PART 1 (COUNCIL DECISION 2002/813/EC)

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

In order to tick one or several possibilities, please use crosses (meaning x or X) into the space provided as (.)

A. General information

1. Details of notification

(a) Member State of notification GE
(b) Notification number B/DE/08/PEI706
(c) Date of acknowledgement of notification 07/08/2008

Title of the project

(a) Clinical Study MI-CP178, "A Phase 1/2a, Randomized, Double-Blind, Placebo-Controlled, Dose-Escalation Study to Evaluate the Safety, Tolerability, Immunogenicity and Vaccine-like Viral Shedding of MEDI-534, a Live, Attenuated Intranasal Vaccine Against Respiratory Syncytial Virus (RSV) and Parainfluenza Virus Type 3 (PIV3), in Healthy 6 to < 24 Month-Old Children and in 2 Month-Old Infants".

(b) Proposed period of release From .01/04/2009. until 31/03/2012

2. Notifier

Name of institution or company:
MedImmune, LLC

3. GMO characterisation

(a) Indicate whether the GMO is a:

viroid		(.)
RNA virus		(x)
DNA virus		(.)
bacterium		(.)
fungus		(.)
animal		
-	mammals	(.)
-	insect	(.)
-	fish	(.)
-	other animal	(.)

specify phylum, class …

(b) Identity of the GMO (genus and species)
Bovine Parainfluenza virus type 3 (bPIV3) plasmid 'backbone': Respirovirus genus Human Parainfluenza virus type 3 (hPIV3) F and HN genes: Respirovirus genus Respiratory Syncytial virus (RSV) F gene: Pneumovirus genus

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

Studies in cell culture and hamsters indicate that the nonessential RSV F gene is maintained without genetic alterations through multiple passage in vitro and in vivo.

b/h PIV3 is attenuated in rhesus monkeys in the upper respiratory tract as judged by restricted replication relative to wild-type hPIV3 (Pennathur 2003). MEDI-534 is genetically stable in vivo and retains RSV F expression after 4 serial passages in the respiratory tract of hamsters.

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

Yes (x) No (.)

If yes, insert the country code(s) BE, ES, FI, GB

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

Yes (x) No (.)

If yes:

- Member State of notification BE
- Notification number B/BE/08/BVW1

Member State of notification: ES

- Notification number: B/ES/08/49

Member State of notification: FI

- Notification number: B/FI/08/1MA

Member State of notification: GB

- Notification number: B/GB/09/R43/01

Please use the following country codes:
Austria AT; Belgium BE; Germany DE; Denmark DK; Spain ES; Finland FI; France FR; United Kingdom GB; Greece GR; Ireland IE; Iceland IS; Italy IT; Luxembourg LU; Netherlands NL; Norway NO; Portugal PT; Sweden SE

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 (x)

If yes:

- Member State of notification …
- Notification number B/../../…

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

MEDI-534 vaccine is a live, attenuated virus that requires a specific host cell for replication. The virus does not persist in the environment and cannot remain infectious outside of a host cell for more than 8 hours. Other factors such as sunlight and heat will further decrease its chance of survival outside a host cell. It is susceptible to common disinfectants and cleaning agents. MEDI-

534 is considered to have minimal potential hazard to clinical site personnel and the environment. No laboratory manipulation of MEDI-534 vaccine or placebo will be conducted at the clinical study sites. Procedures are in place for the shipping, storage, administration and disposal of MEDI-534, and appropriate site training is conducted on study procedures.

MEDI-534 has been shown to be safe and well-tolerated in model animal systems and in clinical studies conducted in the United States in adults and seropositive children. MEDI-534 is a live attenuated viral vaccine, and replication of the vaccine virus in the nasal mucosa is required to generate an immune response. Thus, it is expected that RSV and PIV3 naïve recipients will shed vaccine virus through nasal secretions. Although viral shedding is very restricted in seropositive individuals, the magnitude and duration of viral shedding for MEDI-534 in RSV and PIV3 seronegative children is still unknown. Study procedures are in place to both monitor viral shedding during the trial, as well as to assure that subjects with the potential to come into contact with individuals considered to be at risk for secondary transmission of shed virus will be excluded from participation. Subject safety will be monitored through the duration of the trial.

