SNIF: GMOB-2023-19230

Domain:
GMO

Authorisation type:
Deliberate release of GMO or a combination of GMOs for any other purpose than for placing on the market

Application type:
Summary notification for the release of genetically modified higher organisms other than higher plants in line with Decision 2002/813/EC Annex, Part 1.

Recipient Member State:
Spain

Competent Authority:
National Commission of Biosafety-Ministry of Ecological Transition

Notification number:
B/ES/23/22

Acknowledgement date:
2023-10-10

A- General information

Details of notification

Details of notification
Notification number B/ES/23/22 dated 28 August 2023
Member State of notification
Spain
Title of the project
Phase III, randomized, observer-blind, placebo-controlled, multi-center, multinational study to evaluate the efficacy, immunogenicity, and safety of a Respiratory Syncytial Virus (RSV) vaccine in infants and toddlers.
Proposed period of release
Starting date
2024-05-01
Finishing date
2024-11-30

Notifier

Name of institute or company
Sanofi Pasteur Inc.
Email
Not provided
Phone number
Not provided
Website
Not provided
Address
Not provided
Post code
Not provided
Country
Not provided

GMO characterisation

(a) Indicate whether the GMO is a:

Viroid
No
RNA virus
Yes
DNA virus
No
Bacterium
No
Fungus
No
Animal
No

Other
No

(b) Identity of the GMO (genus and species)

Orthopneumovirus, Respiratory Syncytial Virus

(c) Genetic stability - according to Annex IIIa, II, A(10)

Genetic stability could be affected by two mechanisms: homologous recombination and accumulation of mutational changes, which are influenced by selection pressure. In many organisms, transfer of genetic material could occur by homologous recombination under natural conditions and therefore influence biological evolution at many different levels of the organisms. In most negative sense ribonucleic acid (RNA) viruses including RSV, although sporadic authentic examples indicate that homologous recombination can occur, natural recombination seems to be generally rare or even absent. To investigate the recombination events of RSV, a co-infection in vitro study by two RSV mutants was conducted. It turned out that an RSV variant was identified as a recombined RSV in only one of six coinfections. The isolation of only one single recombinant RSV under optimized experimental conditions suggests that recombination is rare indeed in RSV. As a consequence, it is clear that natural recombination is of low concern for vaccine stability and safety. The second mechanism that could affect the genetic stability is the error-prone nature of replication of RNA virus genomes. Mutation rates vary between RNA viruses, ranging between 106 and 104 per nucleotide site per cell infection, depending on the RNA virus and methods used. The point mutations are very rare in the wild type (WT) RSV A2 strain. For additional information on the evaluation of the genetic stability of RSV NS2/1313/I1314L please see section E2.

Is the same GMO release planned elsewhere in the Community (in conformity with Article 6(1)), by the same notifier?

Yes
Country
Finland
Germany

Has the same GMO been notified for release elsewhere in the Community by the same notifier?

No

Has the same GMO been notified for release or placing on the market outside the Community by the same or other notifier?

Yes
Country
BR
Insert the notification number(s) (if exist)
Not known
Country
IN
Insert the notification number(s) (if exist)
FORM2A-24862227710
Country

ZA
Insert the notification number(s) (if exist)
39.4.1/ Sanofi-Aventis SA - 23/001

Summary of the potential environmental impact of the release of the GMOs.

When considering how the genetic modification to RSV and proposed activities conducted with RSV NS2 /1313/I1314L might lead to harm to humans or the environment, all potential risks were characterized in relation to both the seriousness and likelihood of harm, taking into account the current scientific/technical knowledge and the proposed limits and controls. Both the short- and long-term impact were considered. Credible pathways of potential harm that were considered included exposure of people or animals to RSV NS2/1313/I1314L, potential for persistence of RSV NS2/1313/I1314L and the potential for recombination with other viruses. Potential harms that were considered in relation to these pathways included severe RSV disease and increased disease burden in people. The overall risk linked to the use of RSV vaccine NS2/1313 /I1314L for both humans and the environment is considered negligible. The principal reasons for the conclusion of negligible risks are: i. the attenuation phenotype of the NS2 and 1313/I1314L mutations in terms of reduced ability to replicate in vivo as demonstrated in nonhuman primates and in RSV seronegative children aged 6 to 24 months of age, ii. the absence of shedding of RSV NS2/1313/I1314L in vaccinated seropositive children, iii. there is no known animal reservoir for RSV, iv. the suitability of the proposed limits and controls.

