SUMMARY NOTIFICATION INFORMATION FORMAT FOR THE RELEASE OF GENETICALLY MODIFIED ORGANISMS OTHER THAN HIGHER PLANTS IN ACCORDANCE WITH ARTICLE 11 OF DIRECTIVE 2001/18/EC

MVATG17401
(MVA HIV-B)

EHVA T01/ANRS VRI05 Clinical Trial
(EudraCT No: 2017-003081-27)

Version 1.0
(August 9th 2017)

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A. GENERAL INFORMATION

1. Details of notification

a) Member State of notification **France**
b) Notification number: **B/DE/19/PEI3493**
c) Date of acknowledgement of notification: **13/07/2018**
d) Title of the project **A Phase I/II randomised therapeutic HIV vaccine trial in individuals who started antiretrovirals during primary or chronic infection (EHVA T01/ANRS VRI05)**
e) Proposed period of release **From the start of the clinical trial to the end of release: 24 months (expected Q4 2017 – Q4 2019).**

2. Notifier

Name of institution or company **Inserm-ANRS (French National Institute for Health and Medical Research-ANRS (France REcherche Nord&Sud Sida-hiv Hépatites), Paris, France**

3. GMO characterization

a) Indicate whether the GMO is a:

Viroid	☐
RNA virus	☐
DNA virus	☒
bacterium	☐
fungus	☐
animal	☐

- mammals ☐
- insect ☐
- fish ☐
- other animal ☐ please specify phylum, class

other, please specify (kingdom, phylum and class): **POXVIRIDAE**

b) Identity of the GMO (genus and species) **Modified vaccinia Ankara virus -based anti-HIV vaccine (named MVATG17401 or MVA HIV-B in the clinical trial)**
c) Genetic stability – according to Annex IIIa, II, A(10) **Stable**

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

Yes ☒	No ☐
If yes, insert the country code(s): DE, ES, IT, SW, UK	

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

Yes ☒	No ☐
If yes: – Member State of notification: FR - Notification number: B/FR/13/GT03	

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

Yes ☐	No ☒
If yes: – Member State of notification - Notification number	

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

None

B. INFORMATION RELATING TO THE RECIPIENT OR PARENTAL ORGANISMS FROM WHICH THE GMO IS DERIVED

1. Recipient or parental organism characterization:

a) Indicate whether the recipient or parental organism is a:

Viroid	☐	
RNA virus	☐	
DNA virus	☒	
bacterium	☐	(Production organism)
fungus	☐	
animal	☐	
- mammals	☐	
- insect	☐	
- fish	☐	
-other animal	☐	

(please specify phylum, class) **POXVIRUS**

other, please specify

2. Name (applicable for the Production Organism)

(i) Order and/or higher taxon (for animals)

Poxviridae

(ii) Genus

Orthopoxvirus

(iii) Species **Vaccinia virus (Cowpox)**
(iv) Subspecies
(v) Strain
(vi) pathovar (biotype, ecotype, race, etc.)
(vii) common name **MVA, Modified vaccinia Ankara virus**

3. Geographical distribution of the organism

a) Indigenous to, or otherwise established in, the country where the notification is made:

Yes ☐ No ☐ Not known ☒

b) Indigenous to, or otherwise established in, other EC countries:

(i) Yes ☐

If *yes*, indicate the type of ecosystem in which it is found:

Atlantic ☐
Mediterranean ☐
Arctic ☐
Alpine ☐
Continental ☐

(ii) No ☐

(iii) Not known ☒

c) Is it frequently used in the country where the notification is made?

Yes ☐ No ☐ **NA**

d) Is it frequently kept in the country where the notification is made?