The bPIV3 strain, that serves as a backbone for the MEDI-534 vaccine, naturally occurs in cows and is known to cause respiratory disease in calves. Virulence of MEDI-534 in bovines has not been studied; however, transmission to animals would require the sharing of nasal secretions from vaccinated infants with the animal. …

The study will be conducted at standard healthcare facilities where paediatric vaccines are commonly administered. Clinical sites where the study is to be conducted will be thoroughly evaluated prior to the initiation of the study to ensure that the facilities are sufficient for storing and administering the vaccine, as well as having the appropriate facilities for the collection, processing and storage of human specimens (nasal wash and serum samples). Additionally, the study protocol provisions for some post-dosing study visits to be conducted in the home of subjects. In-home visits will be conducted by qualified and trained site personnel. The visit will consist of a clinical evaluation of the study subject, as well as the collection of human specimens (nasal wash and serum samples). Specimens obtained in the home will be collected according to the study-specific laboratory manual and promptly transported to the clinical site for processing and storage via a secure, refrigerated container. All clinical site personnel involved in the handling or administration of study vaccine will be trained according to the study protocol, and all supportive documentation, including study specific laboratory and clinical trial material manuals. A thorough study-specific training will occur prior to the initiation of the study via a formal local investigator meeting and/or on-site study initiation visit.

Clinical site staff with the responsibility of administering MEDI-534, collection of serum and nasal wash specimens, or the clinical evaluation of study subjects are instructed to follow the World Health Organization (WHO) universal precautions for the prevention of transmission of infectious agents in healthcare settings (WHO Standard Precautions 2006). Clinical site staff should remember that children presenting for illness visits might be infected with wild-type respiratory viruses, and appropriate infection.

It is not anticipated that the study vaccine or any waste associated with study procedures will affect the surrounding ecosystem or environment.

All components of MEDI-534 come from viruses that are ubiquitous throughout the world and all people are serially exposed to RSV and PIV3 on an annual basis.

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

1. Recipient or parental organism characterisation:

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

(select one only)

viroid	(.)
RNA virus	(x)
DNA virus	(.)
bacterium	(.)
fungus	(.)
animal	
- mammals	(.)
- insect	(.)
- fish	(.)
- other animal	(.)
(specify phylum, class)	…

other, specify …

2. Name

(i)	order and/or higher taxon (for animals)	
(ii)	genus	Respirovirus
(iii)	species	Bovine Parainfluenza Virus Type 3
(iv)	subspecies	…
(v)	strain	bPIV3/15626/84
(vi)	pathovar (biotype, ecotype, race, etc.)	…
(vii)	common name	bPIV3

3. Geographical distribution of the organism

(a) Indigenous to, or otherwise established in, the country where the notification is made:
Yes (x) No (.) Not known (.)

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

(i) Yes (x)

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

Atlantic	x
Mediteranean	x
Boreal	..
Alpine	..
Continental	x
Macaronesian	..

(ii) No (.)
(iii) Not known (.)

(c) Is it frequently used in the country where the notification is made?
Yes (x) No (.)

(d) Is it frequently kept in the country where the notification is made?
Yes (x) No (.)

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	(.)
other, specify	…in association with animals

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

5. (a) Detection techniques

Isolation by culture
Fluorescent focus assay (FFA) to measure titers of infectious virus
Sequence analysis (PCR)

(b) Identification techniques

Reverse-Transcriptase Polymerase Chain Reaction (RT-PCR) using virus-specific primers

6. Is the recipient organism classified under existing Community rules relating to the protection of human health and/or the environment?
Yes (x) No (.)
If yes, specify ACDP Classification 1

7. Is the recipient organism significantly pathogenic or harmful in any other way (including its extracellular products), either living or dead?
Yes (x) No (.) Not known (.)

