Pacific island of Yap, and then disseminated from island to island across the Pacific, leading to an extensive outbreak in 2013-2014 in French Polynesia, spreading then to New Caledonia, the Cook Islands, and ultimately, to Easter Island. An Asian lineage virus was subsequently transferred to the Western Hemisphere by routes that remain undetermined (Faria *et al.* (2016) Science. 352(6283):345-349). The virus may be transmitted zoonotically by Aedes aegypti, A. albopictus, and possibly by A. hensilli and A. polynieseinsis (Weaver *et al*. (2016) Antiviral Res. 130:69-80). Additionally, it is thought that other vectors for transmitting the virus may exist, and the virus may be transmitted by blood transfusion, transplacentally, and/or through sexual transmission.

[0005] In late 2015, a significant increase in fetal abnormalities (e.g., microcephaly) and Guillain-Barre syndrome (GBS) in areas of widespread Zika virus infection raised alarm that Zika virus might be much more virulent than originally thought, prompting the World Health Organization (WHO) to declare a Public Health Emergency of International Concern (PHEIC) (Heymann *et al.* (2016) Lancet 387(10020): 719-21). Although the WHO has since declared an end to the PHEIC, Zika continues to pose in particular a significant threat for pregnant women and their unborn babies.

[0006] While Zika virus poses a substantial public health threat, no FDA-approved vaccine or treatment currently exists, and the only preventative measures for controlling Zika virus involve managing mosquito populations.

[0007] In recent efforts to characterize a recombinant Zika virus for the development of a potential vaccine, a non-human cell adapted Zika virus was identified that harbors a mutation in the viral Envelope protein at position 330 (Weger-Lucarelli *et al.* (2017) Journal of Virology 91(1): 1-10). The authors of this study found that full-length infectious cDNA clones of Zika virus strain PRVABC59 were genetically unstable when amplified during cloning, and opted to split the viral genome to address the observed instability, developing and applying a two plasmid system. However, a two plasmid system for the development of a Zika vaccine is less desirable.

SUMMARY

[0008] Thus, there is a need to develop vaccines and immunogenic compositions for treating and/or preventing Zika virus. In one aspect, the present disclosure refers to the use of a vaccine or immunogenic composition inducing a seroconversion rate of at least 60% in

a population and comprising a dose of 1 µg to 40 µg of one antigen from a Zika virus in the manufacture of a medicament for preventing Zika virus infection in a human subject in need thereof, or inducing an immune response against a Zika antigen in a human subject in need thereof, wherein the antigen is an inactivated whole virus and the use comprises that the medicament is to be administered to the human subject. In another aspect, the present disclosure refers to the use of a vaccine or immunogenic composition inducing a seroconversion rate of at least 60% in a population and comprising a dose of 1 µg to 40 µg of one antigen from a Zika virus in the manufacture of a medicament for preventing Zika virus disease in a fetus or newborn in need thereof, wherein the antigen is an inactivated whole virus and the use comprises that the medicament is to be administered to a pregnant human subject or a human subject that intends to become pregnant or woman of childbearing potential. Accordingly, certain examples of the present disclosure relate a vaccine or immunogenic composition comprising a dose of 1µg to 40µg of one antigen from a Zika virus, wherein the antigen is an inactivated whole virus.

[0009] Accordingly, certain examples of the present disclosure relate a vaccine or immunogenic composition comprising a dose of 1µg to 40µg of one antigen from a Zika virus, wherein the Zika virus comprises at least one non-human cell adaptation mutation.

[0010] Accordingly, certain examples of the present disclosure relate a vaccine or immunogenic composition comprising a dose of 1µg to 40µg of one antigen from a Zika virus, the Zika virus having a mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1.

[0011] To meet the above and other needs, the present disclosure is directed, at least in part, to a genetically stable non-human cell adapted Zika virus harboring an adaptation in the Non-structural protein 1 (with a wild-type Envelope protein), allowing for the use of a single virus/viral genome system for vaccine production. The present disclosure is also directed, at least in part, to a Zika virus clonal isolate that is genetically homogenous and/or has been purified away from one or more adventitious agents. Accordingly, the present disclosure provides vaccines and immunogenic compositions useful for treating and/or preventing Zika virus infection (e.g., in humans) that include one or more Zika virus antigens (e.g., one or more antigens from a whole inactivated Zika virus) from a Zika virus harboring at least one non-human cell adaptation mutation (e.g., a mutation in Zika virus Non-structural protein 1) and/or a Zika virus clonal isolate.

[0012] The present disclosure is based, at least in part, on the surprising finding that both high and low dose vaccines comprising one or more antigens from separately derived clonal virus populations of non-human cell adapted Zika virus were able to induce robust immune responses and provide significant protection from Zika virus infection (See Examples 2 and Example 4 below). Clonal isolation of the Zika virus strains also allowed for: 1) the successful purification of the virus away from contaminating agents (e.g., adventitious agents that may be co-purified with the parental strain), and 2) the production of a genetically homogeneous viral population. Moreover, the present disclosure is based, at least in part, on the finding that clonal isolated Zika viruses harboring an adaptation mutation in protein NS1 grew well and predictably in Vero cells to high titer, and surprisingly, were genetically stable/genetically homogenous without any detectable mutations in the viral envelope protein (See Examples 1 and 2 below). While a similar mutation in Zika virus Non-structural protein 1 may have been observed in the genomic sequencing analysis of 1 out of 3 published sequences of Zika virus strain PRVABC59 (Yun et al. Genome Announc. 2016 Aug 18;4(4)), this reference fails to teach or suggest that a mutation in NS1 may improve stability of the virus; fails to teach or suggest that a virus harboring the mutation may be used in the development of an effective vaccine against Zika virus; and fails to teach or suggest that such a vaccine may be effective in inducing a robust immune response and providing significant protection from Zika virus infection when used at both low and high doses. Thus, without wishing to be bound by theory, the inventors of the present disclosure have determined that the adaptation mutation in protein NS1 appeared to enhance genetic stability within the Zika virus, resulting in increased/enhanced replication efficiency. Further, the Zika strain harboring an adaptation mutation in protein NS1 was able to be passaged multiple times without developing further mutations. Such a stable Zika virus strain is advantageous as a master virus seed (MVS), or subsequent seeds derived from the MVS, for vaccine production and manufacturing, as the risk of the master virus seed developing undesirable mutations is reduced. Moreover, without wishing to be bound by theory, the adaptation mutation in protein NS1 of the Zika strain of the present disclosure may also reduce or otherwise inhibit the occurrence of undesirable mutations, such as a mutation within the envelope protein E (Env) of the Zika virus strain.