B. Information relating to the recipient or parental organisms from which the GMO is derived

1. Recipient or parental organism characterisation

Indicate whether the recipient or parental organism is a:

Viroid
No
RNA virus
Yes
DNA virus
No
Bacterium
No
Fungus
No
Animal
No
Other
No

2. Name

(i) Order and/or higher taxon (for animals)
Mononegavirales
(ii) Genus
Orthopneumovirus
(iii) Species
Respiratory Syncytial Virus (RSV)
(iv) Subspecies
Not applicable
(v) Strain
A2
(vi) Pathovar (biotype, ecotype, race, etc.)
Not applicable

3. Geographical distribution of the organism

(a) Indigenous to, or otherwise established in, the country where the notification is made:
yes
(b) Indigenous to, or otherwise established in, other EC countries:
yes
Indicate the type of ecosystem in which it is found:
mediterranean
alpine
continental
macaronesian
pannonian
steppic

atlantic
boreal
black_sea
(c) Is it frequently used in the country where the notification is made?
No
(d) Is it frequently kept in the country where the notification is made?
No

4. Natural habitat of the organism

(a) Is the organism a microorganism ?
Yes
Water
No
Soil, free-living
No
Soil in association with plant-root systems
No
In association with plant leaf/stem systems
No
In association with animals
No
Other
Yes
Specify
Human respiratory tract
(b) Is the organism an animal?
No

5(a) Detection Techniques

Detection Techniques
1) Rapid RSV antigen test: Checks for proteins from the RSV virus, producing results in under an hour. It is from 80% to 90% sensitive in infants and young children but not sensitive in adults. 2) Real-time reverse transcriptase-polymerase chain reaction (rRT-PCR): Highly sensitive in both older children and adults. 3) Viral culture: This test is not as sensitive as the rRT-PCR and antigen tests, and is not usually used in clinical settings.

5(b) Identification Techniques

Identification Techniques
The most common identification techniques include the above rRT-PCR, RSV specific PCR, sequencing, or immunostaining of viral cultures.

6. Is the recipient organism classifies under existing Community rules relating to the protection of human health and/or the environment?

Yes
Specify
RSV is classified in risk group 2, according to the Directive 2000/54/EC of the European Parliament and of the Council (18 September 2000, Directive on the protection of workers from risks related to exposure to biological agents at work).

7. Is the recipient organism significantly pathogenic or harmful in any other way (including its extracellular products), either living or dead?

yes

To which of the following organisms

humans
Yes
animals
No
plants
No
Other
No
Give the relevant information specified under Annex III A, point II, (A)(11)(d) of Directive 2001/18/EC
a. Classification of hazard according to existing Community rules concerning the protection of human health and/or the environment; RSV is classified in risk group 2, according to the Directive 2000/54/EC of the European Parliament and of the Council (18 September 2000, Directive on the protection of workers from risks related to exposure to biological agents at work). b.Generation time in natural ecosystems, sexual and asexual reproductive cycle; RSV has an intracytoplasmic replication cycle and cannot replicate outside a host. RSV infection appears to be limited, as it infects only apical cells in the airway epithelium. People infected with RSV are usually contagious for 3 to 8 days. However, some infants, and people with weakened immune systems, can continue to shed the virus even after they stop showing symptoms, for as long as 4 weeks. c. Information on survival, including seasonability and the ability to form survival structures; RSV is an enveloped virus and is thus very fragile. RSV from infected individual can survive on fomites (including paper tissues, beds, tabletops, and toys) for up to 6h. In addition, RSV can survive on contaminated skin (e.g., hands) for up to 25min. RSV seasonality varies around the world. In Europe, RSV infections exhibit seasonality with an average season starting in the beginning of December, peaking in early February, and continuing until early April with wide variation between countries. d. Pathogenicity: infectivity, toxigenicity, virulence, allergenicity, carrier (vector) of pathogen, possible vectors, host range including non-target organism. Possible activation of latent viruses (proviruses). Ability to colonize other organisms; RSV only causes disease in humans and Chimpanzees. In healthy adults RSV infection is often asymptomatic or is limited to the upper respiratory tract (URT) with symptoms similar to the common cold. RSV is the leading viral cause of lower respiratory tract (LRT) infection in infants and young children having caused approximately 33 million cases of low respiratory illness (LRI) and approximately 118000 deaths worldwide in children <5 years of age in 2015. In Chimpanzees it is presented as a common cold. There is no carrier vector, infection can only spread by coughs or sneezes releasing contaminated droplets into the air. e. Antibiotic resistance, and potential use of these antibiotics in humans and domestic organisms for prophylaxis and therapy; Not applicable: RSV virus does not contain any antibiotic resistance genes. f. Involvement in environmental pr ocesses: primary production, nutrient turnover, decomposition of organic matter, respiration, etc. Not applicable.