If yes:

(a) to which of the following organisms:

humans	(.)
animals	(x)
plants	(.)
other	(.)

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

Pathogenicity, Infectivity and Host Range of bPIV3

Bovine PIV3, the plasmid backbone of MEDI-534, is transmitted through the mucous membranes of the eyes, mouth, or nose. The bPIV3 strain used in the MEDI-534 vaccine naturally occurs in cows and causes respiratory disease in calves.

Nonclinical and Clinical Studies with bPIV3 Vaccines Bovine PIV3 was first considered as a potential vaccine candidate based on its restricted replication in rhesus monkeys and chimpanzees (Coelingh 1988, Skiadopoulos 2003). Studies of bPIV3 in adults, children, and infants demonstrated that bPIV3 is attenuated in humans (Clements 1991, Karron 1995, Karron 1996, Greenberg 2005. A bPIV3 vaccine candidate was evaluated in a series of clinical studies and results demonstrated the safety, tolerability, and immunogenicity of the bPIV3 vaccine virus in humans and suggested that it could be used as a vector for delivering hPIV3 and hRSV antigens for immunization of hPIV3 and RSV naive subjects. These clinical studies demonstrated the safety and tolerability of bPIV3 in adults, children and infants as young as 2 months old. There are no reported cases of bPIV3 transmission to humans in nature. There are also no reported cases of bPIV3 transmission to plants and it is doubtful that an animal virus could initiate an infection in plant cells. bPIV3 is an animal virus and requires live animal cells for replication. It is transmitted by close contact and is inactivated by high temperature and changes in pH when exposed to the environment. …

8. Information concerning reproduction

(a) Generation time in natural ecosystems:
Bovine PIV3 is a live virus which does not persist in the environment. It can only remain infectious outside a host cell for less than 8 hours and is rapidly inactivated by sunlight, heat and pH changes.

(b) Generation time in the ecosystem where the release will take place:
No laboratory manipulation or manufacture of the vaccine will take place at the clinical sites. All vaccine will be administered to subjects within the designated clinical sites, which are healthcare facilities.

(c) Way of reproduction: Sexual .. Asexual x

(c) Factors affecting reproduction:
Bovine PIV3 is a live virus which requires a host cell for replication.

9. Survivability

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

(i)	endospores	(.)
(ii)	cysts	(.)
(iii)	sclerotia	(.)
(iv)	asexual spores (fungi)	(.)
(v)	sexual spores (funghi)	(.)
(vi)	eggs	(.)
(vii)	pupae	(.)
(viii)	larvae	(.)
(ix)	other, specify None	

(b) relevant factors affecting survivability:
bPIV3 requires the presence of a suitable host cell for replication and survival. bPIV3 is susceptible to environmental factors such as UV light and temperature, is susceptible to common disinfectants and will not survive for more than 8 hours at room temperature outside of a host cell.

10. (a) Ways of dissemination
As with other respiratory viruses, bPIV3 is transmitted through direct contact with infected animals or indirect contact with contaminated surfaces or objects. Infection can occur through the mucous membranes of the eyes, mouth, or nose.

(b) Factors affecting dissemination
bPIV3 is susceptible to environmental factors such as sunlight, temperature and also common disinfectants.

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)
None

C. **Information relating to the genetic modification**

1. Type of the genetic modification

(i)	insertion of genetic material	(x)
(ii)	deletion of genetic material	(x)
(iii)	base substitution	(.)
(iv)	cell fusion	(.)
(v)	others, specify …	

2. Intended outcome of the genetic modification
MEDI-534 is a chimeric intranasal live attenuated viral vaccine which expresses hPIV3 fusion protein (F) and hemagglutinin-neuraminidase (HN) in a bPIV3 viral backbone. In addition, the human RSV fusion protein (RSV F) has been engineered into position 2 of the genome. Therefore, the bPIV3 backbone of the vaccine is used to deliver antigens thought to be protective against both RSV and hPIV3 infection.

