gene	complete cds	JQ956373.1	
Human herpesvirus 2 strain 333 (variant AC1) glycoprotein D (US6) gene	complete cds	EU018124.1	ABU45461.1 GI:156072225
Human herpesvirus 2 isolate subject ID VRC11098 specimen 2002_346 glycoprotein D (US6) gene	complete cds	EU029158.1	ABS84899.1 GI:154744645
Human herpesvirus 2 isolate pat4 glycoprotein D (US6) gene	complete cds	KF588421.1 GI:674748162	AIL27723.1 GI:674748163
Human herpesvirus 2 isolate pat10 glycoprotein D (US6) gene	complete cds	KF588427.1	
Human herpesvirus 2 isolate pat9 glycoprotein D (US6) gene	complete cds	KF588426.1	AIL27728.1 GI:674748211

Strain		NCBI Accession No.	Protein Accession No.
Human herpesvirus 2 isolate pat8 glycoprotein D (US6) gene	complete cds	KF588425.1	
Human herpesvirus 2 isolate pat7 glycoprotein D (US6) gene	complete cds	KF588424.1	
Human herpesvirus 2 isolate pat3 glycoprotein D (US6) gene	complete cds	KF588420.1	
Human herpesvirus 2 isolate pat2 glycoprotein D (US6) gene	complete cds	KF588419.1	
Human herpesvirus 2 isolate pat1 glycoprotein D (US6) gene	complete cds	KF588418.1	
Human herpesvirus 2 isolate pat11 glycoprotein D (US6) gene	complete cds	KF588428.1	AIL27730.1 GI:674748224
Human herpesvirus 2 isolate pat12 glycoprotein D (US6) gene	complete cds	KF588429.1	
Human herpesvirus 2 strain 333 (variant A6) glycoprotein D (US6) gene	complete cds	EU018093.1	ABU45435.1 GI:156072168
Human herpesvirus 2 isolate Pt26 virion glycoprotein D (US6) gene	complete cds	JQ956374.1	AFS18221.1 GI:405168231
Human herpesvirus 2 strain 2589 glycoprotein D (US6) gene	complete cds	AY779750.1	AAW23130.1 GI:56698864
Herpes simplex virus type 2 glycoprotein-D gene	complete cds	K02373.1	AAA45842.1 GI:330271
HSV-1			
Human herpesvirus 1 strain TFT401	partial genome	JN420337.1	
Human herpesvirus 1 strain 81L partial genome	KR052508 .1		
Human herpesvirus 1 strain 5-4-2	partial genome	KR011311.1	
Human herpesvirus 1 strain 10-11-3	partial genome	KR011309.1	
Human herpesvirus 1 strain 10-6-2	partial genome	KR011306.1	
Human herpesvirus 1 strain 47M	partial genome	KR011305.1	
Human herpesvirus 1 strain 31XL	partial genome	KR011304.1	
Human herpesvirus 1 strain 10-1-2	partial genome	KR011302.1	
Human herpesvirus 1 strain 10-5-1	partial genome	KR011301.1	
Human herpesvirus 1 strain 76M	partial genome	KR011300.1	

Strain		NCBI Accession No.	Protein Accession No.
Human herpesvirus 1 strain 5-1-1	partial genome	KR011299.1	
Human herpesvirus 1 strain 10-6-1	partial genome	KR011296.1	
Human herpesvirus 1 strain 5-5-2	partial genome	KR011295.1	
Human herpesvirus 1 strain 11M	partial genome	KR011294.1	
Human herpesvirus 1 strain 2-5-3	partial genome	KR011292.1	
Human herpesvirus 1 strain 10-14-1	partial genome	KR011291.1	
Human herpesvirus 1 strain 10-7-1	partial genome	KR011290.1	
Human herpesvirus 1 strain 2-4-2	partial genome	KR011288.1	
Human herpesvirus 1 strain 12-12-67	partial genome	KR011286.1	
Human herpesvirus 1 strain 5-2-1	partial genome	KR011285.1	
Human herpesvirus 1 strain 10-6-3	partial genome	KR011284.1	
Human herpesvirus 1 strain 3M	partial genome	KR011282.1	
Human herpesvirus 1 strain 66S	partial genome	KR011281.1	
Human herpesvirus 1 strain 36L	partial genome	KR011279.1	
Human herpesvirus 1 strain 10-2-2	partial genome	KR011277.1	
Human herpesvirus 1 strain 57M	partial genome	KR011276.1	
Human herpesvirus 1 strain 10-2-3	partial genome	KR011274.1	
Human herpesvirus 1 isolate RE	complete genome	KF498959.1	
Human herpesvirus 1 strain E19	partial genome	HM585511.2	
Human herpesvirus 1 strain CR38	partial genome	HM585508.2	
Human herpesvirus 1 strain E13	partial genome	HM585502.2	
Human herpesvirus 1 strain E08	partial genome	HM585498.2	
Human herpesvirus 1 strain KOS	complete genome	JQ780693.1	
Human herpesvirus 1 strain KOS	complete genome	JQ673480.1	
Human herpesvirus 1 strain OD4	partial genome	JN420342.1	
Human herpesvirus 1 strain KOSc glycoprotein D (US6) gene	complete cds	EF157319.1	
HSV1 glycoprotein D gene	J02217.1		
Herpes simplex virus type 1 glycoprotein D gene	complete cds	L09243.1	
Human herpesvirus 1 strain 12-12-2	partial genome	KR011298.1	

Strain		NCBI Accession No.	Protein Accession No.
Human herpesvirus 1 isolate HSV-1/0116209/India/2011	complete genome	KJ847330.1	
Human herpesvirus 1 strain R62	partial genome	HM585515.2	
Human herpesvirus 1 strain S25	partial genome	HM585513.2	
Human herpesvirus 1 strain KOSc(C2) glycoprotein D (US6) gene	complete cds	EF157322.1	
Human herpesvirus 1 strain KOSc(AC4) glycoprotein D (US6) gene	complete cds	EF157321.1	
Human herpesvirus 1 strain KOSc(AC3	AC6) glycoprote in D (US6) gene	complete cds	
EF157320.1			
Herpes simplex virus type 1 glycoprotein D gene	complete cds	L09244.1	
Herpes simplex virus type 1 glycoprotein D gene	complete cds	L09245.1	

Table 4. Signal Peptides

Description	Sequence	SEQ ID NO:
HuIgG$_k$ signal peptide	METPAQLLFLLLLWLPDTTG	78
IgE heavy chain epsilon -1 signal peptide	MDWTWILFLVAAATRVHS	79
Japanese encephalitis PRM signal sequence	MLGSNSGQRVVFTILLLLVAPAYS	80
VSVg protein signal sequence	MKCLLYLAFLFIGVNCA	81
Japanese encephalitis JEV signal sequence	MWLVSLAIVTACAGA	82