[0013] Accordingly, certain examples of the present disclosure relate to a vaccine or immunogenic composition containing one or more antigen from a Zika virus, where the

Zika virus contains at least one non-human cell adaptation mutation. In some examples, the at least one non-human cell adaptation mutation is in Zika virus Non-structural protein 1 (NS1). In some examples, the at least one adaptation mutation occurs at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1. In some examples, the at least one adaptation mutation is a Trp98Gly mutation.

[0014] Accordingly, certain examples of the present disclosure relate a vaccine or immunogenic composition comprising a dose of 1µg to 40µg of one antigen from a Zika virus, wherein the Zika virus comprises at least one non-human cell adaptation mutation.

[0015] Accordingly, certain examples of the present disclosure relate a vaccine or immunogenic composition comprising a dose of 1µg to 40µg of one antigen from a Zika virus, the Zika virus having a mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1.

[0016] In some examples that may be combined with any of the preceding examples, the at least one adaptation mutation enhances genetic stability as compared to a Zika virus lacking the at least one adaptation mutation. In some examples that may be combined with any of the preceding examples, the at least one adaptation mutation enhances viral replication as compared to a Zika virus lacking the at least one adaptation mutation. In some examples that may be combined with any of the preceding examples, the Zika virus does not comprise a mutation in Envelope protein E (Env).

[0017] In some examples that may be combined with any of the preceding examples, the non-human cell is a mammalian cell. In some examples that may be combined with any of the preceding examples, the non-human cell is a monkey cell. In some examples, the monkey cell is from a Vero cell line. In some examples, the Vero cell line is a WHO Vero 10-87 cell line.

[0018] In some examples that may be combined with any of the preceding examples, the Zika virus is an African lineage virus or an Asian lineage virus. In some examples, the Zika virus is an Asian lineage virus. In some examples, the Zika virus is from strain PRVABC59.

[0019] In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition is a purified antigen vaccine or immunogenic

composition, a subunit vaccine or immunogenic composition, an inactivated whole virus vaccine or immunogenic composition, or an attenuated virus vaccine or immunogenic composition. In some examples, the vaccine or immunogenic composition is an inactivated whole virus vaccine or immunogenic composition. In some examples, the vaccine or immunogenic composition comprises a purified inactivated whole Zika virus. In some examples, the vaccine or immunogenic composition comprises a purified inactivated whole Zika virus comprising a mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1. In some examples, the vaccine or immunogenic composition comprises a purified inactivated whole Zika virus comprising a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1. In some examples, the vaccine or immunogenic composition comprises a purified inactivated whole Zika virus comprising a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO:1, wherein the Zika virus is derived from strain PRVABC59. In some examples, the vaccine or immunogenic composition comprises a purified inactivated whole Zika virus comprising a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO:1, wherein the Zika virus is derived from strain PRVABC59 comprising the genomic sequence according to SEQ ID NO:2. In some examples, the vaccine or immunogenic composition comprises a purified inactivated whole Zika which differs from strain PRVABC59 in a Trp98Gly mutation at position 98 of SEQ ID NO: 1.

[0020] In some examples that may be combined with any of the preceding examples, the virus was chemically inactivated. In some examples, the virus was chemically inactivated with one or more of a detergent, formalin, beta-propiolactone (BPL), binary ethylamine (BEI), acetyl ethyleneimine, methylene blue, and psoralen. In some examples, the virus was chemically inactivated with formalin.

[0021] In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition further contains an adjuvant. In some examples, the adjuvant is selected from aluminum salts, toll-like receptor (TLR) agonists, monophosphoryl lipid A (MLA), synthetic lipid A, lipid A mimetics or analogs, MLA derivatives, cytokines, saponins, muramyl dipeptide (MDP) derivatives, CpG oligos, lipopolysaccharide (LPS) of gram-negative bacteria, polyphosphazenes, emulsions,

virosomes, cochleates, poly(lactide-co-glycolides) (PLG) microparticles, poloxamer particles, microparticles, liposomes, Complete Freund's Adjuvant (CFA), and/or Incomplete Freund's Adjuvant (IFA). In some examples, the adjuvant is an aluminum salt. In some examples, the adjuvant is selected from the group consisting of alum, aluminum phosphate, aluminum hydroxide, potassium aluminum sulfate, and Alhydrogel 85. In some examples, at least 75%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% of the one or more antigens are adsorbed to the adjuvant.

[0022] In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition is a low or medium dose vaccine or immunogenic composition (e.g., containing from about 1µg to about 5µg antigen or 5µg or antigen 2µg antigen or 5µg antigen). In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition is a high dose vaccine or immunogenic composition (e.g., containing about 10µg antigen). In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition contains from about 1 µg to about 25 µg of the one or more antigens, in particular 2 µg, 5 µg or 10 µg, or in particular 10 µg of the one or more antigens. In certain such examples the antigen is a purified inactivated whole virus, such as a Zika virus with a mutation which is a tryptophan to glycine substitution at position 98 of SEQ ID NO:1 or at a position corresponding to position 98 of SEQ ID NO: 1 as described herein. In some examples, the vaccine or immunogenic composition comprises a purified inactivated whole Zika virus comprising a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO:1, wherein the Zika virus is derived from strain PRVABC59. In some examples, the vaccine or immunogenic composition comprises a purified inactivated whole Zika virus comprising a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO:1, wherein the Zika virus is derived from strain PRVABC59 comprising the genomic sequence according to SEQ ID NO:2. In certain such examples the Zika virus is a plaque purified clonal Zika virus isolate. In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition contains from about 0.1 µg to about 100 µg Zika virus antigen or Env. In some examples, the vaccine or immunogenic composition is unadjuvanted. In some examples that may be combined with any of the preceding examples, the Zika virus is a clonal isolate. In some examples, the clonal isolate

is substantially free of one or more adventitious agents (e.g., free of one or more adventitious agents that may be co-purified with the parental strain).