3. (a) Has a vector been used in the process of modification?
Yes (.) No (x)

If no, go straight to question 5.

(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, specify …

(b) Identity of the vector
…

(c) Host range of the vector
…

(d) Presence in the vector of sequences giving a selectable or identifiable phenotype
Yes (.) No (.)

antibiotic resistance (.)
other, specify …

Indication of which antibiotic resistance gene is inserted
…

(e) Constituent fragments of the vector
…

(f) Method for introducing the vector into the recipient organism

(i) transformation (.)
(ii) electroporation (.)
(iii) macroinjection (.)
(iv) microinjection (.)
(v) infection (.)
(vi) other, specify …

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, specify transfection by electroporation

6. Composition of the insert

(a) Composition of the insert
MEDI-534 is a chimeric vaccine which expresses hPIV3 fusion protein (F) and hemagglutinin-neuraminidase (HN) in a bovine PIV3 (bPIV3) viral backbone. In addition, the human RSV fusion protein (RSV F) has been engineered into position 2 of the genome

(b) Source of each constituent part of the insert
bPIV3: PIV3/Kansas/15626/84 strain

hPIV3: Texas/12084/1983 strain
RSV: A2 strain …

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

RSV F protein The RSV fusion (F) protein is a viral surface glycoprotein. It plays a role in cell penetration by the virus and promotes cell to cell spread through the formation of syncytia The F protein is highly conserved between RSV groups (A and B strains). It is also one of the only two RSV components that induce RSV neutralizing antibody and therefore, is an important target of RSV vaccine development.

Human PIV3 HN and F Proteins The human PIV3 hemagglutinin-neuraminidase (HN) protein binds to sialic acid-containing receptors on host cell surfaces, and the fusion (F) protein is involved in the fusion of the viral membrane with the cellular plasma membrane. The F and HN proteins are the only PIV3 antigens that induce neutralizing antibodies, and are important targets of vaccine development for prevention of hPIV3 infection

(d) Location of the insert in the host organism

- on a free plasmid (.)
- integrated in the chromosome (x)
- other, specify …

(e) Does the insert contain parts whose product or function are not known?
Yes (.) No (x)
If yes, specify …

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

1. Indicate whether it is a:

viroid (.)
RNA virus (x)
DNA virus (.)
bacterium (.)
fungus (.)
animal
- mammals (.)
- insect (.)
- fish (.)
- other animal (.)
(specify phylum, class) …

other, specify …

2. Complete name
Human Parainfluenza Virus Type 3

(i)	order and/or higher taxon (for animals)	Paramyxoviridae family
(ii)	family name for plants	…
(iii)	genus	Respirovirus…
(iv)	species	…

(v)	subspecies	…
(vi)	strain	Texas/12084/1983
(vii)	cultivar/breeding line	…
(viii)	pathovar	…
(ix)	common name	hPIV3

Respiratory Syncytial Virus

(j)	order and/or higher taxon (for animals)	Paramyxoviridae family
(ii)	family name for plants	…
(iii)	genus	Pneumovirus
(iv)	species	…
(v)	subspecies	…
(vi)	strain	A2
(vii)	cultivar/breeding line	…
(viii)	pathovar	…
(ix)	common name	RSV

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

Yes (x) No (.) Not known (.)

If yes, specify the following:

RSV and hPIV3 both cause respiratory disease in humans; however, only the HN and F (hPIV3) and F (RSV) genes of these organisms are included in the MEDI-534 vaccine.

(b) to which of the following organisms:

humans	(x)
animals	(.)
plants	(.)
other	..

(b) are the donated sequences involved in any way to the pathogenic or harmful properties of the organism

Yes (x) No (.) Not known (.)