Yes ☐ No ☐ **NA**

4. Natural habitat of the organism

(a) If the organism is a microorganism

Water ☐
soil, free-living ☐
soil in association with plant-root systems ☐
in association with plant leaf/stem systems ☐
in association with animals ☐

other (specify) **No natural host known (Mayr A et al. Infection 1975, 3: 6-14)**

(b) If the organism is an animal: natural habitat or usual agroecosystem:

NA

5. a) Detection techniques

Culture on chicken embryo fibroblasts

5. b) Identification techniques

PCR

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

Yes ☒	No ☐

If yes, specify

Related to Vaccine against smallpox (BSL 1)

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

Yes ☐	No ☒	Not known ☐
If yes: a) to which of the following organisms:	humans ☐ animals ☐ plants ☐ other ☐	
b) give the relevant information specified under Annex III A, point II. (A)(11)(d) of Directive 2001/18/EC		

8. Information concerning reproduction

a) Generation time in natural ecosystems: **NA**
b) Generation time in the ecosystem where the release will take place: **NA**
c) Way of reproduction: Sexual ☐ Asexual ☐ **NA**
d) Factors affecting reproduction: **MVA was derived from the vaccinia strain Ankara by more than 570 serial passages in primary chick embryo fibroblasts, which severely compromised its capacity to replicate in mammalian cells (Mayr et al, 1975; 1998).**

9. Survivability

a) Ability to form structures enhancing survival or dormancy: **NA**

(i) endospores	☐
(ii) cysts	☐
(iii) sclerotia	☐
(iv) asexual spores (fungi)	☐
(v) sexual spores (fungi)	☐
(vi) eggs	☐

(vii) pupae	☐
(viii) larvae	☐
(ix) other, please specify	

b) Relevant factors affecting survivability:

NA

10. a) Ways of dissemination

NA

10. b) Factors affecting dissemination

MVA is incapable of replication in humans and thus there should be no virus shedding from the vaccinated individuals. The genetically modified viral vaccine is not able to survive, disseminate in and/or displace other organisms and is not pathogenic to animals or plants

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)

NA

C. INFORMATION RELATING TO THE GENETIC MODIFICATION

1. Type of the genetic modification

(i) Insertion of genetic material	☒
(ii) Deletion of genetic material	☐
(iii) Base substitution	☐
(iv) Cell fusion	☐
(v) Other, please specify	

2. Intended outcome of the genetic modification

Expression of HIV proteins as antigens for immunization

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

Yes ☒	No ☐
If no, go straight to question 5.	

3. b) If yes, is the vector wholly or partially present in the modified organism?

Yes ☒	No ☐
If no, go straight to question 5.	

4. If the answer to 3 b) is yes, supply the following information

a) Type of vector

plasmid	☒
bacteriophage	☐
virus	☐
cosmid	☐
transposable element	☐

other, please specify

b) Identity of the vector

pTG17401

c) Host range of the vector

E. coli

d) Presence in the vector of sequences giving a selectable or identifiable phenotype

Yes ☒ No ☐

Antibiotic resistance ☐

Other, specify

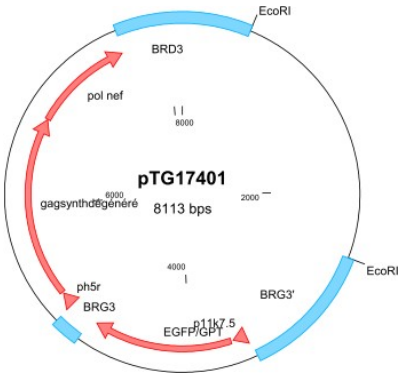
EGFP/GTP: selection marker (Enhanced Green Fluorescent Protein / Xanthine-Guanine Phosphoribosyl Transferase of *E. coli)*

e) Constituent fragments of the vector

The transfer vector (pTG17401) is composed of:

- **The *gag-pol-nef* transcript encoding for the polypeptide GAG-POL-NEF of HIV-1**
- **BRD3 and BRG3 : left and right recombination arms**
- **p11K7.5 and p5HR: early-late vaccinia promoters**
- **EGFP/GTP: selection marker (Enhanced Green Fluorescent Protein / Xanthine-Guanine Phosphoribosyl Transferase of *E. coli)***
- **Repetitive sequence of BRG3 allowing the elimination of the cassette (selection marker) by intragenic recombination**