[0023] Other examples of the present disclosure relate to a vaccine comprising a Zika virus having a mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1. In some examples, the vaccine comprises a purified inactivated whole Zika virus comprising a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO:1, wherein the Zika virus is derived from strain PRVABC59. In some examples, the vaccine comprises a purified inactivated whole Zika virus comprising a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO:1, wherein the Zika virus is derived from strain PRVABC59 comprising the genomic sequence according to SEQ ID NO:2. In certain such examples the Zika virus is a plaque purified clonal Zika virus isolate.

[0024] Other examples of the present disclosure relate to a vaccine or immunogenic composition containing: a) an aluminum salt adjuvant; and b) a purified inactivated whole Zika virus, where the Zika virus contains a non-human cell adaptation mutation, and where the non-human cell adaptation mutation is a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1.

[0025] Other examples of the present disclosure relate to a method of treating or preventing, in particular preventing Zika virus infection in a subject in need thereof, including administering to the subject a therapeutically effective amount of any of the vaccines or immunogenic compositions described herein.

[0026] Other examples of the present disclosure relate to a method for inducing an immune response in a subject in need thereof, including administering to the subject an immunogenic amount of any of the vaccines or immunogenic compositions described herein.

[0027] In one example the present disclosure relates to a method of treating or preventing, in particular preventing Zika virus infection in a subject in need thereof, comprising administering to the subject the vaccine or immunogenic composition.

[0028] In one example the present disclosure relates to a method for inducing an immune response against a Zika virus antigen in a subject in need thereof, comprising administering to the subject the vaccine or immunogenic composition.

[0029] In one example the present disclosure relates to a method of preventing Zika virus disease in a subject in need thereof, comprising administering to the subject the vaccine or immunogenic composition. In this case the disease relates to mild fever, maculopapular rash, conjunctivitis and arthralgia. Furthermore the Zika virus is a neurotropic flavivirus that can potentially cause disease within the central nervous system and Guillain-Barré Syndrome (GBS).

[0030] In one example the present disclosure relates to a method of preventing Zika virus disease in a fetus or newborn in need thereof, comprising administering to the pregnant subject or a subject that intends to become pregnant or woman of childbearing potential the vaccine or immunogenic composition. The Zika disease in this case relates to serious outcomes for the fetus and newborn. The spectrum of congenital anomalies associated with Zika virus infection, known as Congenital Zika Syndrome (CZS), consists of severe microcephaly with partially collapsed skull, cerebral cortices with subcortical calcifications, macular scarring and focal pigmentary retinal mottling, congenital contractures, and marked early hypertonia with symptoms of extrapyramidal involvement.

[0031] In one example the present disclosure relates to a vaccine or immunogenic composition of for use in a method of treating or preventing, in particular preventing Zika virus infection in a subject in need thereof, in a method for inducing an immune response against a Zika virus antigen in a subject in need thereof, and in a method of preventing Zika virus disease in a subject, fetus or newborn in need thereof.

[0032] In one example the present disclosure relates to the use of the vaccine or immunogenic composition in the manufacture of a medicament for a method of treating or preventing, in particular preventing Zika virus infection in a subject in need thereof, a method for inducing an immune response in a subject in need thereof, and for a method of preventing Zika virus disease in a subject fetus or newborn in need thereof.

[0033] Accordingly, certain examples of the present disclosure relates to a method of treating or preventing, in particular preventing Zika virus infection in a subject population in need thereof, comprising administering to individual subjects of said subject population a

vaccine or immunogenic composition comprising a Zika antigen, wherein the vaccine or immunogenic composition is administered as a first (prime) and a second (boost) administration about 1 to about 16 weeks apart, and wherein the administration of the vaccine or immunogenic composition induces 14 and/or 28 days after the boost administration geometric mean neutralizing antibodies titers in a population of at least 20 flavivirus naïve subjects and/or at least 20 Zika virus seronegative subjects of greater than 300, or greater than 500, or greater than 1000, or greater than 1500, or greater than 2000, or greater than 3000, as determined by the plaque reduction neutralization test (PRNT).

[0034] Accordingly, certain examples of the present disclosure relates to a method of treating or preventing, in particular preventing Zika virus infection in a subject population in need thereof, comprising administering to individual subjects of said subject population a vaccine or immunogenic composition comprising one antigen from a Zika virus, wherein the vaccine or immunogenic composition is administered as a first (prime) and a second (boost) administration about 1 to about 16 weeks apart, and wherein the administration of the vaccine or immunogenic composition induces 28 days after the boost administration geometric mean neutralizing antibodies titers in a population of at least 20 flavivirus naïve subjects and/or at least 20 Zika virus seronegative subjects, which are at least 10 times, or at least 15 times, or at least 20 times, or at least 25 times higher than the geometric mean neutralizing antibodies titers induces 28 days after the prime administration, as determined by the plaque reduction neutralization test (PRNT). The boost administration thus provides for very high geometric mean neutralizing antibodies titers responsible for a long term protection

[0035] Accordingly, certain examples of the present disclosure relate a method of treating or preventing, in particular preventing Zika virus infection in a subject population in need thereof, comprising administering to individual subjects of said subject population a vaccine or immunogenic composition comprising one antigen from a Zika virus, wherein the vaccine or immunogenic composition is administered as a single dose or prime administration, and wherein the administration of the vaccine or immunogenic composition induces 14 and/or 28 days after the single dose or prime administration geometric mean neutralizing antibodies titers in a population of at least 20 flavivirus naïve subjects and/or at least 20 Zika virus seronegative subjects of greater than 10, or greater than 50, or greater than 100, or greater than 200, or greater than 250, as determined by the plaque

reduction neutralization test (PRNT). The high geometric mean neutralizing antibodies titers indicate an early onset of protection which is beneficial in an outbreak situation or a traveler visiting an endemic area within a short period of time from the administration of the vaccine or immunogenic composition.

[0036] Within the meaning of this disclosure PRNT refers to Zika virus Neutralizing antibody titers determined by a plaque reduction neutralization test (PRNT) as described previously (See Sun, W. et al. Protection of Rhesus monkeys against dengue virus challenge after tetravalent live attenuated dengue virus vaccination. J. Infect. Dis. 193, 1658–1665 (2006). Muthumani K, Griffin BD, Agarwal S, et al. In vivo protection against ZIKV infection and pathogenesis through passive antibody transfer and active immunisation with a prMEnv DNA vaccine. NPJ Vaccines 2016; 1: 16021). The Zika strain used for PRNT assay development was PRVABC59.