If yes, give the relevant information under Annex III A, point II(A)(11)(d):

Although the RSV F and hPIV3 HN and F proteins are involved in host cell binding and penetration of the viruses, they have been inserted into a bPIV3 plasmid backbone within the MEDI-534 vaccine. This bPIV3 plasmid backbone vaccine has been demonstrated to be attenuated in humans due to its host range restriction as described previously in Section 7 of this application.

Infectivity of RSV and hPIV3 in Humans Both wild-type RSV and hPIV3 are transmitted through direct contact with respiratory secretions of an infected individual or can be transmitted through fomites. Infection can occur when infectious material contacts mucous membranes of the eyes. Both RSV and hPIV3 are ubiquitous and all individuals can expect to be serially exposed to both viruses throughout life.

Virulence of RSV and hPIV3 in Humans RSV is an important respiratory pathogen of infants and young children, causing annual epidemics of bronchiolitis and pneumonia

worldwide. Severe RSV illness commonly occurs among infants with primary infection in the first year of life. RSV is estimated to cause as much as 90% of all childhood bronchiolitis and up to 40% of all pediatric pneumonias (Hall 2000).

hPIV3 is responsible for a spectrum of respiratory symptoms and is second only to RSV as a cause of bronchiolitis and pneumonia among infants and young children (Chanock 2001). Most children are infected with hPIV3 by 2 years of age, but because infection does not confer complete protective immunity, reinfection occurs throughout life (Glezen 1984).

Both RSV and hPIV3 are human pathogens. Non-human primates and rodents are also permissive to hRSV and hPIV3 infection. There is no evidence that RSV and PIV3 activate latent viruses (eg, proviruses) from the cellular genome following infection; RSV and PIV3 are RNA viruses that replicate in the cytoplasm and would not be expected to activate latent viruses. No evidence exists of RSV or hPIV3 being transmitted from humans to animals or vice versa. RSV and hPIV3 are human RNA viruses that are not expected to infect microbes or plant cells which lack receptors for hPIV3 and RSV F and HN proteins, which are the proteins responsible for attachment and entry into host cells.

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 to exposure to biological agents at work?

Yes (x) No (.)

If yes, specify ACDP Classification 2

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

Yes (.) No (.) Not known (.)

Paramyxoviruses replicate in the cell cytoplasm and do not integrate into the host genome; recombination events have not been reported in nature. There is minimal risk of gene exchange between circulating wild-type and vaccine virus (Bukreyev 2006)

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 (x) Not known (.)

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 (.) Unknown (x)

Specify …

(c) is the GMO in any way different from the recipient as far as dissemination is concerned?

Yes (.) No (.) Not known (x)

Specify While bPIV3 has been shown to be poorly transmissible in seronegative children and infants, the magnitude and duration of viral shedding of MEDI-534 in RSV and PIV3 seronegative children is still unknown.

(d) is the GMO in any way different from the recipient as far as pathogenicity is concerned?

Yes (.) No (.) Not known (.)

Specify …

2. Genetic stability of the genetically modified organism

Studies in cell culture and hamsters indicate that the nonessential RSV F gene is maintained without genetic alterations through multiple passages in vitro and in vivo. b/h PIV3 is attenuated in rhesus monkeys in the upper respiratory tract as judged by restricted replication relative to wild-type hPIV3 (Pennathur 2003). MEDI-534 is genetically stable in vivo and retains RSV F expression after 4 serial passages in the respiratory tract of hamsters.

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

Yes (.) No (.) Unknown (x)

(a) to which of the following organisms?