8. Information concerning reproduction

(a) Generation time in natural ecosystems:
RSV has an intracytoplasmic replication cycle and cannot replicate outside a host. RSV infection appears to be limited, as it infects only apical cells in the airway epithelium. People infected with RSV are usually contagious for 3 to 8 days. However, some infants, and people with weakened immune systems, can continue to shed the virus even after they stop showing symptoms, for as long as 4 weeks.
(b) Generation time in the ecosystem where the release will take place:

The generation time will be the same as in the natural ecosystem in section 8a above.

(c) Way of reproduction

Asexual

(d) Factors affecting reproduction:

Host immune response: A robust immune response will limit viral replication. Viral load: The amount of virus present in an individual can affect RSV replication. Co-infections with other respiratory viruses can affect RSV replication.

9. Survivability

(a) Ability to form structures enhancing survival or dormancy:

(i) endospores

No

(ii) cysts

No

(iii) sclerotia

No

(iv) asexual spores (fungi)

No

(v) sexual spores (fungi)

No

(vi) eggs

No

(vii) pupae

No

(viii) larvae

No

Other

Yes

Specify

RSV does not form structures enhancing survival or dormancy.

(b) Relevant factors affecting survivability

RSV is a fragile, lipid-enveloped virus sensitive to desiccation with an intra-cytoplasmic replication cycle. It does not replicate outside the host and its infectious potential rapidly decreases in the external environment. RSV can survive on fomites (including paper tissues, beds, table tops and toys) for up to 6h. In addition, RSV can survive on contaminated skin (e.g. hands) for up to 25min.

10(a) Ways of dissemination

RSV can spread when an infected person coughs or sneezes, releasing contaminated droplets into the air. Transmission usually occurs when these droplets come into contact with (or inoculate) another person's eyes, nose, or mouth.

10(b) Factors affecting dissemination

RSV can spread when an infected person coughs or sneezes, releasing contaminated droplets into the air. Dissemination is affected by the titers of virus shed by the infected person. Dissemination is also affected by the external environmental factors such as temperature and humidity; in Europe, RSV infections exhibit seasonality with an average season starting in the beginning of December, peaking in early February and continuing until early April with wide variation between countries.

11. Previous genetic modifications of the recipient or parental organism already notified for release in the country where the notification is made (give notification numbers)

Not applicable

C. Information relating to the genetic modification

1. Type of the genetic modification

Insertion of genetic material
No
Deletion of genetic material
Yes
Base substitution
Yes
Cell fusion
No
Other
No

2. Intended outcome of the genetic modification

RSV NS2/1313/I1314L has three deletions: a) A 112 nt (nucleotide) phenotypically silent deletion in the SH noncoding sequence. Engineered to stabilize the complementary deoxyribonucleic acid (DNA) during propagation in bacteria. b) The attenuating deletion of the entire NS2 gene. The RSV nonstructural protein, NS2, suppresses the production of interferon / and also suppresses the cells ability to establish an antiviral state. Deletion of the NS2 gene attenuates RSV and also potentially provides increased immunogenicity. The NS2 protein was recently implicated in pathogenic effects resulting in distal airway obstruction and its deletion may increase vaccine tolerability. c) The attenuating 3 nt deletion of codon 1313 of the L gene. Deletion of codon 1313 (nt 12,434 to 12,436) in the L gene (1313) yielded a temperature sensitive attenuating mutation (shutoff temperature of 37C). Replication of RSV with 1313 was reduced about 50-fold in nasal turbinates, and 150-fold in lungs when compared to wild type RSV. RSV NS2/1313/I1314L has 8 base substitutions: a) 5 silent nucleotide substitutions in the last 4 codons of the SH Open Reading Frame (ORF). Engineered to stabilize the complementary DNA during propagation in bacteria. b) Two missense nucleotide substitutions in codon 1314 of the ORF encoding the L polymerase.The 1313 mutation is susceptible to a second-site compensatory mutation at codon 1314 unless stabilized by an I1314L mutation (codon ATA to CTG, nt 12,437 to 12,439).

3(a) Has a vector been used in the process of modification?

Yes
3(b) If yes, is the vector wholly or partially present in the modified organism?
No

6. Composition of the insert

(a) Composition of the insert

There is no insert in RSV NS2/1313/I1314L. The vector used to construct RSV NS2/1313/I1314L is the RSV A2 WT genome into which the above mutations were engineered by deletions or substitutions. No foreign DNA was inserted into RSV NS2/1313/I1314L genome. Therefore, all of the below is Not Applicable.

(b) Source of each constituent part of the insert

Not applicable

(c) Intended function each constituent part of the insert in the GMO

Not applicable

(d) Location of the insert in the host organism

On a free plasmid
No
Integrated in the chromosome
No
Other
Yes
Specify
Not applicable

(e) Does the insert contain parts whose product or function are not known?