[0037] Within the meaning of this disclosure seropositivity is defined as titer ≥ 10 as determined by the plaque reduction neutralization test (PRNT); Zika virus seronegative subjects (titer <10) as determined by the plaque reduction neutralization test (PRNT), Seroconversion is defined as: Zika virus seronegative subjects (titer <10) have titer ≥10 post-vaccination as determined by the plaque reduction neutralization test (PRNT); Results < 10 as determined by the plaque reduction neutralization test (PRNT) are assigned a titer of 5; Titers ≥ 10 (limit of detection) and < 26 (lower limit of quantification) as determined by the plaque reduction neutralization test (PRNT) are assigned a value of 13.

[0038] Flavivirus naïve subjects for the present disclosure are defined to be subjects without detectable serum antibodies against a panel of flaviviruses, as measured by a reactive antibody based assay (Luminex). Flavivirus screening assay is based on a luminex platform to simultaneously detect multiple target antigens in the same sample. This bead based assay is sensitive, specific and reproducible. For ZIK101, the antigens targeted are Zika, Dengue, Yellow fever, JEV, USUV, SLEV and WNV. Due to cross reactivity among Flaviviruses, the current antigen set would help detect any prior Flavivirus exposure. References for luminex concept are: Dias D, Van Doren J, Schlottmann S, Kelly S, Puchalski D, Ruiz W, Boerckel P, Kessler J, Antonello JM, Green T, Brown M, Smith J, Chirmule N, Barr E, Jansen KU, Esser MT. 2005. Optimization and validation of a multiplexed Luminex assay to quantify antibodies to neutralizing epitopes on human

papillomaviruses 6, 11, 16, and 18. Clin. Diagn. Lab. Immunol. 12:959–969 [PMC free article] [PubMed]. Ayouba A et al Development of a Sensitive and Specific Serological Assay Based on Luminex Technology for Detection of Antibodies to Zaire Ebola Virus. J Clin Microbiol. 2017 Dec 28;55(1):165-176. doi: 10.1128/JCM.01979-16.

[0039] Witin the meaning of this disclosure endemic is defined as areas with risk of infection as defined by the Centers for Disease Control and Prevention, such as for example as of March 2018, namely: Asia: Bangladesh, Burma (Myanmar), Cambodia, India, Indonesia, Laos, Malaysia, Maldives, Pakistan, Philippines, Singapore, Thailand, Timor-Leste (East Timor), Vietnam.The Pacific Islands: Fiji, Marshall Islands, Papua New Guinea, Samoa, Solomon Islands, Tonga. The Caribbean: Anguilla; Antigua and Barbuda; Aruba; Barbados; Bonaire; British Virgin Islands; Cuba; Curaçao; Dominica; Dominican Republic; Grenada; Haiti; Jamaica; Montserrat; the Commonwealth of Puerto Rico, a US territory; Saba; Saint Kitts and Nevis; Saint Lucia; Saint Martin; Saint Vincent and the Grenadines; Sint Eustatius; Sint Maarten; Trinidad and Tobago; Turks and Caicos Islands; US Virgin Islands. North America: Mexico Central America: Belize, Costa Rica, El Salvador, Guatemala, Honduras, Nicaragua, Panama South America: Argentina, Bolivia, Brazil, Colombia, Ecuador, French Guiana, Guyana, Paraguay, Peru, Suriname, Venezuela Africa: Angola, Benin, Burkina-Faso, Burundi, Cameroon, Cape Verde, Central African Republic, Chad, Congo (Congo-Brazzaville), Côte d'Ivoire, Democratic Republic of the Congo (Congo-Kinshasa), Equatorial Guinea, Gabon, Gambia, Ghana, Guinea, Guinea-Bissau, Kenya, Liberia, Mali, Niger, Nigeria, Rwanda, Senegal, Sierra Leone, South Sudan, Sudan, Tanzania, Togo, Uganda. This areas may change.

[0040] Accordingly, certain examples of the present disclosure relate to a method for inducing an immune response in a subject population in need thereof, comprising administering to individual subjects of said subject population a vaccine or immunogenic composition comprising a Zika antigen, wherein the vaccine or immunogenic composition is administered as a first (prime) and a second (boost) administration about 1 to about 16 weeks apart, and wherein the administration of the vaccine or immunogenic composition induces 14 and/or 28 days after the boost administration geometric mean neutralizing antibodies titers in a subject population of at least 20 flavivirus naïve subjects and/or at least 20 Zika virus seronegative subjects of greater than 300, or greater than 500, or greater than 1000, or greater than 1500, or greater than 2000, or greater than 3000, or

greater than 5000, or greater than 10,000, as determined by the reporter virus particle neutralization assay (RVP).

[0041] Accordingly, certain examples of the present disclosure relate to a method of treating or preventing, in particular preventing Zika virus infection in a subject population in need thereof, comprising administering to individual subjects of said subject population a vaccine or immunogenic composition comprising one antigen from a Zika virus, wherein the vaccine or immunogenic composition is administered as a single dose or prime administration, and wherein the administration of the vaccine or immunogenic composition induces 14 and/or 28 days after the single dose or prime administration geometric mean neutralizing antibodies titers in a population of at least 20 flavivirus naïve subjects and/or at least 20 Zika virus seronegative subjects of greater than 300, or greater than 500, greater than 1000, or greater than 2000, as determined by the reporter virus particle neutralization assay (RVP). The high geometric mean neutralizing antibodies titers indicate an early onset of protection which is beneficial in an outbreak situation or a traveler visiting an endemic area within a short period of time from the administration of the vaccine or immunogenic composition.

[0042] Within the meaning of this disclosure Reporter virus particle (RVP) neutralization assay refers to Zika Neutralizing antibody titers were analyzed by titration of serum samples with a constant amount of Zika RVPs in Vero cells grown in 96-well plates. RVPs contained the prME proteins of Zika (strain SPH2012) and a Dengue-based Renilla luciferase reporter. Briefly, sera were heat inactivated at 56°C for 30 min, diluted, and then incubated at 37°C with RVPs. The serum/RVP mixture was then mixed with Vero cells and incubated for 72 hours at 37°C ± 2°C/ 5% CO2 before detection with luciferase substrate. Data was analyzed using JMP11 non-linear 4 parameter analysis, normalized to a positive tracking control and effective dose 50% (EC50) was reported.