humans (x (Not known))
animals (x (Not known))
plants (.)
other …

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

MEDI-534 has demonstrated attenuation properties, genetic stability and an acceptable safety profile in relevant animal models. Data from two completed clinical studies in adults and seropositive children 1-9 years of age show that MEDI-534 was safe and well tolerated and demonstrated restricted replication in seropositive subjects. MEDI-534 is also currently being evaluated in an on-going study in RSV and PIV3 seronegative children 6 to <24 months of age, and a preliminary evaluation after 2 doses at the lowest dose indicates that the vaccine was well tolerated in this population. The bPIV3 Kansas/15626/84 strain that serves as a backbone for the MEDI-534 vaccine naturally occurs in cows and has been shown to cause respiratory disease in calves. Virulence of MEDI-534 in bovines has not been studied; however, transmission to animals would require the sharing of nasal secretions from vaccinated infants with the animal. MEDI-534 is capable of infecting humans, non-human primates, bovines, hamsters and neonatal ferrets, however, its replication has been shown to be attenuated in humans and non-human primates due to the host range restriction of the bPIV3 backbone. No latent viruses were detected in the master virus seed used for production of MEDI-534. There is no evidence that MEDI-534 activates latent viruses (eg, proviruses) from the cellular genome following infection; bPIV3 is an RNA virus that replicates in the cytoplasm and would not be expected to activate latent viruses. No evidence exists of MEDI-534 being transmitted from humans to animals or vice versa.

4. Description of identification and detection methods

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

(b) Techniques used to identify the GMO
…

F. Information relating to the release

1. Purpose of the release (including any significant potential environmental benefits that may be expected)
MEDI-534 is a live, attenuated intranasal vaccine being investigated for the prevention of lower respiratory tract illness caused by respiratory syncytial virus (RSV) and parainfluenza virus type 3 (PIV3) in healthy young infants. This study is being conducted to evaluate the safety and immune response of this vaccine in healthy 6 to <24 month-old children and in 2 month-old infants.

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 (x)
If yes, specify …

3. Information concerning the release and the surrounding area

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

The study vaccine will be administered at healthcare facilities where paediatric immunizations are commonly administered. Additionally, the study protocol provisions for some post-dosing study visits to be conducted in the home of subjects. In-home visits will be conducted by qualified and trained site personnel. The visit will consist of a clinical evaluation of the study subject, as well as the collection of human specimens (nasal wash and serum samples). Specimens obtained in the home will be collected according to the study-specific laboratory manual and promptly transported to the clinical site for processing and storage via a secure, refrigerated container.

The following are clinical study sites within Germany where study vaccine will be administered:

Site 1

The GMO will be administered at Universitätskinderklinik Ruhr-Universität Bochum, Alexandrinenstr.5, D-44791 Bochum

The GMO will be administered at Sana Klinikum Lichtenberg, Gotlindestr. 2-20, D-10365 Berlin

Site 3

The GMO will be administered at Charité, Universitätsmedizin Berlin**,** Augustenburger Platz 1, D-13353 Berlin

Site 4

The GMO will be administered at Universitätsklinikum Freiburg, Mathildenstraße 1, D-79106 Freiburg

Site 5

The GMO will be administered at Asklepios Klinik Sankt Augustin, Arnold-Janssen-Straße 29, D-53757 Sankt Augustin

Site 6

The GMO will be administered at Universitätsklinik der Johannes Gutenberg-Universität Mainz, Langenbeckstr. 1 /Geb. 106, D-55131 Mainz

(b) Size of the site (m^2):not applicable … m^2
(i) actual release site (m^2): … m^2
(ii) wider release site (m^2): … m^2

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

The study vaccine will be administered at standard healthcare facilities where paediatric vaccines are commonly administered. It is not anticipated that the study vaccine or any waste associated with study procedures will affect the surrounding ecosystem.

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

The bPIV3 Kansas/15626/84 strain that serves as a backbone for the MEDI-534 vaccine naturally occurs in cows and has been shown to cause respiratory disease in calves. Virulence of MEDI-534 in bovines has not been studied; however, transmission to animals would require the sharing of nasal secretions from vaccinated infants with the animal.

4. Method and amount of release

(a) Quantities of GMOs to be released:

It is estimated that a total of 42 MEDI-534 kits (126 total 0.2 ml syringes) and 42 Placebo kits (126 total 0.2 ml syringes) will be distributed to clinical sites in Germany

(b) Duration of the operation:

It is anticipated that enrollment in the EU will begin in April 2009 and will be completed by March 2012.