Map of the transfer vector (pTG17401)



f) Method for introducing the vector into the recipient organism

(i) transformation ☐
(ii) electroporation ☐
(iii) macroinjection ☐
(iv) microinjection ☐

(v) infection ☐

(vi) other, please specify: **Homologous recombination**

5. If the answer to question B.3 (a) and (b) is no, what was the method used in the process of modification?

(i) transformation ☐
(ii) microinjection ☐
(iii) microencapsulation ☐
(iv) macroinjection ☐

(v) other, please specify

6. Information on the insert

a) Composition of the insert

The insert is composed of the synthetic gene: *gag-pol-nef* of HIV-1

b) Source of each constituent part of the insert

Synthetic plasmid

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

Immunization against HIV-1 proteins

d) Location of the insert in the host organism

- on a free plasmid ☐
- integrated in the chromosome ☐

- other, please specify

Inserted in the MVA genome, within the deletion III site of the MVA

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

Yes ☐ No ☒

If yes, please specify

D. INFORMATION ON THE ORGANISM(S) FROM WHICH THE INSERT IS DERIVED

1. Indicate whether it is a:

Viroid	☐
RNA virus	☒
DNA virus	☐
bacterium	☐
fungus	☐
animal	☐
- mammals	☐
- insect	☐
- fish	☐
- other animal	☐ (please specify phylum, class) ***RETROVIRIDAE***
	other, please specify:

2. Complete name

(i) order and/or higher taxon (for animals)
(ii) family name (for plants)
(iii) genus
(iv) species
(v) subspecies

(vi) strain
(vii) cultivar/breeding line
(viii) pathovar Not applicable
(ix) common name

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

Yes ☒	No ☐	Not known ☐
If *yes*, please specify the following		
a) to which of the following organisms?	Humans ☒ animals ☐ plants ☐ other ☐	
b) are the donated sequences involved in any way to the pathogenic or harmful properties of the organism?		
Yes ☐	No ☒	Not known ☐
If yes, give the relevant information under Annex III A, point II (A), 11(d):		

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?