[0043] Accordingly, certain examples of the present disclosure relate a method of treating or preventing, in particular preventing Zika virus infection in a subject population in need thereof, comprising administering to individual subjects of said subject population a vaccine or immunogenic composition comprising a Zika antigen, wherein the vaccine or immunogenic composition is administered as a first (prime) and a second (boost) administration about 1 to about 16 weeks apart and wherein the administration of the vaccine or immunogenic composition induces 14 and/or 28 days after the boost

administration a seroconversion rate of 100% in a population of at least 20 seronegative subjects.

[0044] Accordingly, certain examples of the present disclosure relate to a method of treating or preventing, in particular preventing Zika virus infection in a subject population in need thereof, comprising administering to individual subjects of said subject population a vaccine or immunogenic composition comprising one antigen from a Zika virus, wherein the vaccine or immunogenic composition is administered as single dose or prime administration and wherein the administration of the vaccine or immunogenic composition induces 14 and/or 28 days after the single dose or prime administration a seroconversion rate of 60%, 70%, 80% or 90% in a population of at least 20 Zika virus seronegative subjects. The high seroconversion rate indicates an early onset of protection, which is beneficial in an outbreak situation or a traveler visiting an endemic area within a short period of time from the administration of the vaccine or immunogenic composition.

[0045] The above methods are to be understood to also relate to corresponding uses of a vaccine or immunogenic composition comprising one antigen from a Zika virus as disclosed herein for the manufacture of a medicament for the treating or preventing Zika virus infection.

[0046] The above methods are to be understood to also relate to a vaccine or immunogenic composition comprising one antigen from a Zika virus as disclosed herein for use the treating or preventing Zika virus infection.

[0047] In some examples the administration is intramuscular or subcutaneous. In some examples the administration includes the administration of two doses of the vaccine or immunogenic composition as described herein (e.g. 10 µg purified inactivated whole virus, such as a Zika virus with a mutation which is a tryptophan to glycine substitution at position 98 of SEQ ID NO:1 or at a position corresponding to position 98 of SEQ ID NO: 1 as described herein) given about 1 to about 16 weeks apart (first (prime) and a second (boost) administration). In certain such examples the Zika virus is a plaque purified clonal Zika virus isolate.

[0048] Other examples of the present disclosure relate to the vaccines or immunogenic compositions as described herein for use in treating or preventing Zika virus infection in a subject in need thereof, for use in inducing an immune response in a subject in need thereof

and for use in preventing Zika virus disease in a subject, fetus or newborn in need thereof. In some examples the administration is intramuscular or subcutaneous. In some examples the administration includes the administration of two doses of the vaccine or immunogenic composition as described herein (e.g. 10 µg purified inactivated whole virus, such as a Zika virus with a mutation which is a tryptophan to glycine substitution at position 98 of SEQ ID NO:1 or at a position corresponding to position 98 of SEQ ID NO: 1 as described herein) given about 1 to about 16 weeks apart (first (prime) and a second (boost) administration). In certain such examples the Zika virus is a plaque purified clonal Zika virus isolate.

[0049] Other examples of the present disclosure relate to the use of the vaccines or immunogenic compositions as described herein in the manufacture of a medicament for treating or preventing Zika virus infection in a subject in need thereof for inducing an immune response in a subject in need thereof and in preventing Zika virus disease in a subject, fetus or newborn in need thereof. In some examples the administration is intramuscular or subcutaneous. In some examples the administration includes the administration of two doses of the vaccine or immunogenic composition as described herein (e.g. 10 µg purified inactivated whole virus, such as a Zika virus with a mutation which is a tryptophan to glycine substitution at position 98 of SEQ ID NO:1 or at a position corresponding to position 98 of SEQ ID NO: 1 as described herein) given about 1 to about 16 weeks apart (first (prime) and a second (boost) administration). In certain such examples the Zika virus is a plaque purified clonal Zika virus isolate.

[0050] In some examples that may be combined with any of the preceding examples, the subject is a human. In some examples, the subject is pregnant or intends to become pregnant or woman of childbearing potential.

[0051] In some examples that may be combined with any of the preceding examples, administration of the vaccine or immunogenic composition induces a protective immune response in the subject. In some examples, the protective immune response induced in the subject is greater than a protective immune response induced in a corresponding subject administered a vaccine or immunogenic composition containing one or more antigens from a Zika virus lacking the at least one non-human cell adaptation mutation. In some examples that may be combined with any of the preceding examples, administration of the vaccine or immunogenic composition induces the generation of neutralizing antibodies to Zika virus in the subject. In some examples, the concentration of neutralizing antibodies generated in the

subject is higher than a concentration of neutralizing antibodies generated in a corresponding subject administered a vaccine or immunogenic composition comprising one or more antigens from a Zika virus lacking the at least one non-human cell adaptation mutation.

[0052] In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition is administered by a route selected from subcutaneous administration, transcutaneous administration, intradermal administration, subdermal administration, intramuscular administration, peroral administration, intranasal administration, buccal administration, intraperitoneal administration, intravaginal administration, anal administration and/or intracranial administration. In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition is administered one or more times. In some examples, the vaccine or immunogenic composition is administered as a first (prime) and a second (boost) administration. In some examples, the second (boost) administration is administered at least 28 days after the first (prime) administration. In some examples the administration includes the administration of two doses of the vaccine or immunogenic composition as described herein (e.g. 10 µg purified inactivated whole virus, such as a Zika virus with a mutation which is a tryptophan to glycine substitution at position 98 of SEQ ID NO:1 or at a position corresponding to position 98 of SEQ ID NO: 1 as described herein) given about 1 to about 16 weeks apart (first (prime) and a second (boost) administration). In certain such examples the Zika virus is a plaque purified clonal Zika virus isolate.

[0053] In some examples that may be combined with any of the preceding examples, the virus preparation is mixed with an adjuvant. In some examples, the adjuvant is selected from aluminum salts, toll-like receptor (TLR) agonists, monophosphoryl lipid A (MLA), synthetic lipid A, lipid A mimetics or analogs, MLA derivatives, cytokines, saponins, muramyl dipeptide (MDP) derivatives, CpG oligos, lipopolysaccharide (LPS) of gram-negative bacteria, polyphosphazenes, emulsions, virosomes, cochleates, poly(lactide-co-glycolides) (PLG) microparticles, poloxamer particles, microparticles, liposomes, Complete Freund's Adjuvant (CFA), and/or Incomplete Freund's Adjuvant (IFA). In some examples, the adjuvant is an aluminum salt. In some examples, the adjuvant is selected from alum, aluminum phosphate, aluminum hydroxide, potassium aluminum sulfate, and/or Alhydrogel 85. In some examples, at least 75%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% of one or more antigens in the virus preparation are adsorbed to the adjuvant.