Table 5. Flagellin Nucleic Acid Sequences

Name	Sequence	SEQ ID NO:
NT (5' UTR, ORF, 3' UTR)	TCAAGCTTTTGGACCCTCGTACAGAAGCTAATACGACTCACTATAGGGAA ATAAGAGAGAAAAGAAGAGTAAGAAGAAATATAAGAGCCACCATGGCA CAAGTCATTAATACAAACAGCCTGTCGCTGTTGACCCAGAATAACCTGAA CAAATCCCAGTCCGCACTGGGCACTGCTATCGAGCGTTTGTCTTCCGGTCT GCGTATCAACAGCGCGAAAGACGATGCGGCAGGACAGGCGATTGCTAAC CGTTTTACCGCGAACATCAAAGGTCTGACTCAGGCTTCCCGTAACGCTAA CGACGGTATCTCCATTGCGCAGACCACTGAAGGCGCGCTGAACGAAATC AACAACAACCTGCAGCGTGTGCGTGAACTGGCGGTTCAGTCTGCGAATGG TACTAACTCCCAGTCTGACCTCGACTCCATCCAGGCTGAAATCACCCAGC GCCTGAACGAAATCGACCGTGTATCCGGCCAGACTCAGTTCAACGGCGTG AAAGTCCTGGCGCAGGACAACACCCTGACCATCCAGGTTGGTGCCAACG ACGGTGAAACTATCGATATTGATTTAAAAGAAATCAGCTCTAAAACACTG GGACTTGATAAGCTTAATGTCCAAGATGCCTACACCCCGAAAGAAACTGC TGTAACCGTTGATAAAACTACCTATAAAAATGGTACAGATCCTATTACAG CCCAGAGCAATACTGATATCCAAACTGCAATTGGCGGTGGTGCAACGGG GGTTACTGGGGCTGATATCAAATTTAAAGATGGTCAATACTATTTAGATG TTAAAGGCGGTGCTTCTGCTGGTGTTTATAAAGCCACTTATGATGAAACT ACAAAGAAAGTTAATATTGATACGACTGATAAAACTCCGTTGGCAACTGC GGAAGCTACAGCTATTCGGGGAACGGCCACTATAACCCACAACCAAATT GCTGAAGTAACAAAAGAGGGTGTTGATACGACCACAGTTGCGGCTCAAC TTGCTGCAGCAGGGGTTACTGGCGCCGATAAGGACAATACTAGCCTTGTA AAACTATCGTTTGAGGATAAAAACGGTAAGGTTATTGATGGTGGCTATGC	83

	AGTGAAAATGGGCGACGATTTCTATGCCGCTACATATGATGAGAAAACA GGTGCAATTACTGCTAAAACCACTACTTATACAGATGGTACTGGCGTTGC TCAAACTGGAGCTGTGAAATTTGGTGGCGCAAATGGTAAATCTGAAGTTG TTACTGCTACCGATGGTAAGACTTACTTAGCAAGCGACCTTGACAAACAT AACTTCAGAACAGGCGGTGAGCTTAAAGAGGTTAATACAGATAAGACTG AAAACCCACTGCAGAAAATTGATGCTGCCTTGGCACAGGTTGATACACTT CGTTCTGACCTGGGTGCGGTTCAGAACCGTTTCAACTCCGCTATCACCAA CCTGGGCAATACCGTAAATAACCTGTCTTCTGCCCGTAGCCGTATCGAAG ATTCCGACTACGCAACCGAAGTCTCCAACATGTCTCGCGCGCAGATTCTG CAGCAGGCCGGTACCTCCGTTCTGGCGCAGGCGAACCAGGTTCCGCAAA ACGTCCTCTCTTTACTGCGTTGATAATAGGCTGGAGCCTCGGTGGCCATG CTTCTTGCCCCTTGGGCCTCCCCCCAGCCCCTCCTCCCCTTCCTGCACCCG TACCCCCGTGGTCTTTGAATAAAGTCTGAGTGGGCGGC	
ORF Sequence, NT	ATGGCACAAGTCATTAATACAAACAGCCTGTCGCTGTTGACCCAGAATAA CCTGAACAAATCCCAGTCCGCACTGGGCACTGCTATCGAGCGTTTGTCTT CCGGTCTGCGTATCAACAGCGCGAAAGACGATGCGGCAGGACAGGCGAT TGCTAACCGTTTTACCGCGAACATCAAAGGTCTGACTCAGGCTTCCCGTA ACGCTAACGACGGTATCTCCATTGCGCAGACCACTGAAGGCGCGCTGAAC GAAATCAACAACAACCTGCAGCGTGTGCGTGAACTGGCGGTTCAGTCTGC GAATGGTACTAACTCCCAGTCTGACCTCGACTCCATCCAGGCTGAAATCA CCCAGCGCCTGAACGAAATCGACCGTGTATCCGGCCAGACTCAGTTCAAC GGCGTGAAAGTCCTGGCGCAGGACAACACCCTGACCATCCAGGTTGGTG CCAACGACGGTGAAACTATCGATATTGATTTAAAAGAAATCAGCTCTAAA ACACTGGGACTTGATAAGCTTAATGTCCAAGATGCCTACACCCCGAAAGA AACTGCTGTAACCGTTGATAAAACTACCTATAAAAATGGTACAGATCCTA TTACAGCCCAGAGCAATACTGATATCCAAACTGCAATTGGCGGTGGTGCA ACGGGGGTTACTGGGGCTGATATCAAATTTAAAGATGGTCAATACTATTT AGATGTTAAAGGCGGTGCTTCTGCTGGTGTTTATAAAGCCACTTATGATG AAACTACAAAGAAAGTTAATATTGATACGACTGATAAAACTCCGTTGGCA ACTGCGGAAGCTACAGCTATTCGGGGAACGGCCACTATAACCCACAACC AAATTGCTGAAGTAACAAAAGAGGGTGTTGATACGACCACAGTTGCGGC TCAACTTGCTGCAGCAGGGGTTACTGGCGCCGATAAGGACAATACTAGCC TTGTAAAACTATCGTTTGAGGATAAAAACGGTAAGGTTATTGATGGTGGC TATGCAGTGAAAATGGGCGACGATTTCTATGCCGCTACATATGATGAGAA AACAGGTGCAATTACTGCTAAAACCACTACTTATACAGATGGTACTGGCG TTGCTCAAACTGGAGCTGTGAAATTTGGTGGCGCAAATGGTAAATCTGAA GTTGTTACTGCTACCGATGGTAAGACTTACTTAGCAAGCGACCTTGACAA ACATAACTTCAGAACAGGCGGTGAGCTTAAAGAGGTTAATACAGATAAG ACTGAAAACCCACTGCAGAAAATTGATGCTGCCTTGGCACAGGTTGATAC ACTTCGTTCTGACCTGGGTGCGGTTCAGAACCGTTTCAACTCCGCTATCAC CAACCTGGGCAATACCGTAAATAACCTGTCTTCTGCCCGTAGCCGTATCG AAGATTCCGACTACGCAACCGAAGTCTCCAACATGTCTCGCGCGCAGATT CTGCAGCAGGCCGGTACCTCCGTTCTGGCGCAGGCGAACCAGGTTCCGCA AAACGTCCTCTCTTTACTGCGT	84
mRNA Sequence (assumes T100 tail)	G*GGGAAAUAAGAGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCAC CAUGGCACAAGUCAUUAAUACAAACAGCCUGUCGCUGUUGACCCAGAA UAACCUGAACAAAUCCCAGUCCGCACUGGGCACUGCUAUCGAGCGUUU GUCUUCCGGUCUGCGUAUCAACAGCGCGAAAGACGAUGCGGCAGGACA GGCGAUUGCUAACCGUUUUACCGCGAACAUCAAAGGUCUGACUCAGGC UUCCCGUAACGCUAACGACGGUAUCUCCAUUGCGCAGACCACUGAAGG CGCGCUGAACGAAAUCAACAACAACCUGCAGCGUGUGCGUGAACUGGC GGUUCAGUCUGCGAAUGGUACUAACUCCCAGUCUGACCUCGACUCCAU CCAGGCUGAAAUCACCCAGCGCCUGAACGAAAUCGACCGUGUAUCCGG CCAGACUCAGUUCAACGGCGUGAAAGUCCUGGCGCAGGACAACACCCU GACCAUCCAGGUUGGUGCCAACGACGGUGAAACUAUCGAUAUUGAUUU AAAAGAAAUCAGCUCUAAAACACUGGGACUUGAUAAGCUUAAUGUCCA AGAUGCCUACACCCCGAAAGAAACUGCUGUAACCGUUGAUAAAACUAC CUAUAAAAAUGGUACAGAUCCUAUUACAGCCCAGAGCAAUACUGAUAU CCAAACUGCAAUUGGCGGUGGUGCAACGGGGGUUACUGGGGCUGAUAU	85