No

D. Information on the organism(s) from which the insert is derived

1. Indicate whether it is a:

Viroid
No
RNA virus
No
DNA virus
No
Bacterium
No
Fungus
No
Animal
No
Other
Yes
Specify kingdom, phylum and class
There is no insert in RSV NS2/1313/I1314L. The vector used to construct RSV NS2/1313/I1314L is the RSV A2 WT genome into which the above mutations were engineered by deletions or substitutions. No foreign DNA was inserted into RSV NS2/1313/I1314L genome. Therefore, all of the below is Not Applicable.

2. Complete name

(i) Order and/or higher taxon (for animals)
Not applicable
(ii) Family name (for plants)
Not applicable
(iii) Genus
Not applicable
(iv) Species
Not applicable
(v) Subspecies
Not applicable
(vi) Strain
Not applicable
(vii) Cultivar/Breeding line
Not applicable
(viii) Pathovar
Not applicable
(ix) Common name
Not applicable

3. Is the organism significantly pathogenic or harmful in any other way (including its extracellular products), either living or dead?

no
(b) are the donated sequences involved in any way to the pathogenic or harmful properties of the organism?

no

Give the relevant information specified under Annex III A, point II, (A)(11)(d) of Directive 2001/18/EC

Not applicable

4. Is the donor organism classified under existing Community rules relating to the protection of human health and the environment, such as Directive 90/679/EEC on the protection of workers from risks related to exposure to biological agents at work?

No

5. Do the donor and recipient organism exchange genetic material naturally?

no

E. Information relating to the genetically modified organism

1. Genetic traits and phenotypic characteristics of the recipient or parental organism which have been changed as a result of the genetic modification

(a) Is the GMO different from the recipient as far as survivability is concerned?
no
Specify
RSV is a fragile, lipid-enveloped virus sensitive to desiccation with an intra-cytoplasmic replication cycle. It does not replicate outside the host and its infectious potential rapidly decreases in the external environment. RSV can survive on fomites (including paper tissues, beds, table tops and toys) for up to 6h. In addition, RSV can survive on contaminated skin (e.g. hands) for up to 25min. RSV NS2/1313/I1314L is highly attenuated, particularly in the lower respiratory tract, as compared to parental WT RSV A2, but this does not affect its survivability outside the host.
(b) Is the GMO in any way different from the recipient as far as mode and/or rate of reproduction is concerned?
yes
Specify
RSV NS2/1313/I1314L attenuation results in much lower peak titers than the parental RSV A2. RSV NS2 /1313/I1314L is capable of efficient reproduction only in the upper airway while the parental virus often spreads to the lower airway.
(c) Is the GMO in any way different from the recipient as far as dissemination is concerned?
yes
Specify
Attenuation of RSV NS2/1313/I1314L results in much lower titers of shed virus than the WT A2, reducing the likelihood of spread beyond the initial host.
(d) Is the GMO in any way different from the recipient as far as pathogenicity is concerned?
yes
Specify
RSV NS2/1313/I1314L is a live-attenuated vaccine candidate designed to be much less pathogenic than WT RSV.

2. Genetic stability of the genetically modified organism

The genetic stability of RSV NS2/1313/I1314L Pre-Master Seed (P3) was assessed by serial passage on SP serum-free Vero cells up to five times (passage 8) under serum-free conditions. The pre-Master Seed Lot (pre-MSL) as well as virus at passage 8 (5 passages beyond Pre-Master Seed) was then sequenced by high throughput sequencing (HTS) to assess the genetic stability of RSV NS2/1313/I1314L and to examine if subpopulations of viral mutants with compensatory mutations emerged during serial passage at the cut-off of 10%. 99.7% full length coverage was obtained and the sequence of both the pre-MSL and P8 virus conformed to the expected sequence. The only mutation was a single nucleotide substitution found at position 14456 of the viral genome that represents a change from a Thymine (T) to an Adenine (A). This mutation is located in a low complexity region close to a homopolymer of Thymine and Adenine and further results in a homopolymer of 6 consecutive Adenine. The mutation in a non-coding region. No other sequence changes were observed in the Pre-MSL at passage 3 and at passage 8. This mutation T14456A may have existed in the original virus clone picked. The genetic stability of RSV NS2/1313/I1314L also was assessed in children by the National Institutes of Health (NIH). RT-PCR and partial sequence analysis of nasal wash (NW) isolates obtained at the peak of vaccine shedding from 18 RSV-seronegative vaccinees confirmed the presence of the NS2 deletion and the 1313 and I1314L mutations. Briefly, to verify the presence and genetic stability of the attenuating elements, viral RNA was obtained from a single passage of NW fluid on Vero cells. The presence of the NS2 gene deletion was verified by agarose gel electrophoresis, confirming an 855

base pair RT-PCR amplicon spanning the deletion. The presence of the 1313 deletion and the I1314L mutation was confirmed by sequence analysis of a 758 base pair PCR fragment of the L gene.