[0054] In some examples that may be combined with any of the preceding examples, the at least one non-human cell adaptation mutation is in Zika virus NS1. In some examples, the at least one adaptation mutation occurs at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1. In some examples, the at least one adaptation mutation is a Trp98Gly mutation. In some examples that may be combined with any of the preceding examples, the at least one adaptation mutation enhances genetic stability as compared to a Zika virus lacking the at least one adaptation mutation. In some examples that may be combined with any of the preceding examples, the at least one adaptation mutation enhances viral replication as compared to a Zika virus lacking the at least one adaptation mutation. In some examples that may be combined with any of the preceding examples, the Zika virus does not comprise a mutation in Envelope protein E (Env).

[0055] In some examples that may be combined with any of the preceding examples, the population of Zika viruses is heterogeneous. In some examples that may be combined with any of the preceding examples, the population of Zika viruses comprises a Zika virus clinical isolate. In some examples, the Zika virus clinical isolate is from strain PRVABC59. In some examples that may be combined with any of the preceding examples, the population of Zika viruses comprises a Zika virus that has been previously passaged one or more times in cell culture. In some examples that may be combined with any of the preceding examples, the inoculum comprises human serum. In some examples that may be combined with any of the preceding examples, the inoculum comprises one or more adventitious agents. In some examples, the Zika virus clonal isolate is substantially free of the one or more adventitious agents.

[0056] In some examples that may be combined with any of the preceding examples, the methods further include one or more additional plaque purifications of the Zika virus clonal isolate. In some examples, the Zika virus clonal isolate is further plaque purified two or more times. In some examples that may be combined with any of the preceding examples, the methods further include passaging the Zika virus clonal isolate one or more times in cell culture. In some examples, the Zika virus clonal isolate is passaged two or more times.

[0057] In some examples that may be combined with any of the preceding examples, the methods further include formulating a vaccine or immunogenic composition comprising one or more antigens from the Zika virus clonal isolate. In some examples, the vaccine or immunogenic composition is a purified antigen vaccine or immunogenic composition, a subunit vaccine or immunogenic composition, an inactivated whole virus vaccine or immunogenic composition, or an attenuated virus vaccine or immunogenic composition. In some examples, the vaccine or immunogenic composition is a purified inactivated whole

virus vaccine or immunogenic composition. In some examples, the Zika virus clonal isolate was chemically inactivated. In some examples, the Zika virus clonal isolate was chemically inactivated with one or more of a detergent, formalin, beta-propiolactone (BPL), binary ethylamine (BEI), acetyl ethyleneimine, methylene blue, and psoralen. In some examples, the Zika virus clonal isolate was chemically inactivated with formalin.

[0058] In some examples that may be combined with any of the preceding examples, the methods further include admixing the vaccine or immunogenic composition with an adjuvant. In some examples, the adjuvant is selected from aluminum salts, toll-like receptor (TLR) agonists, monophosphoryl lipid A (MLA), synthetic lipid A, lipid A mimetics or analogs, MLA derivatives, cytokines, saponins, muramyl dipeptide (MDP) derivatives, CpG oligos, lipopolysaccharide (LPS) of gram-negative bacteria, polyphosphazenes, emulsions, virosomes, cochleates, poly(lactide-co-glycolides) (PLG) microparticles, poloxamer particles, microparticles, liposomes, Complete Freund's Adjuvant (CFA), and Incomplete Freund's Adjuvant (IFA). In some examples, the adjuvant is an aluminum salt. In some examples, the adjuvant is selected from alum, aluminum phosphate, aluminum hydroxide, potassium aluminum sulfate, and Alhydrogel 85. In some examples, at least 75%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% of the one or more antigens are adsorbed to the adjuvant. In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition comprises 1 µg to about 40 µg of the purified inactivated whole virus, or 1 µg to about 30 µg of the purified inactivated whole virus, or 1 µg to about 20 µg of the purified inactivated whole virus, in particular 2 µg, or 5 µg, or 10 µg, or 15 µg or 20 µg or in particular 10 µg purified inactivated whole virus , such as a Zika virus with a mutation which is a tryptophan to glycine substitution at position 98 of SEQ ID NO:1 or at a position corresponding to position 98 of SEQ ID NO: 1 as described herein. In some examples, the vaccine or immunogenic composition comprises 1 µg to about 30 µg of the purified inactivated whole virus, in particular 2 µg, 5 µg or 10 µg, or in particular 10 µg of a purified inactivated whole Zika virus comprising a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO:1, wherein the Zika virus is derived from strain PRVABC59. In some examples, the vaccine or immunogenic composition comprises 1 µg to about 30 µg of the purified inactivated whole virus, in particular 2 µg, 5 µg or 10 µg, or in particular 10 µg of a purified inactivated whole Zika virus comprising a Trp98Gly mutation at position 98 of SEQ ID NO: 1, or at a

position corresponding to position 98 of SEQ ID NO:1, wherein the Zika virus is derived from strain PRVABC59 comprising the genomic sequence according to SEQ ID NO:2.

[0059] In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition comprises from about 0.1 µg Env to about 100 µg Env. In certain such examples the Zika virus is a plaque purified clonal Zika virus isolate .In some examples, the vaccine or immunogenic composition is unadjuvanted.

[0060] In some examples that may be combined with any of the preceding examples, the Zika virus clonal isolate is a homogenous genetic population. In some examples, the Zika virus clonal isolate does not contain a mutation in Envelope protein E (Env). In some examples, the Zika virus clonal isolate contains at least one mutation. In some examples, the at least one mutation is in Zika virus Non-structural protein 1 (NS1). In some examples, the at least one mutation occurs at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1. In some examples, the at least one mutation is a Trp98Gly mutation. In some examples the at least one mutation is not in Envelope protein E (Env). In some examples, the at least one mutation enhances genetic stability as compared to a Zika virus lacking the at least one mutation. In some examples, the at least one mutation enhances viral replication as compared to a Zika virus lacking the at least one mutation.