	CAAAUUUAAAGAUGGUCAAUACUAUUUAGAUGUUAAAGGCGGUGCUUC UGCUGGUGUUUAUAAAGCCACUUAUGAUGAAACUACAAAGAAAGUUAA UAUUGAUACGACUGAUAAAACUCCGUUGGCAACUGCGGAAGCUACAGC UAUUCGGGGAACGGCCACUAUAACCCACAACCAAAUUGCUGAAGUAAC AAAAGAGGGUGUUGAUACGACCACAGUUGCGGCUCAACUUGCUGCAGC AGGGGUUACUGGCGCCGAUAAGGACAAUACUAGCCUUGUAAAACUAUC GUUUGAGGAUAAAAACGGUAAGGUUAUUGAUGGUGGCUAUGCAGUGA AAAUGGGCGACGAUUUCUAUGCCGCUACAUAUGAUGAGAAAACAGGUG CAAUUACUGCUAAAACCACUACUUAUACAGAUGGUACUGGCGUUGCUC AAACUGGAGCUGUGAAAUUUGGUGGCGCAAAUGGUAAAUCUGAAGUU GUUACUGCUACCGAUGGUAAGACUUACUUAGCAAGCGACCUUGACAAA CAUAACUUCAGAACAGGCGGUGAGCUUAAAGAGGUUAAUACAGAUAAG ACUGAAAACCCACUGCAGAAAAUUGAUGCUGCCUUGGCACAGGUUGAU ACACUUCGUUCUGACCUGGGUGCGGUUCAGAACCGUUUCAACUCCGCU AUCACCAACCUGGGCAAUACCGUAAAUAACCUGUCUUCUGCCCGUAGC CGUAUCGAAGAUUCCGACUACGCAACCGAAGUCUCCAACAUGUCUCGC GCGCAGAUUCUGCAGCAGGCCGGUACCUCCGUUCUGGCGCAGGCGAAC CAGGUUCCGCAAAACGUCCUCUCUUUACUGCGUUGAUAAUAGGCUGGA GCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCUCC UCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGU GGGCGGCAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAA AAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAA AAAAAAAAAAAAAUCUAG	
Flagellin mRNA Sequences		
NT (5' UTR, ORF, 3' UTR)	UCAAGCUUUUGGACCCUCGUACAGAAGCUAAUACGACUCACUAUAGGG AAAUAAGAGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUG GCACAAGUCAUUAAUACAAACAGCCUGUCGCUGUUGACCCAGAAUAAC CUGAACAAAUCCCAGUCCGCACUGGGCACUGCUAUCGAGCGUUUGUCU UCCGGUCUGCGUAUCAACAGCGCGAAAGACGAUGCGGCAGGACAGGCG AUUGCUAACCGUUUUACCGCGAACAUCAAAGGUCUGACUCAGGCUUCC CGUAACGCUAACGACGGUAUCUCCAUUGCGCAGACCACUGAAGGCGCG CUGAACGAAAUCAACAACAACCUGCAGCGUGUGCGUGAACUGGCGGUU CAGUCUGCGAAUGGUACUAACUCCCAGUCUGACCUCGACUCCAUCCAG GCUGAAAUCACCCAGCGCCUGAACGAAAUCGACCGUGUAUCCGGCCAG ACUCAGUUCAACGGCGUGAAAGUCCUGGCGCAGGACAACACCCUGACC AUCCAGGUUGGUGCCAACGACGGUGAAACUAUCGAUAUUGAUUUAAAA GAAAUCAGCUCUAAAACACUGGGACUUGAUAAGCUUAAUGUCCAAGAU GCCUACACCCCGAAAGAAACUGCUGUAACCGUUGAUAAAACUACCUAU AAAAAUGGUACAGAUCCUAUUACAGCCCAGAGCAAUACUGAUAUCCAA ACUGCAAUUGGCGGUGGUGCAACGGGGGUUACUGGGGCUGAUAUCAAA UUUAAAGAUGGUCAAUACUAUUUAGAUGUUAAAGGCGGUGCUUCUGCU GGUGUUUAUAAAGCCACUUAUGAUGAAACUACAAAGAAAGUUAAUAU UGAUACGACUGAUAAAACUCCGUUGGCAACUGCGGAAGCUACAGCUAU UCGGGGAACGGCCACUAUAACCCACAACCAAAUUGCUGAAGUAACAAA AGAGGGUGUUGAUACGACCACAGUUGCGGCUCAACUUGCUGCAGCAGG GGUUACUGGCGCCGAUAAGGACAAUACUAGCCUUGUAAAACUAUCGUU UGAGGAUAAAAACGGUAAGGUUAUUGAUGGUGGCUAUGCAGUGAAAA UGGGCGACGAUUUCUAUGCCGCUACAUAUGAUGAGAAAACAGGUGCAA UUACUGCUAAAACCACUACUUAUACAGAUGGUACUGGCGUUGCUCAAA CUGGAGCUGUGAAAUUUGGUGGCGCAAAUGGUAAAUCUGAAGUUGUU ACUGCUACCGAUGGUAAGACUUACUUAGCAAGCGACCUUGACAAACAU AACUUCAGAACAGGCGGUGAGCUUAAAGAGGUUAAUACAGAUAAGACU GAAAACCCACUGCAGAAAAUUGAUGCUGCCUUGGCACAGGUUGAUACA CUUCGUUCUGACCUGGGUGCGGUUCAGAACCGUUUCAACUCCGCUAUC ACCAACCUGGGCAAUACCGUAAAUAACCUGUCUUCUGCCCGUAGCCGU AUCGAAGAUUCCGACUACGCAACCGAAGUCUCCAACAUGUCUCGCGCG CAGAUUCUGCAGCAGGCCGGUACCUCCGUUCUGGCGCAGGCGAACCAG GUUCCGCAAAACGUCCUCUCUUUACUGCGUUGAUAAUAGGCUGGAGCC UCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCUCCUCC	86

	CCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGUGGGCGGC	
ORF Sequence, NT	AUGGCACAAGUCAUUAAUACAAACAGCCUGUCGCUGUUGACCCAGAAUAACCUGAACAAAUCCCAGUCCGCACUGGGCACUGCUAUCGAGCGUUUGUCUUCCGGUCUGCGUAUCAACAGCGCGAAAGACGAUGCGGCAGGACAGGCGAUUGCUAACCGUUUUACCGCGAACAUCAAAGGUCUGACUCAGGCUUCCCGUAACGCUAACGACGGUAUCUCCAUUGCGCAGACCACUGAAGGCGCGCUGAACGAAAUCAACAACAACCUGCAGCGUGUGCGUGAACUGGCGGUUCAGUCUGCGAAUGGUACUAACUCCCAGUCUGACCUCGACUCCAUCCAGGCUGAAAUCACCCAGCGCCUGAACGAAAUCGACCGUGUAUCCGGCCAGACUCAGUUCAACGGCGUGAAAGUCCUGGCGCAGGACAACACCCUGACCAUCCAGGUUGGUGCCAACGACGGUGAAACUAUCGAUAUUGAUUUAAAAGAAAUCAGCUCUAAAACACUGGGACUUGAUAAGCUUAAUGUCCAAGAUGCCUACACCCCGAAAGAAACUGCUGUAACCGUUGAUAAAACUACCUAUAAAAAUGGUACAGAUCCUAUUACAGCCCAGAGCAAUACUGAUAUCCAAACUGCAAUUGGCGGUGGUGCAACGGGGGUUACUGGGGCUGAUAUCAAAUUUAAAGAUGGUCAAUACUAUUUAGAUGUUAAAGGCGGUGCUUCUGCUGGUGUUUAUAAAGCCACUUAUGAUGAAACUACAAAGAAAGUUAAUAUUGAUACGACUGAUAAAACUCCGUUGGCAACUGCGGAAGCUACAGCUAUUCGGGGAACGGCCACUAUAACCCACAACCAAAUUGCUGAAGUAACAAAAGAGGGUGUUGAUACGACCACAGUUGCGGCUCAACUUGCUGCAGCAGGGGUUACUGGCGCCGAUAAGGACAAUACUAGCCUUGUAAAACUAUCGUUUGAGGAUAAAAACGGUAAGGUUAUUGAUGGUGGCUAUGCAGUGAAAAUGGGCGACGAUUUCUAUGCCGCUACAUAUGAUGAGAAAACAGGUGCAAUUACUGCUAAAACCACUACUUAUACAGAUGGUACUGGCGUUGCUCAAACUGGAGCUGUGAAAUUUGGUGGCGCAAAUGGUAAAUCUGAAGUUGUUACUGCUACCGAUGGUAAGACUUACUUAGCAAGCGACCUUGACAAACAUAACUUCAGAACAGGCGGUGAGCUUAAAGAGGUUAAUACAGAUAAGACUGAAAACCCACUGCAGAAAAUUGAUGCUGCCUUGGCACAGGUUGAUACACUUCGUUCUGACCUGGGUGCGGUUCAGAACCGUUUCAACUCCGCUAUCACCAACCUGGGCAAUACCGUAAAUAACCUGUCUUCUGCCCGUAGCCGUAUCGAAGAUUCCGACUACGCAACCGAAGUCUCCAACAUGUCUCGCGCGCAGAUUCUGCAGCAGGCCGGUACCUCCGUUCUGGCGCAGGCGAACCAGGUUCCGCAAAACGUCCUCUCUUUACUGCGU	87
mRNA Sequence (assumes T100 tail)	G*GGGAAAUAAGAGAGAAAAGAAGAGUAAGAAGAAAUAUAAGAGCCACCAUGGCACAAGUCAUUAAUACAAACAGCCUGUCGCUGUUGACCCAGAAUAACCUGAACAAAUCCCAGUCCGCACUGGGCACUGCUAUCGAGCGUUUGUCUUCCGGUCUGCGUAUCAACAGCGCGAAAGACGAUGCGGCAGGACAGGCGAUUGCUAACCGUUUUACCGCGAACAUCAAAGGUCUGACUCAGGCUUCCCGUAACGCUAACGACGGUAUCUCCAUUGCGCAGACCACUGAAGGCGCGCUGAACGAAAUCAACAACAACCUGCAGCGUGUGCGUGAACUGGCGGUUCAGUCUGCGAAUGGUACUAACUCCCAGUCUGACCUCGACUCCAUCCAGGCUGAAAUCACCCAGCGCCUGAACGAAAUCGACCGUGUAUCCGGCCAGACUCAGUUCAACGGCGUGAAAGUCCUGGCGCAGGACAACACCCUGACCAUCCAGGUUGGUGCCAACGACGGUGAAACUAUCGAUAUUGAUUUAAAAGAAAUCAGCUCUAAAACACUGGGACUUGAUAAGCUUAAUGUCCAAGAUGCCUACACCCCGAAAGAAACUGCUGUAACCGUUGAUAAAACUACCUAUAAAAAUGGUACAGAUCCUAUUACAGCCCAGAGCAAUACUGAUAUCCAAACUGCAAUUGGCGGUGGUGCAACGGGGGUUACUGGGGCUGAUAUCAAAUUUAAAGAUGGUCAAUACUAUUUAGAUGUUAAAGGCGGUGCUUCUGCUGGUGUUUAUAAAGCCACUUAUGAUGAAACUACAAAGAAAGUUAAUAUUGAUACGACUGAUAAAACUCCGUUGGCAACUGCGGAAGCUACAGCUAUUCGGGGAACGGCCACUAUAACCCACAACCAAAUUGCUGAAGUAACAAAAGAGGGUGUUGAUACGACCACAGUUGCGGCUCAACUUGCUGCAGCAGGGGUUACUGGCGCCGAUAAGGACAAUACUAGCCUUGUAAAACUAUCGUUUGAGGAUAAAAACGGUAAGGUUAUUGAUGGUGGCUAUGCAGUGAAAAUGGGCGACGAUUUCUAUGCCGCUACAUAUGAUGAGAAAACAGGUGCAAUUACUGCUAAAACCACUACUUAUACAGAUGGUACUGGCGUUGCUCAAACUGGAGCUGUGAAAUUUGGUGGCGCAAAUGGUAAAUCUGAAGUUGUUACUGCUACCGAUGGUAAGACUUACUUAGCAAGCGACCUUGACAAA	88