3. Is the GMO significantly pathogenic or harmfull in any way (including its extracellular products), either living or dead?

no

(b) Give the relevant information under Annex III A, point II (A)(11)(d) and II(C)(2)(i):

No adverse systemic or local effects were observed following repeated nasal administrations of Live-attenuated Respiratory Syncytial Virus vaccine candidate in non-human primates.

4. Description of identification and detection methods

(a) Techniques used to detect the GMO in the environment

RSV can be detected either by the presence of viral nucleic acids, for example genomic detection using quantitative or standard RT-PCR, or by presence of replication competent virus by viral culture, for example by plaque assay.

(b) Techniques used to identify the GMO

A quantitative Reverse Transcriptase Polymerase Chain Reaction (qRT-PCR) assay was developed to specifically detect and quantify RSV NS2/1313/I1314L in nasal swab samples. The qRT-PCR assay was designed using the Light Cycler Probe system from Sigma. This system consists of two hybridization probes that are designed so that they bind the target 1-5 nucleotides apart. Probe 1 (donor) is labeled on the 3 end with a donor reporter. Probe 2 (acceptor) is labeled at the 5 end with an acceptor reporter. During the annealing step, the PCR primers and the LightCycler Probes hybridize to their specific target regions, bringing the probes in close proximity. When this happens, the donor dye is excited by the LightCycler and energy is transferred from the donor to the acceptor dye. The acceptor reporters emission is detected by the Light Cycler at 640 nm. If the probes bind, but are not in close proximity, no signal is produced. The Light Cycler probes for the RSV NS2/1313/I1314L qRT-PCR assay target the deletion site of the NS2 gene in RSV NS2. Probe 1 binds before the deletion and Probe 2 binds across the deletion site. While the primers and probes may bind WT RSV A2 due to high sequence similarity, the two probes would not bind in close enough proximity to create signal as the NS2 gene is over 500 nucleotides long, making this method highly specific. If the probes are not in close proximity, as is the case with wild type RSV, no signal is produced. Additionally, RSV NS2/1313/I1314L can also be identified by viral culture at different temperatures as it exhibits moderate temperature sensitivity at elevated temperatures. This test assesses viral replication at different temperatures (i.e. 34C, 36C and 38C) by plaque assay to characterize the temperature sensitive phenotype of the virus.

F. Information relating to the release

1. Purpose of the release (including any significant potential environmental benefits that may be expected)

Sanofi is developing a prophylactic RSV NS2/1313/I1314L live attenuated vaccine candidate to prevent RSV. RSV NS2/1313/I1314L will be administered by intranasal route to subjects participating in the Phase III, randomized, observer-blind, placebo-controlled, multi-center, multinational study to evaluate the efficacy, immunogenicity, and safety of a Respiratory Syncytial Virus vaccine in infants and toddlers. Each participant in the clinical trial will receive 2 doses of Live-attenuated Respiratory Syncytial Virus vaccine candidate at a concentration of 6.4 log10 Plaque Forming Units (PFU) per dose at a 23-month interval or a placebo. Study duration will be between 20 to 21 months for each participant. It is anticipated that a maximum of 5334 subjects will be enrolled during this international multi-center study. While the candidate vaccine addresses an urgent need for protecting human health, there is no expected benefit for the environment.

2. Is the site of the release different from the natural habitat or from the ecosystem in which the recipient or parental organism is regularly used, kept or found?

No

3. Information concerning the release and the surrounding area

(a) Geographical location (administrative region and where appropriate grid reference):

The clinical trial will be conducted at the following clinical sites: CHUS H. Clnica U. de Santiago, Avenida Choupana, s/n,Santiago de Compostela, 15706; Hospital HM Puerta del Sur, Avenida Carlos V 70, Mstoles, Madrid; Hospital Universitario de Navarra, Irunlarrea, 3, Servicio de Farmacia, Ensayos Clnicos, Pamplona, Navarra; Hospital Quirnsalud Barcelona, Calle Marquesa Vilallonga 22, Servicio de Farmacia, Barcelona; Instituto Hispalense de Pediatria, C/ Jardn de la isla, n 6 acceso I.Edif. Expolocal, Sevilla.

(b) Size of the site (m2)

(i) actual release site (m2)
No specific size for the release, immunizations are going to be performed in separate examination rooms.
(ii) wider release area (m2)
No specific size for the release, immunizations are going to be performed in separate examination rooms.