Yes ☒	No ☐

If yes, please specify: **Human immunodeficiency virus: Risk class BSL 3**

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

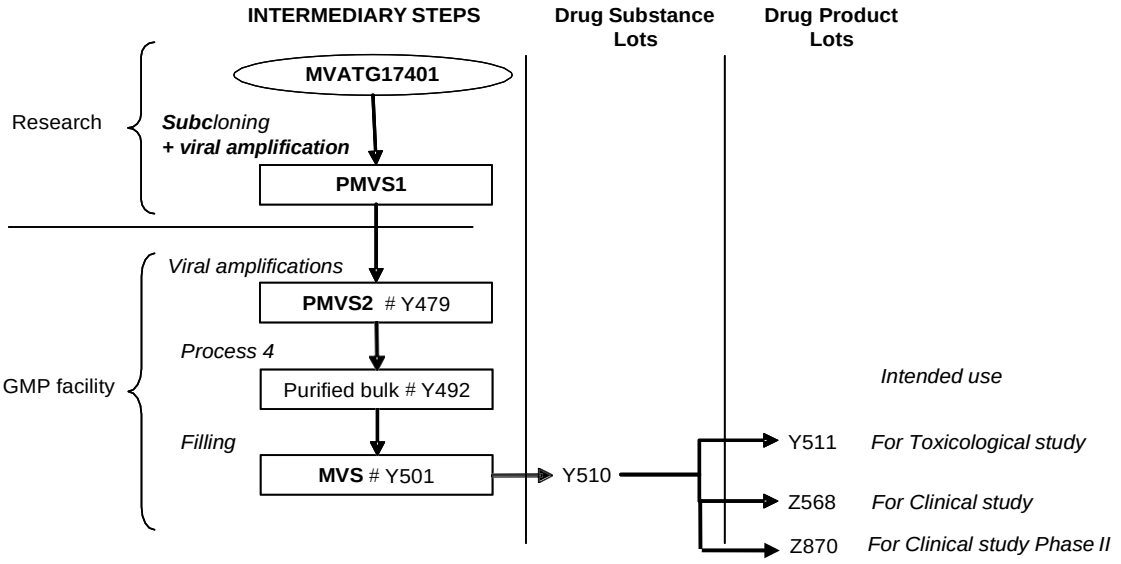
Yes ☐ (virus to human)	No ☒	Not known ☐

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? Yes ☐ No ☒ Not known ☐ Please specify:
b) Is the GMO in any way different from the recipient as far as mode and/or rate of *reproduction* is concerned? Yes ☐ No ☒ Not known ☐ Please specify:
c) Is the GMO in any way different from the recipient as far as *dissemination* is concerned? Yes ☐ No ☒ Not known ☐ Please specify:
d) Is the GMO in any way different from the recipient as far as *pathogenicity* is concerned? Yes ☐ No ☒ Not known ☐ Please specify:

2. Genetic stability of the genetically modified organism



Filliation of the different lots and their use

- **The MVATG17401 was amplified on CEF to produce the Pre-Master Virus Seed 1 (PMVS1). Controls have been performed on the PMVS1 and on isolated cloned obtained after 6 passages of the PMVS1 on CEF. The genetic stability has been performed by PCR and sequencing on PMVS1 and confirmed 100% genetic integrity on 100 tested viral clones. Stability on the protein was tested by Western Blot and showed 98% stability.**

- **Analysis of the genetic stability of recombinant vector MVATG17401 at a passage equivalent to the clinical batch (= lot Y511) has been performed. These results were used to validate the Master Virus Seed (MVS) stock. The proportion of viruses expressing the HIV gag-pol-nef fusion protein of recombinant MVATG17401 was determined by Western Blot. The fusion protein, consisting of 795 amino acids, was detected at the expected molecular weight of 90 KDa. Genetic integrity was confirmed by characterization of the genetic insert by PCR and sequencing of the coding and non-coding regions of the expression cassette and flanking viral sequences.**

Characterization of the insert by PCR and sequencing of the genetic insert and deletion II and III regions has been performed directly on viral DNA isolated from the toxicological lot Y511. All results are in compliance with specifications. The proportion of viruses expressing the fusion protein has been determined in a sample of the toxicological batch, collected before filling: 100% of the clones express the fusion protein. Therefore, the results are in full compliance with specifications.

In addition to these tests, expression of the HIV fusion protein has been assessed in samples from MVS (Y501), Tox lot (Y511) and clinical lots (Z568 and Z870). The fusion protein was detected in all samples.

These results demonstrate that vector MVATG17401 is genetically stable.

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

Yes ☐	No ☒	Not known ☐

If *yes*,

a) to which of the following organisms?:

humans	☐
animals	☐
plants	☐
other	☐

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

All data collected so far concur to demonstrate that MVA-based vaccines are unable to survive, disseminate in and/or displace other organisms and is not pathogenic to animals or plants.

4. Description of identification and detection methods

a) Techniques used to detect the GMO in the environment

The recombinant virus may be detected using a PCR.

b) Techniques used to identify the GMO

The identity of the MVA virus may be checked using PCR probes specific for both MVA and the *gag*, *pol* and *nef* inserts.

F. INFORMATION RELATING TO THE RELEASE

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

MVAT17401 (MVA HIV-B) will be used in the phase I/II EHVA T01/ANRS VRI05 clinical trial in combination with a non-GMO vaccine to assess the impact of this therapeutic vaccination upon viral control following analytic treatment interruption, in presence or not of a monoclonal antibody, versus placebo. It will be administered intramuscularly at the following dosage level 1 x 10^8 pfu/mL in expected 96 HIV-1 infected participants (44 minimum – 96 maximum).

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?