	CAUAACUUCAGAACAGGCGGUGAGCUUAAAGAGGUUAAUACAGAUAAG ACUGAAAACCCACUGCAGAAAAUUGAUGCUGCCUUGGCACAGGUUGAU ACACUUCGUUCUGACCUGGGUGCGGUUCAGAACCGUUUCAACUCCGCU AUCACCAACCUGGGCAAUACCGUAAAUAACCUGUCUUCUGCCCGUAGC CGUAUCGAAGAUUCCGACUACGCAACCGAAGUCUCCAACAUGUCUCGC GCGCAGAUUCUGCAGCAGGCCGGUACCUCCGUUCUGGCGCAGGCGAAC CAGGUUCCGCAAAACGUCCUCUCUUUACUGCGUUGAUAAUAGGCUGGA GCCUCGGUGGCCAUGCUUCUUGCCCCUUGGGCCUCCCCCCAGCCCCUCC UCCCCUUCCUGCACCCGUACCCCCGUGGUCUUUGAAUAAAGUCUGAGU GGGCGGCAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAA AAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAAA AAAAAAAAAAAAAUCUAG	

Table 6. Flagellin Amino Acid Sequences

Name	Sequence	SEQ ID NO:
ORF Sequence, AA	MAQVINTNSLSLLTQNNLNKSQSALGTAIERLSSGLRINSAKDDAAGQAIANR FTANIKGLTQASRNANDGISIAQTTEGALNEINNNLQRVRELAVQSANGTNS QSDLDSIQAEITQRLNEIDRVSGQTQFNGVKVLAQDNTLTIQVGANDGETIDI DLKEISSKTLGLDKLNVQDAYTPKETAVTVDKTTYKNGTDPITAQSNTDIQT AIGGGATGVTGADIKFKDGQYYLDVKGGASAGVYKATYDETTKKVNIDTTD KTPLATAEATAIRGTATITHNQIAEVTKEGVDTTTVAAQLAAAGVTGADKD NTSLVKLSFEDKNGKVIDGGYAVKMGDDFYAATYDEKTGAITAKTTTYTDG TGVAQTGAVKFGGANGKSEVVTATDGKTYLASDLDKHNFRTGGELKEVNT DKTENPLQKIDAALAQVDTLRSDLGAVQNRFNSAITNLGNTVNNLSSARSRI EDSDYATEVSNMSRAQILQQAGTSVLAQANQVPQNVLSLLR	89
Flagellin-GS linker-circumsporozoite protein (CSP)	MAQVINTNSLSLLTQNNLNKSQSALGTAIERLSSGLRINSAKDDAAGQAIANR FTANIKGLTQASRNANDGISIAQTTEGALNEINNNLQRVRELAVQSANSTNSQ SDLDSIQAEITQRLNEIDRVSGQTQFNGVKVLAQDNTLTIQVGANDGETIDID LKQINSQTLGLDTLNVQQKYKVSDTAATVTGYADTTIALDNSTFKASATGLG GTDQKIDGDLKFDDTTGKYYAKVTVTGGTGKDGYYEVSVDKTNGEVTLAG GATSPLTGGLPATATEDVKNVQVANADLTEAKAALTAAGVTGTASVVKMS YTDNNGKTIDGGLAVKVGDDYYSATQNKDGSISINTTKYTADDGTSKTALN KLGGADGKTEVVSIGGKTYAASKAEGHNFKAQPDLAEAAATTTENPLQKID AALAQVDTLRSDLGAVQNRFNSAITNLGNTVNNLTSARSRIEDSDYATEVSN MSRAQILQQAGTSVLAQANQVPQNVLSLLRGGGGSGGGGSMMAPDPNANP NANPNANPNANPNANPNANPNANPNANPNANPNANPNANPNANPNANPNA NPNANPNANPNANPNANPNANPNKNNQGNGQGHNMPNDPNRNVDENANA NNAVKNNNNEEPSDKHIEQYLKKIKNSISTEWSPCSVTCGNGIQVRIKPGSAN KPKDELDYENDIEKKICKMEKCSSVFNVVNS	125
Flagellin-RPVT linker-circumsporozoite protein (CSP)	MMAPDPNANPNANPNANPNANPNANPNANPNANPNANPNANPNANPNANP NANPNANPNANPNANPNANPNANPNANPNANPNKNNQGNGQGHNMPNDP NRNVDENANANNAVKNNNNEEPSDKHIEQYLKKIKNSISTEWSPCSVTCGN GIQVRIKPGSANKPKDELDYENDIEKKICKMEKCSSVFNVVNSRPVTMAQVI NTNSLSLLTQNNLNKSQSALGTAIERLSSGLRINSAKDDAAGQAIANRFTANI KGLTQASRNANDGISIAQTTEGALNEINNNLQRVRELAVQSANSTNSQSDLD SIQAEITQRLNEIDRVSGQTQFNGVKVLAQDNTLTIQVGANDGETIDIDLKQIN SQTLGLDTLNVQQKYKVSDTAATVTGYADTTIALDNSTFKASATGLGGTDQ KIDGDLKFDDTTGKYYAKVTVTGGTGKDGYYEVSVDKTNGEVTLAGGATS PLTGGLPATATEDVKNVQVANADLTEAKAALTAAGVTGTASVVKMSYTDN NGKTIDGGLAVKVGDDYYSATQNKDGSISINTTKYTADDGTSKTALNKLGG ADGKTEVVSIGGKTYAASKAEGHNFKAQPDLAEAAATTTENPLQKIDAALA QVDTLRSDLGAVQNRFNSAITNLGNTVNNLTSARSRIEDSDYATEVSNMSRA QILQQAGTSVLAQANQVPQNVLSLLR	126

EQUIVALENTS

Those skilled in the art will recognize, or be able to ascertain using no more than routine experimentation, many equivalents to the specific embodiments of the disclosure described herein. Such equivalents are intended to be encompassed by the following claims.

All references, including patent documents, disclosed herein are incorporated by reference in their entirety.

What is claimed is:

CLAIMS

1. A herpes simplex virus (HSV) vaccine, comprising:

at least one ribonucleic acid (RNA) polynucleotide having an open reading frame encoding at least one HSV antigenic polypeptide or an immunogenic fragment thereof, and a pharmaceutically acceptable carrier.

2. The HSV vaccine of claim 1, wherein the at least one antigenic polypeptide is selected from HSV-2 glycoprotein B or an immunogenic fragment thereof, HSV-2 glycoprotein C or an immunogenic fragment thereof, HSV-2 glycoprotein D or an immunogenic fragment thereof, HSV-2 glycoprotein E or an immunogenic fragment thereof, HSV-2 glycoprotein IS or an immunogenic fragment thereof, and HSV-2 ICP4 protein or an immunogenic fragment thereof.

3. The HSV vaccine of claim 1, wherein the at least one antigenic polypeptide is selected from HSV-2 glycoprotein C or an immunogenic fragment thereof, HSV-2 glycoprotein D or an immunogenic fragment thereof, and a combination of HSV-2 glycoprotein C and HSV-2 glycoprotein D or an immunogenic fragment thereof.

4. The vaccine of any one of claims 1-3, wherein the vaccine comprises at least one RNA polynucleotide having an open reading frame encoding at least two HSV antigenic polypeptides or immunogenic fragments thereof selected from HSV-2 glycoprotein B or an immunogenic fragment thereof, HSV-2 glycoprotein C or an immunogenic fragment thereof, HSV-2 glycoprotein D or an immunogenic fragment thereof, HSV-2 glycoprotein E or an immunogenic fragment thereof, HSV-2 glycoprotein IS or an immunogenic fragment thereof, and HSV-2 ICP4 protein or an immunogenic fragment thereof.