[0061] Other examples of the present disclosure relate to a vaccine or immunogenic composition containing one or more antigens from a plaque purified clonal Zika virus isolate. In some examples, the plaque purified clonal Zika virus isolate was plaque purified from cells contacted with an inoculum comprising a population of Zika viruses. In some examples, the cells are non-human cells. In some examples, the cells are insect cells. In some examples, the insect cells are mosquito cells. In some examples, the cells are mammalian cells. In some examples, the mammalian cells are monkey cells. In some examples, the monkey cells are from a Vero cell line. In some examples, the Vero cell line is a WHO Vero 10-87 cell line.

[0062] In some examples that may be combined with any of the preceding examples, the population of Zika viruses was heterogeneous. In some examples that may be combined with any of the preceding examples, the population of Zika viruses comprised a Zika virus clinical isolate. In some examples, the Zika virus clinical isolate is from strain PRVABC59.

In some examples that may be combined with any of the preceding examples, the population of Zika viruses comprised a Zika virus that had been previously passaged one or more times in cell culture. In some examples that may be combined with any of the preceding examples, the inoculum comprised human serum. In some examples that may be combined with any of the preceding examples, the inoculum comprised one or more adventitious agents. In some examples, the plaque purified clonal Zika virus isolate is substantially free of the one or more adventitious agents.

[0063] In some examples that may be combined with any of the preceding examples, the plaque purified clonal Zika virus isolate is modified as compared to a wild-type Zika virus. In some examples that may be combined with any of the preceding examples, the plaque purified clonal Zika virus isolate is a homogenous genetic population. In some examples that may be combined with any of the preceding examples, the plaque purified clonal Zika virus isolate does not include a mutation in Envelope protein E (Env). In some examples that may be combined with any of the preceding examples, the plaque purified clonal Zika virus isolate comprises at least one mutation. In some examples, the at least one mutation is in Zika virus Non-structural protein 1 (NS1). In some examples, the at least one mutation occurs at position 98 of SEQ ID NO: 1, or at a position corresponding to position 98 of SEQ ID NO: 1. In some examples, the at least one mutation is a Trp98Gly mutation. In some examples, the at least one mutation is not in Zika virus Envelope protein E (Env). In some examples, the at least one mutation enhances genetic stability as compared to a Zika virus lacking the at least one mutation. In some examples, the at least one mutation enhances viral replication as compared to a Zika virus lacking the at least one mutation. In some examples that may be combined with any of the preceding examples, the plaque purified clonal Zika virus isolate is an African lineage virus or an Asian lineage virus. In some examples, the plaque purified clonal Zika virus isolate is an Asian lineage virus.

[0064] In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition is a purified antigen vaccine or immunogenic composition, a subunit vaccine or immunogenic composition, an inactivated whole virus vaccine or immunogenic composition, or an attenuated virus vaccine or immunogenic composition. In some examples, the vaccine or immunogenic composition is an inactivated whole virus vaccine or immunogenic composition. In some examples that may be combined with any of the preceding examples, the plaque purified clonal Zika virus isolate was

chemically inactivated. In some examples, the plaque purified Zika virus was chemically inactivated with one or more of a detergent, formalin, beta-propiolactone (BPL), binary ethylamine (BEI), acetyl ethyleneimine, methylene blue, and psoralen. In some examples, the plaque purified clonal Zika virus isolate was chemically inactivated with formalin.

[0065] In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition further comprises an adjuvant. In some examples, the adjuvant is selected from aluminum salts, toll-like receptor (TLR) agonists, monophosphoryl lipid A (MLA), synthetic lipid A, lipid A mimetics or analogs, MLA derivatives, cytokines, saponins, muramyl dipeptide (MDP) derivatives, CpG oligos, lipopolysaccharide (LPS) of gram-negative bacteria, polyphosphazenes, emulsions, virosomes, cochleates, poly(lactide-co-glycolides) (PLG) microparticles, poloxamer particles, microparticles, liposomes, Complete Freund's Adjuvant (CFA), and Incomplete Freund's Adjuvant (IFA). In some examples, the adjuvant is an aluminum salt. In some examples, the adjuvant is selected from alum, aluminum phosphate, aluminum hydroxide, potassium aluminum sulfate, and Alhydrogel 85. In some examples, at least 75%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% of the one or more antigens are adsorbed to the adjuvant. In some examples that may be combined with any of the preceding examples, the vaccine or immunogenic composition contains from 0.1 µg to about 25 µg of the purified inactivated whole virus, such as a Zika virus with a mutation which is a tryptophan to glycine substitution at position 98 of SEQ ID NO:1 as described herein, in particular 2 µg, 5 µg or 10 µg, or in particular 10 µg purified inactivated whole virus or about 0.1 µg Env to about 100 µg Env. In certain such examples the Zika virus is a plaque purified clonal Zika virus isolate. In some examples, the vaccine or immunogenic composition is unadjuvanted.

[0066] It is to be understood that one, some, or all of the properties of the various examples described above and herein may be combined to form other examples of the present disclosure. These and other aspects of the present disclosure will become apparent to one of skill in the art. These and other examples of the present disclosure are further described by the detailed description that follows.

Brief description of The drawings

[0067] FIG. 1 shows bright field microscopy images of Vero cell monolayers mock infected (top) or infected with ZIKAV strain PRVABC59 (bottom).

[0068] FIG. 2 shows growth kinetics of ZIKAV PRVABC59 P1 on Vero cell monolayers, as determined by TCID50.

[0069] FIG. 3 shows potency assay testing (TCID50) of Zika virus PRVABC59 P5 clones a-f.

[0070] FIG. 4 shows bright-field microscopy images depicting the cytopathic effect (CPE) of growth of Zika virus PRVABC59 P6 clones a-f on Vero cell monolayers.

[0071] FIG. 5 shows potency assay testing (TCID50) of Zika virus PRVABC59 P6 clones a-f

[0072] FIG. 6 shows an amino acid sequence alignment comparing the envelope glycoprotein sequence of Zika virus near residue 330 from Zika virus strains PRVABC59 P6e (SEQ ID NO: 8) and PRVABC59 (SEQ ID NO: 9) with several other flaviviruses (WNV (SEQ ID NO: 10); JEV (SEQ ID NO: 11); SLEV (SEQ ID NO: 12); YFV (SEQ ID NO: 13); DENV 1 16007 (SEQ ID NO: 14); DENV 2 16681 (SEQ ID NO: 15); DENV 3 16562 (SEQ IDNO: 16); and DENV 4 1036 (SEQ ID NO: 17)).