Yes ☒	No ☐

If yes, please specify:

MVA has no natural host

3. Information concerning the release and the surrounding area

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

The clinical study using MVA HIV-B is conducted in Germany, Italy, Spain, Switzerland, UK and France
Up to 13 clinical sites could be implicated:
Confirmed sites:

- **ZIM, 1. Med. Klinik (O28), Universitätsklinikum Hamburg-Eppendorf (Hamburg, Germany),**
- **UOC Immunodeficienze Virali, Istituto Nazionale Malattie Infettive Lazaro Spallanzani (Roma, Italy)**
- **Servicio de Infecciones, Hospital Clinic de Barcelona (Barcelona, Spain),**
- **Infectious Diseases Division – BH 07, Centre Hospitalier Universitaire Vaudois (Lausanne, Switzerland),**
- **St Mary's Campus, Winston Churchill Wing, Imperial College (London, UK),**
- **St Stephen's Clinical Research, Chelsea and Westminster Hospital (London, UK),**
- **Unité Immunologie Clinique, Hôpital Henri Mondor (Créteil, France).**

Expected sites (to be confirmed):

- **Service Maladies Infectieuses et Tropicales, Hôpital Avicenne (Bobigny, France)**
- **Service des Maladies Infectieuses et Tropicales, Hôpital Saint-Antoine (Paris, France)**
- **Service des Maladies Infectieuses et Tropicales, Hôpital Bichat - Claude Bernard (Paris, France)**
- **Service des Maladies Infectieuses et Tropicales, Hôpital Saint-Louis (Paris, France)**
- **Service de Médecine Interne-Maladies Infectieuses, Hopital Bicêtre (Le Kremlin-Bicêtre, France)**
- **Service Immunologie Clinique, Hôpital HEGP (Paris, France).**

b) Size of the site (m^2): **NA**

(i) actual release site (m^2):

(ii) wider release area (m^2):

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

None

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

None

4. Method and amount of release

a) Quantities of GMOs to be released:

A maximum of 96 participants will receive 1 shot of 0,5 ml of MVA HIV-B vaccine (1 x 10^8 pfu/mL) intramuscularly at week 12 of the clinical trial.

b) Duration of the operation:

The EHVA T01/ANRS VRI05 clinical trial duration is 114 weeks and the duration of GMO release is 66 weeks.

The immunizations will occur in an outpatient setting and participants will be closely observed for 60 minutes after the administration.

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

Procedures are available to administer the vaccine.
The sponsor of the clinical trial will provide a procedure detailing the precautions for confinement and inactivation of waste in accordance with the GMO guidelines. All concerned hospital staff will receive appropriate information from the sponsor before the start of the trial. Gloves will be worn for GMO handling.
The vaccination will take place in a containment zone of Class 1 (the lowest level of risk). Dressing will be placed over the site of injection for 60 minutes. All waste potentially in contact with the GMO will be inactivated according to the rules of a containment zone of Class 1. Disposal of all waste shall be traced appropriately.
All these documents contain the appropriate measures to avoid spread of the GMO in the environment.

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

Hospital conditions

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

All data collected so far concur to demonstrate that MVA-based vaccines are unable to survive, disseminate in and/or displace other organisms and is not pathogenic to animals or plants.

MVA and its recombinant derivatives were found to be safe in normal mice (Ramirez et al. 2000), (Hanke et al. 2002), (Ramirez et al. 2003), (Chen et al.2008) newborn and irradiated mice (Meyer at al. 1991), (Werner et al. 1980), SCID mice (Hanke et al. 2005) and immunosuppressed macaques (Stittelaer et al. 2002), (Hanke et al. 2005).

The MVA HIV-B (MVATG17401) has been released during a first multicenter phase I/II clinical trial, which was hold in four clinical centers in France between 2014 and 2015 (ANRS VRI01 clinical trial).

The advice of the HCB (Haut Conseil de Biotechnologie) required in France by the Ministry for superior education and research to classify the GMO is: the virus has no capacity to significantly multiply in primary human cells. No infectious viral particles are produced by the GMO but the production of pseudo-particles is possible. MVA vector has been already used in several clinical trials in different countries. MVA virus could persist during several days at the injection site but its capacity to disseminate is low. No transmission, contagion or pathogenicity has been observed.