5. The vaccine of any one of claims 1-4, wherein the vaccine comprises at least two RNA polynucleotides, each having an open reading frame encoding at least one HSV antigenic polypeptide or an immunogenic fragment thereof selected from HSV-2 glycoprotein B or an immunogenic fragment thereof, HSV-2 glycoprotein C or an immunogenic fragment thereof, HSV-2 glycoprotein D or an immunogenic fragment thereof, HSV-2 glycoprotein E or an immunogenic fragment thereof, HSV-2 glycoprotein IS or an

immunogenic fragment thereof, and HSV-2 ICP4 protein or an immunogenic fragment thereof, wherein the hMPV antigenic polypeptide encoded by one of the open reading frames differs from the hMPV antigenic polypeptide encoded by another of the open reading frames.

6. The vaccine of any one of claims 1-5, wherein the at least one antigenic polypeptide comprises an amino acid sequence identified by any one of SEQ ID NO: 24-53 or 66-77.

7. The vaccine of any one of claims 1-6, wherein the at least one RNA polypeptide is encoded by a nucleic acid sequence identified by any one of SEQ ID NO: 1-23 or 54-64, and/or wherein the at least one RNA polypeptide comprises a nucleic acid sequence identified by any one of SEQ ID NO: 90-124 or comprises a fragment of a nucleic acid sequence identified by any one of SEQ ID NO: 90-124.

8. The vaccine of any one of claims 1-7, wherein the at least one antigenic polypeptide has an amino acid sequence that has at least 95% identity to an amino acid sequence identified by any one of SEQ ID NO: 24-53 or 66-77.

9. The vaccine of any one of claims 1-8, wherein the at least one antigenic polypeptide has an amino acid sequence that has 95%-99% identity to an amino acid sequence identified by any one of SEQ ID NO: 24-53 or 66-77.

10. The vaccine of any one of claims 1-8, wherein the at least one antigenic polypeptide has an amino acid sequence that has at least 90% identity to an amino acid sequence of SEQ ID NO: 24-53 or 66-77 and wherein the antigenic polypeptide or immunogenic fragment thereof has membrane fusion activity, attaches to cell receptors, causes fusion of viral and cellular membranes, and/or is responsible for binding of the virus to a cell being infected.

11. The vaccine of any one of claims 1-8, wherein the at least one antigenic polypeptide has an amino acid sequence that has 90%-99% identity to an amino acid sequence of SEQ ID NO: 24-53 or 66-77 and wherein the antigenic polypeptide or immunogenic fragment thereof has membrane fusion activity, attaches to cell receptors, causes fusion of viral and cellular membranes, and/or is responsible for binding of the virus to a cell being infected.

12. The vaccine of any one of claims 1-11, wherein the the at least one RNA polynucleotide has less than 80% identity to wild-type mRNA sequence.

13. The vaccine of any one of claims 1-11, wherein the the at least one RNA polynucleotide has at least 80% identity to wild-type mRNA sequence, but does not include wild-type mRNA sequence.

14. The vaccine of any one of claims 1-13, wherein the at least one antigenic polypeptide has membrane fusion activity, attaches to cell receptors, causes fusion of viral and cellular membranes, and/or is responsible for binding of the virus to a cell being infected.

15. The vaccine of any one of claims 1-13, wherein the at least one RNA polynucleotide comprises the at least one chemical modification.

16. The vaccine of claim 15, wherein the chemical modification is selected from pseudouridine, N1-methylpseudouridine, N1-ethylpseudouridine, 2-thiouridine, 4'-thiouridine, 5-methylcytosine, 5-methyluridine, 2-thio-1-methyl-1-deaza-pseudouridine, 2-thio-1-methyl-pseudouridine, 2-thio-5-aza-uridine, 2-thio-dihydropseudouridine, 2-thio-dihydrouridine, 2-thio-pseudouridine, 4-methoxy-2-thio-pseudouridine, 4-methoxy-pseudouridine, 4-thio-1-methyl-pseudouridine, 4-thio-pseudouridine, 5-aza-uridine, dihydropseudouridine, 5-methoxyuridine and 2'-O-methyl uridine.

17. The vaccine of claim 15 or 16, wherein the chemical modification is in the 5-position of the uracil.

18. The vaccine of any one of claims 15-17, wherein the chemical modification is a N1-methylpseudouridine or N1-ethylpseudouridine.

19. The vaccine of any one of claims 15-18, wherein at least 80% of the uracil in the open reading frame have a chemical modification.

20. The vaccine of claim 19, wherein at least 90% of the uracil in the open reading frame have a chemical modification.

21. The vaccine of claim 20, wherein 100% of the uracil in the open reading frame have a chemical modification.

22. The vaccine of any one of claims 1-21, wherein at least one RNA polynucleotide further encodes at least one 5' terminal cap.

23. The vaccine of claim 22, wherein the 5' terminal cap is 7mG(5')ppp(5')NlmpNp.

24. The vaccine of any one of claims 1-23, wherein at least one antigenic polypeptide or immunogenic fragment thereof is fused to a signal peptide selected from: a HuIgGk signal peptide (METPAQLLFLLLLWLPDTTG; SEQ ID NO: 78); IgE heavy chain epsilon-1 signal peptide (MDWTWILFLVAAATRVHS; SEQ ID NO: 79); Japanese encephalitis PRM signal sequence (MLGSNSGQRVVFTILLLLVAPAYS; SEQ ID NO: 80), VSVg protein signal sequence (MKCLLYLAFLFIGVNCA; SEQ ID NO: 81) and Japanese encephalitis JEV signal sequence (MWLVSLAIVTACAGA; SEQ ID NO: 82).

25. The vaccine of claim 24, wherein the signal peptide is fused to the N-terminus of at least one antigenic polypeptide.

26. The vaccine of claim 24, wherein the signal peptide is fused to the C-terminus of at least one antigenic polypeptide.

27. The vaccine of any one of claims 1-26, wherein the antigenic polypeptide or immunogenic fragment thereof comprises a mutated N-linked glycosylation site.

28. The vaccine of any one of claims 1-27 formulated in a nanoparticle.

29. The vaccine of claim 28, wherein the nanoparticle is a lipid nanoparticle.

30. The vaccine of claim 28 or 29, wherein the nanoparticle has a mean diameter of 50-200 nm.

31. The vaccine of claim 29 or 30, wherein the lipid nanoparticle comprises a cationic lipid, a PEG-modified lipid, a sterol and a non-cationic lipid.

32. The vaccine of claim 31, wherein the lipid nanoparticle carrier comprises a molar ratio of about 20-60% cationic lipid, 0.5-15% PEG-modified lipid, 25-55% sterol, and 25% non-cationic lipid.

33. The vaccine of claim 31 or 32, wherein the cationic lipid is an ionizable cationic lipid and the non-cationic lipid is a neutral lipid, and the sterol is a cholesterol.