[0073] FIG. 7 shows an amino acid sequence alignment comparing the NS1 protein sequence of Zika virus near residue 98 from Zika virus strains PRVABC59 P6e (SEQ ID NO: 18) and PRVABC59 (SEQ ID NO: 19) with several other flaviviruses (WNV (SEQ ID

CLAIMS

What is claimed is:

1. Use of a vaccine or immunogenic composition inducing a seroconversion rate of at least 60% in a population and comprising a dose of 1 µg to 40 µg of one antigen from a Zika virus in the manufacture of a medicament for preventing Zika virus infection in a human subject in need thereof, or inducing an immune response against a Zika antigen in a human subject in need thereof,

wherein the antigen is an inactivated whole virus and the use comprises that the medicament is to be administered to the human subject.

2. The use of claim 1, wherein the subject is pregnant or intends to become pregnant or is a woman of childbearing potential.

3. Use of a vaccine or immunogenic composition inducing a seroconversion rate of at least 60% in a population and comprising a dose of 1 µg to 40 µg of one antigen from a Zika virus in the manufacture of a medicament for preventing Zika virus disease in a fetus or newborn in need thereof, wherein the antigen is an inactivated whole virus and the use comprises that the medicament is to be administered to a pregnant human subject or a human subject that intends to become pregnant or woman of childbearing potential.

4. The use of any one of claims 1 to 3, wherein the seroconversion rate is measured 14 and/or 28 days after a single dose or prime administration.

5. The use of any one of claims 1 to 4, wherein the seroconversion rate is induced in a population of at least 20 Zika virus seronegative subjects.

6. The use according to any one of claims 1 to 5, wherein the antigen is purified and optionally wherein the main peak of the purified antigen as measured in size exclusion chromatography is more than 85% of the total area under the curve in the size exclusion chromatography.

7. The use of any one of claims 1 to 6, wherein the vaccine or immunogenic composition comprises a dose of about 1 µg to about 20 µg, preferably of about 1 µg to about 15 µg, such as of about 2 µg or of about 5 µg or of about 10 µg of purified inactivated whole virus.

8. The use of any one of claims 1 to 7, wherein the vaccine or immunogenic composition further comprises an adjuvant, preferably wherein the adjuvant is an aluminum salt.

9. The use of claim 8, wherein the vaccine or immunogenic composition comprises as an adjuvant from about 100 µg to about 600 µg of an aluminum salt, or from about 100 µg to about 450 µg of an aluminum salt, or from about 100 µg to about 300 µg of an aluminum salt, or from about 150 µg to about 250 µg of an aluminum salt, or from about 100 µg to about 225 µg of an aluminum salt, or about 200 µg of an aluminum salt.

10. The use of claim 8 or 9, wherein the adjuvant is aluminum hydroxide.

11. The use of any one of claims 8 to 10, wherein at least 75%, at least 85%, at least 90%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% of the antigen are adsorbed to the adjuvant.

12. The use of any one of claims 1 to 11, wherein the Zika virus is a clonal isolate, in particular from a plaque purified clonal Zika virus isolate.

13. The use of any one of claims 1 to 12, the vaccine or immunogenic composition comprising

- a dose of about 10 µg of purified inactivated whole virus,
- about 200 µg aluminum hydroxide,
- a buffer; and optionally
- a sugar such as sucrose.

14. The use of any one of claims 1 to 13, wherein severe adverse events are avoided, preferably wherein fever of 39 °C and higher is avoided.

15. The use of any one of claims 1 to 14, wherein the use comprises that the medicament is to be administered intramuscularly or subcutaneously.

16. The use of any one of claims 1 to 15, wherein the use comprises that the medicament is to be administered

one time, or

more than one time, preferably as a first (prime) and a second (boost) administration, more preferably wherein the prime and boost administration takes place from about 1 to

about 16 weeks apart or from about 1 to about 6 weeks apart or from about 1 to about 4 weeks apart,
optionally wherein the vaccine or immunogenic composition comprises a dose of about 10µg.

17. The use of any one of claims 1 to 15, wherein the use comprises that the medicament is to be administered as first (prime) and second (boost) administration, preferably wherein the prime and boost administration takes place about 1 week, about 2 weeks, about 3 weeks, about 4 weeks, about 5 weeks, about 6 weeks, about 7 weeks, about 8 weeks, about 9 weeks, about 10 weeks, about 11 weeks, about 12 weeks, or about 16 weeks apart, more preferably wherein the prime and boost administration takes place from 25-30 days apart.

18. The use of any one of claims 1 to 17, wherein the subject is 18 to 29 years of age or 30 to 49 years of age, preferably wherein the subject is a female subject, optionally wherein the subject is from a Zika endemic region or from a Zika non-endemic region travelling to an endemic region.

19. The use of any one of claims 1 to 18, wherein the use comprises that the medicament is to be administered as a first (prime) and a second (boost) administration about 1 to about 16 weeks apart and wherein the administration of the medicament induces 14 and/or 28 days after the boost administration a seroconversion rate of 100% in a population of at least 20 Zika virus seronegative subjects.

20. The use of any one of claims 1 to 18, wherein the use comprises that the medicament is to be administered as single dose or prime administration and wherein the administration of the medicament induces 14 and/or 28 days after the single dose or prime administration a seroconversion rate of at least 70%, at least 80% or at least 90% in a population of at least 20 Zika virus seronegative subjects.

21. The use according to any one of claims 1 to 18, wherein the use comprises that the medicament is to be administered as a first (prime) and a second (boost) administration about 1 to about 16 weeks apart, and wherein the administration of the medicament induces 14 and/or 28 days after the boost administration geometric mean neutralizing antibody titers in a population of at least 20 flavivirus naïve subjects and/or at least 20 Zika virus seronegative subjects, which are at least 10 times, or at least 15 times, or at least 20 times, or at least 25 times higher than the geometric mean neutralizing antibody titers induced 28 days after the prime administration, as determined by the plaque reduction neutralization test (PRNT).