(c) Proximity to internationally recognised biotopes or protected areas (including drinking water reservoirs), which could be affected:

The primary release is the moment where the vaccine is administered to the participant. No release is anticipated outside the examination rooms. Containment measures during the administration of RSV NS2 /1313/I1314L to subjects will exclude the release of RSV NS2/1313/I1314L into the environment. Personal protective equipment will be used to avoid exposure to RSV NS2/1313/I1314L to the medical personnel involved in the administration of the product. Therefore, the likelihood that RSV NS2/1313/I1314L will be released to the proximity of significant biotopes, protected areas or drinking water supplies as possible potential sites, which could be affected, is negligible.

(d) Flora and fauna including crops, livestock and migratory species which may potentially interact with the GMO

Not relevant

4. Method and amount of release

(a) Quantities of GMOs to be released:

In the notified country RSV NS2/1313/I1314L will be administered to maximally 2700 subjects who will receive maximum 2 doses of 6.4 log10 PFU per dose.

(b) Duration of the operation:

Intranasal immunization of the subjects will take a few minutes.

(c) Methods and procedures to avoid and/or minimise the spread of the GMOs beyond the site of the release

At the clinical sites, considerable care will be taken to ensure that RSV NS2/1313/I1314L is contained and that personnel and the areas are not exposed. After administration of the vaccine the instructions for the disposal of the intranasal device used for administration are as follows: Discard the intranasal device used for administration after usage as per protocol. The intranasal device used for administration (medical device) is designed for single-use. Do not re-use the device system. Discard the prepared device if it would not be used within 6 hours after the preparation. Do not freeze and re-use the prepared device. All used materials will be destroyed on site, in specific containers, at the end of each vaccination.

5. Short description of average environmental conditions (weather, temperature etc.)

Not applicable: given that RSV NS2/1313/I1314L is prepared for administration and given to subjects in a clinical environment, it is not anticipated that RSV NS2/1313/I1314L will be released into the environment.

6. Relevant data regarding previous releases carried out with the same GMO, if any, specially related to the potential environmental and human health impacts from the release

The safety and immunogenicity of the investigational RSV vaccine has been evaluated in 7 clinical trials so far. Completed or Ongoing Phase I and II Studies: - Sponsor NIAID (Study CIR 288): Phase I (USA) age group: 4-59 months, objective (population): Safety and Immunogenicity (S+ or S-), RSV vaccine Recipients (Total in study): 15 (22), 30 (45), RSV vaccine Dose: 1 admin (5.0 log10 PFU/dose), 1 dose (6.0 log10 PFU /dose), Timeframe: Jun 2013 Apr 2023 (est). - Sponsor NIAID (Study IMPAACT 2018 / 38405): Phase I (USA), age group: 6-24 months, objective (population): Safety, Immunogenicity and Infectivity (S-), RSV vaccine Recipients (Total in study): 25 (62), RSV vaccine Dose: 2 doses (6.0 log10 PFU/dose), Timeframe: Sept 2017 Oct 2020. - Sponsor NIAID (Study CIR 321*): See IMPAACT 2018 / 38405. - Sponsor NIAID (Study IMPAACT 2021 / 38530): Phase I/II (USA), age group: 6-25 months, objective (population): Safety and Immunogenicity (S-), RSV vaccine Recipients (Total in study): 40 (160), RSV vaccine Dose: 2 doses (6.0 log10 PFU/dose), Timeframe: May 2019 Apr 2023 (est). - Sponsor Sanofi (Study VAD00001): Phase I/II (USA / Chile / Honduras), age group:6-18 months, objective (population): Safety, Immunogenicity, Infectivity, Dose-Finding (R+ or R-), RSV vaccine Recipients (Total in study): 155 (259), RSV vaccine Dose: 1 dose or 2 doses (5.6 log10 PFU/dose), 1 dose or 2 doses (6.2 log10 PFU/dose), Timeframe: Sept 2020 -*