34. The vaccine of any one of claims 31-33, wherein the cationic lipid is selected from 2,2-dilinoleyl-4-dimethylaminoethyl-[1,3]-dioxolane (DLin-KC2-DMA), dilinoleyl-methyl-4-dimethylaminobutyrate (DLin-MC3-DMA), and di((Z)-non-2-en-1-yl) 9-((4-(dimethylamino)butanoyl)oxy)heptadecanedioate (L319).

35. The vaccine of any one of claims 1-34, wherein the nanoparticle has a polydispersity value of less than 0.4.

36. The vaccine of any one of claims 1-35, wherein the nanoparticle has a net neutral charge at a neutral pH value.

37. The vaccine of any one of claims 1-36 further comprising an adjuvant.

38. The vaccine of claim 37, wherein the adjuvant is a flagellin protein or peptide.

39. The vaccine of claim 38, wherein the flagellin protein or peptide comprises an amino acid sequence identified by any one of SEQ ID NO: 89, 125 or 126.

40. The vaccine of any one of claims 1-39, wherein the open reading frame is codon-optimized.

41. The vaccine of any one of claims 1-40, wherein the vaccine is multivalent.

42. The vaccine of any one of claims 1-41 formulated in an effective amount to produce an antigen-specific immune response.

43. A method of inducing an antigen-specific immune response in a subject, the method comprising administering to the subject the vaccine of any one of claims 1-42 in an amount effective to produce an antigen-specific immune response in the subject.

44. The method of claim 43, wherein the antigen specific immune response comprises a T cell response or a B cell response.

45. The method of claim 43 or 44, wherein the subject is administered a single dose of the vaccine.

46. The method of claim 43 or 44, wherein the subject is administered a booster dose of the vaccine.

47. The method of any one of claims 43-46, wherein the vaccine is administered to the subject by intradermal injection or intramuscular injection.

48. The method of any one of claims 43-47, wherein an anti-antigenic polypeptide antibody titer produced in the subject is increased by at least 1 log relative to a control.

49. The method of any one of claims 43-47, wherein an anti-antigenic polypeptide antibody titer produced in the subject is increased by 1-3 log relative to a control.

50. The method of any one of claims 43-49, wherein the anti-antigenic polypeptide antibody titer produced in the subject is increased at least 2 times relative to a control.

51. The method of any one of claims 43-50, wherein the anti-antigenic polypeptide antibody titer produced in the subject is increased 2-10 times relative to a control.

52. The method of any one of claims 48-51, wherein the control is an anti-antigenic polypeptide antibody titer produced in a subject who has not been administered a vaccine against the virus.

53. The method of any one of claims 48-51, wherein the control is an anti-antigenic polypeptide antibody titer produced in a subject who has been administered a live attenuated vaccine or an inactivated vaccine against the virus.

54. The method of any one of claims 48-51, wherein the control is an anti-antigenic polypeptide antibody titer produced in a subject who has been administered a recombinant protein vaccine or purified protein vaccine against the virus.

55. The method of any one of claims 48-51, wherein the control is an anti-antigenic polypeptide antibody titer produced in a subject who has been administered a VLP vaccine against the virus.

56. The method of any one of claims 43-55, wherein the effective amount is a dose equivalent to an at least 2-fold reduction in the standard of care dose of a recombinant protein vaccine or a purified protein vaccine against the virus, and wherein an anti-antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a recombinant protein vaccine or a purified protein vaccine against the virus, respectively.

57. The method of any one of claims 43-55, wherein the effective amount is a dose equivalent to an at least 2-fold reduction in the standard of care dose of a live attenuated vaccine or an inactivated vaccine against the virus, and wherein an anti-antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a live attenuated vaccine or an inactivated vaccine against the virus, respectively.

58. The method of any one of claims 43-55, wherein the effective amount is a dose equivalent to an at least 2-fold reduction in the standard of care dose of a VLP vaccine against the virus, and wherein an anti-antigenic polypeptide antibody titer produced in the subject is equivalent to an anti-antigenic polypeptide antibody titer produced in a control subject administered the standard of care dose of a VLP vaccine against the virus.

59. The method of any one of claims 43-58, wherein the effective amount is a total dose of 50 μg-1000 μg.

60. The method of claim 59, wherein the effective amount is a dose of 25 μg, 100 μg, 400 μg, or 500 μg administered to the subject a total of two times.

61. The method of any one of claims 43-60, wherein the efficacy of the vaccine against the virus is greater than 65%.

62. The method of any one of claims 43-61, wherein the vaccine immunizes the subject against the virus for up to 2 years.

63. The method of any one of claims 43-61, wherein the vaccine immunizes the subject against the virus for more than 2 years.

64. The method of any one of claims 43-63, wherein the subject has been exposed to the virus, wherein the subject is infected with the virus, or wherein the subject is at risk of infection by the virus.

65. The method of any one of claims 43-63, wherein the subject is immunocompromised.

66. The vaccine of any one of claims 1-42 for use in a method of inducing an antigen specific immune response in a subject, the method comprising administering to the subject the vaccine in an amount effective to produce an antigen specific immune response in the subject.

67. Use of the vaccine of any one of claims 1-42 in the manufacture of a medicament for use in a method of inducing an antigen specific immune response in a subject, the method comprising administering to the subject the vaccine in an amount effective to produce an antigen specific immune response in the subject.

68. An engineered nucleic acid encoding at least one RNA polynucleotide of a vaccine of any one of claims 1-43.

69. A pharmaceutical composition for use in vaccination of a subject comprising an effective dose of mRNA encoding a herpes simplex virus (HSV) antigen,

wherein the effective dose is sufficient to produce detectable levels of antigen as measured in serum of the subject at 1-72 hours post administration.

5

70. The composition of claim 69, wherein the cut off index of the antigen is 1-2.

71. A pharmaceutical composition for use in vaccination of a subject comprising an effective dose of mRNA encoding a herpes simplex virus (HSV) antigen,

10 wherein the effective dose is sufficient to produce a 1,000- 10,000 neutralization titer produced by neutralizing antibody against said antigen as measured in serum of the subject at 1-72 hours post administration.

Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing filed or furnished:

   a. (means)

   ☐ on paper

   ☒ in electronic form

   b. (time)

   ☐ in the international application as filed

   ☒ together with the international application in electronic form

   ☐ subsequently to this Authority for the purposes of search

2. ☐ In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

   The sequence listing filed with this international application was not used for the purpose of this search and opinion.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US2016/058322

A. CLASSIFICATION OF SUBJECT MATTER

A61K 39/245 (2006.01) A61K 31/7105 (2006.01) A61K 31/7115 (2006.01) A61P 31/22 (2006.01)

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

Databases consulted: WPIAP, EPODOC, MEDLINE, HCAPLUS, EMBASE, BIOSIS, PUBMED, ESPACENET, PATENTSCOPE, internal IP Australia Databases

Search terms used: herpes simplex, HSV, herpesviridae, herpetoviridae, human herpes virus, HHV, glycoprotein, ICP4, ribonucleic acid, RNA, mRNA, vaccine, immunity, immunogen, antigen, and related terms; Modernatx, Moderna Therapeutics (applicant search); Helen Lockhart, Giuseppe Ciaramella, Shinu John, Andrew J Bett, Danilo Casimiro (inventor search).