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

Procedures are in place for transport to and from and storage at clinical sites, administration of the vaccine and clinical sample collections, waste handling, and monitoring of viral shedding from study subjects during the trial. These procedures contain the appropriate measures to avoid the spread of the MEDI-534 vaccine in the environment.

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

Germany has a climate with warm to hot summers and colder, wetter winters.

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.

Overview of Results from Previous Releases of MEDI-534

MEDI-534 (the GMO) has been evaluated in two completed and one ongoing Phase 1 studies within the United States. Additionally, an international Phase1/2a study has been launched in children 2-24 months of age, and enrollment has been initiated in the United States. A list and description of these studies is provided in Table 1. Study MI-CP119 was a Phase 1 study performed to evaluate the safety and tolerability of MEDI-534 in healthy adults. Study MI-

CP130 was a Phase 1 study performed to evaluate the safety and tolerability of MEDI-534 in healthy RSV and PIV3 seropositive children 1 to 9 years of age. MEDI-534 is also being evaluated in an ongoing Phase 1 study (MI-CP149) to evaluate the safety, tolerability, immunogenicity and vaccine shedding in seronegative children 6 to <24 months of age. All of these studies have been/are being conducted within the United States under an Investigational New Drug Application (IND). MEDI-534 has not yet been released outside of the United States.

In these studies, MEDI-534 was administered intranasally to healthy adults, children, and infants. Previous and ongoing clinical studies with MEDI-534 are designed to assess safety, viral shedding, and immunogenicity of this potential vaccine candidate against infection with respiratory syncytial virus (RSV) and parainfluenza virus type 3 (PIV3). These respiratory viruses are spread via large respiratory droplets. Close contact is required for transmission. Both RSV and hPIV3 are ubiquitous in the general population and all individuals can expect to be serially exposed to both viruses throughout life.

Overall, the data from clinical testing in healthy adults and seropositive children 1 to 9 years of age (study MI-CP119 and study MI-CP130, respectively) suggest that MEDI-534 has an acceptable safety profile and restricted replication in seropositive subjects. The immunogenicity, shedding, and safety profiles in the adult and seropositive paediatric population are similar to those previously described with bPIV3 (Clements, 1991; Karron, 1995), the parental virus from which MEDI-534 is derived. The safety profile of MEDI-534 in seronegative young children is also expected to be similar to that of bPIV3 vaccine recipients.

Adverse events included minor reactogenicity events of the upper respiratory tract such as runny nose/nasal congestion, sore throat, and cough. Delivery by an intranasal route of administration may result in epistaxis. Epistaxis was reported in two children (MEDI-534 vs placebo, p=0.49) who received the highest dose (10^6 $TCID_{50}$) of MEDI-534 in Study MI-CP130, and subjects may be at increased risk for developing this adverse event. Systemic symptoms related to low-grade viral illness may potentially be observed and include fever, chills or feverishness, muscle aches, and fatigue (malaise/lethargy). Some respiratory viruses are known to precipitate wheezing illnesses, including asthma exacerbations. Wheezing was observed in two adults who received MEDI-534 in Study MI-CP119. No wheezing events were observed in MEDI-534 recipients during clinical testing in seropositive RSV and PIV3 paediatric populations in Study MI-CP130. Whether MEDI-534 will be associated with an increased risk of wheezing in seronegative RSV and PIV3 children is unknown.

Natural infection with both RSV and PIV3 in young children can result in significant lower respiratorytract infections, including pneumonia, bronchitis, bronchiolitis, croup, and upper respiratory tract infections that can be complicated with acute otitis media. Acute otitis media was diagnosed in two children (p=0.49) who received the highest dose (10^6 $TCID_{50}$) of MEDI-534 in Study MI-CP130 and subjects may be at increased risk for developing this adverse event.