For conducting a clinical trial, no particular requirement has been made by the Authority in terms of patient maintenance in specific hospital structure, other than the times needed for injections which are realized in a classic hospital room. All materials used for GMO (syringes, needles, gauze...) are treated according to the rules of a containment zone of Class 1.

Data collected during this first clinical trial with MVA HIV-B (ANRS VRI01) confirm the safety of this GMO vaccine.

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) Not applicable

(i)	order and/or higher taxon (for animals)
(ii)	family name (for plants)
(iii)	genus

(iv)	species
(v)	subspecies
(vi)	strain
(vii)	cultivar/breeding line
(viii)	pathovar
(ix)	common name

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

NA

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

None

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

Yes ☐	No ☒	Not known ☐
Please give details		

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

None

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

Not applicable

(i) order and/or higher taxon (for animals)
(ii) family name (for plants)
(iii) genus
iv) species
(v) subspecies
(vi) strain

(vii) cultivar/breeding line
(viii) pathovar
(ix) common name

7. Likelihood of genetic exchange in vivo

a) from the GMO to other organisms in the release ecosystem: **None**
b) from other organisms to the GMO: **None**
c) likely consequences of gene transfer: **None**

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.):

Vaccine trials with this or similar MVA constructs have been performed in USA, UK, Germany, Holland, Kenya and Uganda without detectable environmental impact.

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

NA

H. INFORMATION RELATING TO MONITORING

1. Methods for monitoring the GMOs

The GMO is confined in the pharmacy in sealed vials. The product is only drawn at the time of injection. All waste is inactivated prior to disposal and all waste is traced. In absence of a specific accident (the breakage of a vial), no dispersal in the environment is envisaged during the procedure. **The vaccine can be monitored by PCR**

2. Methods for monitoring ecosystem effects

MVA is incapable of replication in humans and thus there should be no virus shedding from the vaccinated individuals. The genetically modified viral vaccine is not able to survive, disseminate in and/or displace other organisms and is not pathogenic to animals or plants.

3. Methods for detecting transfer of the donated genetic material from the GMO to other organisms

NA

4. Size of the monitoring area (m^2)

NA

5. Duration of the monitoring

NA

6. Frequency of the monitoring

NA

I. INFORMATION ON POST-RELEASE AND WASTE TREATMENT

1. Post-release treatment of the site

Surfaces that have been used during vaccination will be cleaned using 2° chloride-solution or other disinfecting solution available at the hospital (i.e. Surfanios).

All materials used during vaccination (used GMO ampoules, syringue, gauze...) will be treated according to the instructions given by the sponsor (Inserm-ANRS) following the national recommendations (if any) and the European Directive 2009/41/EC.

2. Post-release treatment of the GMOs

At the end of the study, all unused vaccine ampoules will be destroyed according to procedures for hospital wastes. Previously, these ampoules will be decontaminated by autoclave (121°C, 20 min).

3. a) Type and amount of waste generated

Material: empty sealed ampoules, syringe, needle, gauze dressing,…
Gloves (single use)

3. b) Treatment of waste

Decontamination and destruction according to approved procedures for GMO given by the sponsor

J. INFORMATION ON EMERGENCY RESPONSE PLANS

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

In case of broken vaccine ampoule, the hospital staff must clean the surface (GMO and broken glass) immediately using gloves, absorbent material and 2° chloride-solution or other commercial disinfectant solution (i.e. Surfanios).
Blood and vaccine that could go out from the injection site during any of the vaccination procedures will be recovered using adhesive gauze dressing which will be applied on the injection site during 30 minutes.
All materials (absorbent material, empty or broken ampoules, syringe, gauze...) used during the cleaning procedures will be decontaminated and/or destroyed according to procedures for the GMO destruction.

2. Methods for removal of the GMO(s) of the areas potentially affected

As described above (J.1).

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

NA

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

NA