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
	Documents are listed in the continuation of Box C	

[X] Further documents are listed in the continuation of Box C [X] See patent family annex

* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
11 January 2017	11 January 2017
Name and mailing address of the ISA/AU	**Authorised officer**
AUSTRALIAN PATENT OFFICE PO BOX 200, WODEN ACT 2606, AUSTRALIA Email address: pct@ipaustralia.gov.au	Safiea Goolam AUSTRALIAN PATENT OFFICE (ISO 9001 Quality Certified Service) Telephone No. 0262832521

INTERNATIONAL SEARCH REPORT C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		International application No. **PCT/US2016/058322**
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2013/055905 A1 (NOVARTIS AG) 18 April 2013 Abstract, [29], Table 1, claim 6, claims 16-21, [63], [79]-[83], Example 1	1-71
X	WO 2012/051211 A2 (NOVARTIS AG) 19 April 2012 Abstract, [10]-[12], [56]-[58], [87], [90]-[96], [106]-[112], [169]-[172]	1-71
X	WO 2008/011609 A2 (VICAL INCORPORATED; UNIVERSITY OF WASHINGTON) 24 January 2008 Abstract, page 1 lines 18-23, page 19 lines 1-29, page 20 lines 15-27, page 50 lines 14-22, page 78 line 30-page 80 line 19, page 81 lines 14-27, adjoining paragraph at pages 81-82, page 18 lines 15026, page 22 line 20-page 23 line 14	1-71
X	WO 2011/106607 A2 (JUVARIS BIOTHERAPEUTICS, INC.; THE TRUSTEES OF THE UNIVERSITY OF PENNSYLVANIA) 01 September 2011 Abstract, [0008]-[0011], [0064]-[0077], [0096]-[0098]	1-71
P,A	WO 2015/164674 A1 (MODERNA THERAPEUTICS, INC.) 29 October 2015 Whole document	1-71

INTERNATIONAL SEARCH REPORT Information on patent family members	International application No. **PCT/US2016/058322**

This Annex lists known patent family members relating to the patent documents cited in the above-mentioned international search report. The Australian Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

Patent Document/s Cited in Search Report		**Patent Family Member/s**	
Publication Number	**Publication Date**	**Publication Number**	**Publication Date**
WO 2013/055905 A1	18 April 2013	WO 2013055905 A1	18 Apr 2013
		AU 2012322704 A1	10 Apr 2014
		CA 2872033 A1	18 Apr 2013
		CN 104105504 A	15 Oct 2014
		EP 2768530 A1	27 Aug 2014
		JP 2015527871 A	24 Sep 2015
		MX 2014004214 A	07 May 2014
		RU 2014118727 A	20 Nov 2015
		US 2014271829 A1	18 Sep 2014
WO 2012/051211 A2	19 April 2012	WO 2012051211 A2	19 Apr 2012
		AU 2011316707 A1	09 May 2013
		AU 2016238966 A1	03 Nov 2016
		BR 112013008700 A2	21 Jun 2016
		CA 2814386 A1	19 Apr 2012
		CL 2013000984 A1	11 Oct 2013
		CN 103269713 A	28 Aug 2013
		CN 103269713 B	20 Jan 2016
		EP 2627351 A2	21 Aug 2013
		JP 2013544504 A	19 Dec 2013
		JP 2016096827 A	30 May 2016
		KR 20130117781 A	28 Oct 2013
		MX 2013003939 A	17 Oct 2013
		RU 2013121582 A	20 Nov 2014
		US 2014030292 A1	30 Jan 2014
		ZA 201302548 B	25 Sep 2014
WO 2008/011609 A2	24 January 2008	WO 2008011609 A2	24 Jan 2008
		AU 2007275047 A1	24 Jan 2008
		CA 2658484 A1	24 Jan 2008
		EP 2521786 A2	14 Nov 2012
		EP 2521786 B1	24 Jun 2015
		HK 1177952 A1	24 Mar 2016
		JP 2009544289 A	17 Dec 2009
		JP 5872755 B2	01 Mar 2016
		JP 2013138680 A	18 Jul 2013
		JP 2016052322 A	14 Apr 2016

Due to data integration issues this family listing may not include 10 digit Australian applications filed since May 2001.

INTERNATIONAL SEARCH REPORT Information on patent family members	International application No. **PCT/US2016/058322**

This Annex lists known patent family members relating to the patent documents cited in the above-mentioned international search report. The Australian Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

Patent Document/s Cited in Search Report		Patent Family Member/s	
Publication Number	**Publication Date**	**Publication Number**	**Publication Date**
		US 2008102087 A1	01 May 2008
		US 7628993 B2	08 Dec 2009
		US 2010160418 A1	24 Jun 2010
		US 7879339 B2	01 Feb 2011
		US 2010160419 A1	24 Jun 2010
		US 7935352 B2	03 May 2011
		US 2010040651 A1	18 Feb 2010
		US 8263087 B2	11 Sep 2012
		US 2010158949 A1	24 Jun 2010
		US 8293248 B2	23 Oct 2012
		US 2013273108 A1	17 Oct 2013
		US 8828408 B2	09 Sep 2014
		US 2013273107 A1	17 Oct 2013
		US 8834894 B2	16 Sep 2014
		US 2012328656 A1	27 Dec 2012
		US 8840903 B2	23 Sep 2014
		US 2013202640 A1	08 Aug 2013
		US 8840904 B2	23 Sep 2014
		US 2012328657 A1	27 Dec 2012
		US 8852610 B2	07 Oct 2014
		US 2012328658 A1	27 Dec 2012
		US 8852611 B2	07 Oct 2014
		US 2014370055 A1	18 Dec 2014
		US 9161973 B2	20 Oct 2015
		US 2015010596 A1	08 Jan 2015
		US 9205146 B2	08 Dec 2015
		US 2015010597 A1	08 Jan 2015
		US 2015030631 A1	29 Jan 2015

Due to data integration issues this family listing may not include 10 digit Australian applications filed since May 2001.

INTERNATIONAL SEARCH REPORT Information on patent family members	International application No. **PCT/US2016/058322**

This Annex lists known patent family members relating to the patent documents cited in the above-mentioned international search report. The Australian Patent Office is in no way liable for these particulars which are merely given for the purpose of information.

Patent Document/s Cited in Search Report		**Patent Family Member/s**	
Publication Number	**Publication Date**	**Publication Number**	**Publication Date**
WO 2011/106607 A2	01 September 2011	WO 2011106607 A2	01 Sep 2011
		US 2013171234 A1	04 Jul 2013
		US 9089537 B2	28 Jul 2015
WO 2015/164674 A1	29 October 2015	WO 2015164674 A1	29 Oct 2015
		AU 2015249553 A1	10 Nov 2016
		CA 2946751 A1	29 Oct 2015
		SG 11201608798Y A	29 Nov 2016
		US 2016317647 A1	03 Nov 2016
		US 2016331828 A1	17 Nov 2016

End of Annex

Due to data integration issues this family listing may not include 10 digit Australian applications filed since May 2001.