April 2023. - Sponsor Sanofi (Study VAD00014): Puerto Rico, age group:6-24 months, objective (population): Safety, immunogenicity, transmissibility (R+ or R-), RSV vaccine Recipients (Total in study): 50 (100), RSV vaccine Dose: 2 doses of 6.2 log10 PFU /dose, Timeframe: Feb 2023 Dec 2023 (est). - Sponsor Sanofi (Study VAD00012): Japan, age group:6-24 months, objective (population): Safety and immunogenicity, RSV vaccine Recipients (Total in study): 12 (18), RSV vaccine Dose: 2 doses of 6.2 log10 PFU/dose, Timeframe: May 2023 Dec 2023. Legend: S+ (RSV seropositive); S- (RSV seronegative); R+ (RSV experienced) R- (RSV nave) the portion of IMPAACT 2018, executed at Johns Hopkins University Center for Immunization Research (CIR); **This is the anticipated number, based on the VAD00001 study design; as full unblinding has not taken place, the final number of IP recipients is unknown. Because of the restricted shedding of the attenuated virus, the quantities that will be released in the environment by vaccinated individuals will be negligible. Due to the attenuation, and because most humans have pre-existing immunity against wt RSV, it is unlikely that the RSV NS2/1313/I1314L vaccine will cause a measurable infection in adults and older children and if it happened, it would most likely be asymptomatic. Recombination of the RSV NS2/1313/I1314L virus with wt RSV is highly unlikely. RSV is a negative stranded non-segmented RNA virus for which recombination is generally rare or even absent. Being a non-segmented virus, RSV cannot recombine through re-assortment like influenza viruses. In this light, it is highly unlikely that RSV NS2/1313/I1314L will regain the complete genetic material for the NS2 gene deletion through recombination. The temperature sensitive attenuating deletion of codon 1313 in the polymerase (L) is stabilized by substitution of leucine (L) for isoleucine (I) at codon 1314.

G. Interactions of the GMO with the environment and potential impact on the environment, if significantly different from the recipient or parent organism

1. Name of target organisms (if applicable)

Applicable?
Yes
(i) Order and/or higher taxon (for animals)
Primates
(ii) Family name (for plants)
Not applicable
(iii) Genus
Homo
(iv) Species
Homo sapiens
(v) Subspecies
Not applicable
(vi) Strain
Not applicable
(vii) Cultivar/Breeding line
Not applicable
(viii) Pathovar
Not applicable
(ix) Common name
Not applicable

2. Anticipated mechanism and result of interaction between the released GMOs and the target organism (if applicable)

Applicable?
Yes
Specify
The RSV NS2/1313/I1314L is a live, attenuated, temperature sensitive vaccine. The expressed viral proteins, principally those encoded by the F and G genes, contain antigenic determinants for neutralization. Therefore, these proteins act as immunogens and their expression via the RSV NS2/1313/I1314L virus induces a protective immune response against wt RSV. For information on previous releases of this GMO please section A6.

3. Any other potentially significant interactions with other organisms in the environment

Even under laboratory conditions, attenuated RSV strains do not cause disease and replicate only at low levels in animal models such as mice and non-human primates. Thus, it is unlikely that RSV NS2/1313 /I1314L could infect and cause disease in wild mammals. The only significant impact could be on chimpanzees as this is only species besides humans known to experience RSV disease. The attenuation of RSV NS2/1313/I1314L was initially established in chimpanzes where innoculation resulted in a self-limiting infection with low levels of viral shedding which makes further transmission cycles unlikely. The consequence of potential survival and dissemination of RSV NS2/1313/I1314L outside the host is negligible.

4. Is post-release selection such as increased competitiveness, increased invasiveness for the GMO likely to occur?

no

5. Types of ecosystems to which the GMO could be disseminated from the site of release and in which it could become established

There is no ecosystem in which the vaccine strain could become established.

6. Complete name of non-target organisms which (taking into account the nature of the receiving environment) may be unintentionally significantly harmed by the release of the GMO

(i) Order and/or higher taxon (for animals)
Not applicable
(ii) Family name (for plants)
Not applicable
(iii) Genus
Not applicable
(iv) Species
Not applicable
(v) Subspecies
Not applicable
(vi) Strain
Not applicable
(vii) Cultivar/Breeding line
Not applicable
(viii) Pathovar
Not applicable
(ix) Common name
Not applicable

7. Likelihood of genetic exchange in vivo

(a) from the GO to other organisms in the release ecosystem:
Natural recombination of negative-sense RNA viruses including RSV in the environment appears to be vanishingly rare or altogether absent. Should genetic exchange from the GMO to wt RSV occur, it would only serve to attenuate the wt virus.
(b) from other organisms to the GMO:
In the event of genetic exchange from wt RSV to the GMO, recombination could in the worst case scenario, only result in the full reversion of the GMO to wt RSV which is naturally and constantly circulating in the environment.
(c) likely consequences of gene transfer**
The consequences of the potential gene transfer to or from the GMO are negligible, as such transfer could only occur with wt RSV, at worst reverting the GMO to wt (please see 7a and 7b above).

8. Give references to relevant results (if available) from studies of the behaviour and characteristics of the GMO and its ecological impact carried out in simulated natural environments (e.g. microcosms etc.):

9. Possible environmentally significant interactions with biogeochemical processes (if different from the recipient or parental organism)

H. Information relating to monitoring

H. Information relating to monitoring

1. Methods for monitoring the GMOs
The intended function of RSV NS2/1313/I1314L is to induce an RSV specific immune responses, which will be measured by assessment of immune responses against RSV. In addition, subjects participating in the clinical trial using RSV NS2/1313/I1314L will be monitored for clinical assessment (e.g. physical examinations), and adverse event monitoring. The presence of virus in nasal secretions will be monitored during the phase I/II studies.
2. Methods for monitoring ecosystem effects
Ecosystems effects will not be monitored as RSV NS2/1313/I1314L is not naturally present in any ecosystem.
3. Methods for detecting transfer of the donated genetic material from the GMO to other organisms
It is highly unlikely that transfer of donor genetic material from RSV NS2/1313/I1314L will be transferred to other organisms.
4. Size of the monitoring area (m2)
Not applicable
5. Duration of the monitoring
Not applicable
6. Frequency of the monitoring
Not applicable

I. Information on post release and waste treatment

I. Information on post release and waste treatment

1. Post-release treatment of the site
The rooms in the medical facility used to prepare and administer the vaccine will be cleaned before and after manipulation with a standard desinfectant active against RSV. Surfaces will be decontaminated and cleaned after use with standard desinfectant active against RSV. After administration of the vaccine the intranasal device will be disposed and all used materials will be destroyed on site, in specific containers, at the end of each vaccination.
2. Post-release treatment of the GMOs
Materials in contact with the RSV NS2/1313/I1314L will be considered as contaminated and all wastes (including vaccine vials, syringes, vial access cannulas, and nasal devices) will be placed in bins suitable for biohazardous waste directly following administration of the vaccine. Decontamination of RSV NS2/1313 /I1314L will be managed like any other enveloped virus. Decontamination by heat or chemicals (bleach, ethanol/isopropyl alcohol, detergents) can be envisaged. A few minutes at 100C or with contact chemicals have been shown to achieve full decontamination. Therefore, autoclaving or incinerating, which are common processes of decontamination, are fully applicable to the RSV NS2/1313/I1314L vaccine.
3(a) Type and amount of waste generated
The generated waste consists of: Vial containing remaining vaccine, Syringes, Vial access cannulas, Device, Any other consumable material directly in contact with the vaccine.
3(b) Treatment of waste
All equipment, supplies including gloves, and receptacles in contact with RSV NS2/1313/I1314L will be directly handled and disposed of in bins suitable for biohazardous waste. Partially used empty boxes will be monitored by the Clinical Research Associate (CRA). In that case, they will be kept on site in a secure place and well identified and will be destroyed on site after monitoring by the CRA at the end of each cohort as biohazardous waste . The destruction will be documented on site. If destruction on site is not possible, the vials will be returned for destruction to the Sponsor, or to a third-party vendor where applicable at room temperature along with the applicable form provided by CRA. Prior to the return of unused and unusable products (expired, break in the cold chain) the responsible site personnel will account for all study vaccine and the CRA will monitor product accountability on the Investigational Product Dispensing and Reconciliation Form.

J. Information on emergency response plans

J. Information on emergency response plans

1. Methods and procedures for controlling the dissemination of the GMO(s) in case of unexpected spread

RSV NS2/1313/I1314L has the same structure and physical properties as parental RSV, which is a fragile, lipid-enveloped virus sensitive to desiccation with an intracytoplasmic replication cycle. As all enveloped viruses, RSV is sensitive to detergents and solvents. Like all viruses, RSV does not replicate or survive outside the host cell and is sensitive to heat and ultraviolet radiation. RSV NS2/1313/I1314L, like parental RSV, is susceptible to common disinfectants such as 70% ethanol, various detergents including 0.1% sodium deoxycholate, sodium dodecyl sulphate, and Triton X-100, and to 1% sodium hypochlorite, formaldehyde (5% formalin), 2% glutaraldehyde, 1% iodine and is inactivated by heat.

2. Methods for removal of the GMOs of the areas potentially affected

See section 1

3. Methods for disposal or sanitation of plants, animals, soils etc. that could be exposed during or after the spread

See section 1

4. Plans for protecting human health and the enironment in the event of an undesirable effect

RSV NS2/1313/I1314L is a vaccine developed to protect humans from RSV infection. Data from Phase I clinical trials indicate that RSV NS2/1313/I1314L was well tolerated with rates of upper respiratory symptoms comparable to placebo and no incidence of lower respiratory tract infection in vaccinees. Sanofi Pasteur Inc. will continue to monitor the safety profile of RSV NS2/1313/I1314L vaccine during future clinical trials and after product launch by conducting: Intensive monitoring of the safety profile in vaccinees during clinical trials. Routine pharmacovigilance practices allowing a comprehensive, continuous and global overview of post-licensure safety profile.