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 organism (if applicable)

(i)	order and/or higher taxon (for animals)	Bovidae
(ii)	family name for plants	…
(iii)	genus	*Bos*
(iv)	species	*taurus*
(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)
…

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

4. Is post-release selection such as increased competitiveness, increased invasiveness for the GMO likely to occur?
Yes (.) No (.) Not known (.)
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
…

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)	…
(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:
Recombination events are extremely rare for paramyxoviruses and have not been reported in nature. There is minimal risk of gene exchange between circulating wild-type and vaccine virus (Bukreyev 2006).

(b) from other organisms to the GMO:
See 7a above.

(c) likely consequences of gene transfer:
Paramyxoviruses replicate in the cell cytoplasm and do not integrate into the host genome. Recombination events are extremely rare for paramyxoviruses and have not been reported in nature. There is minimal risk of gene exchange between circulating

wild-type and vaccine virus (Bukreyev 2006). Furthermore, the absence of any human gene sequences and the absence of any MEDI-534 encoded retroviral polymerase means that the genome of MEDI-534 remains as RNA throughout its lifecycle. Without the ability to convert RNA to DNA and the absence of human gene sequences, integration of any MEDI-534 genetic material into the host genome is highly improbable.

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 stimulated natural environments (e.g. microcosms, etc.):
No studies have been conducted on the ecological impact of MEDI-534 on simulated natural environments.

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

There is no evidence to suggest that MEDI-534 will have any impact on agricultural production, general ecology, environmental quality and pollution in the area of release. MEDI-534 is not an agriculture genetically modified organism. It is an RNA animal virus that replicates in mammalian cells. MEDI-534 cannot infect microbes and plant cells which lack receptors for hPIV3F and HN proteins, which are the proteins responsible for attachment and entry of MEDI-534 into host cells. MEDI-534 does not persist in the environment. It can only remain infectious outside a host cell for no more than 8 hours and is rapidly inactivated by UV, heat and pH changes. It is susceptible to common disinfectants and cleaning agent, such as 1% sodium hypochlorite, 70% ethanol, 2% glutaraldehyde and detergents.

Bovine PIV3 (the backbone for MEDI-534) is antigenically related to hPIV3 but is not virulent in humans. The bPIV3 Kansas/15626/84 strain that serves as a backbone for the MEDI-534 vaccine naturally occurs in cows. Bovine PIV3 is endemic throughout Europe and is among the most frequently diagnosed virus in bovine respiratory disease cases. The bPIV3 infection on its own is usually uncomplicated and subclinical, associated with mild clinical illness only. In a clinical study, 3-week-old calves were directly challenged with bPIV3 and developed only very mild clinical signs (Vangeel 2007). However, under natural conditions, bPIV3 infection is usually accompanied by infection with other respiratory viruses, bacteria and/or mycoplasmas. It is believed that bPIV3 plays a significant role in the bovine respiratory disease complex by predisposing the respiratory tract of cattle to secondary infections (Todd 1975).

H. Information relating to monitoring

1. Methods for monitoring the GMOs
The study will be monitored by MedImmune or its designee on a regular basis throughout the study period in accordance with general monitoring principles set forth in ICH E5. Safety of the study subjects, including monitoring for RSV enhanced disease, will be evaluated throughout the duration of the study. Viral shedding will be evaluated during the study through the collection and evaluation of nasal wash samples. Immune response to the MEDI-534 vaccine will be assessed during the study.

2. Methods for monitoring ecosystem effects
The dissemination and impact of MEDI-534 on ecosystems is limited because dissemination requires close contact with infected nasal secretions. The study will be conducted at standard healthcare facilities where paediatric vaccines are normally administered. It is not anticipated that the study vaccine or any waste associated with study procedures will affect the surrounding ecosystem; therefore monitoring of ecosystem effects is not planned. MEDI-534 is a Biosafety Level 1 organism according to guidelines published by the United States Centers for Disease

Control and Prevention (CDC), and is therefore considered to have minimal potential hazard to laboratory personnel and the environment. Standard universal precautions that are mandated in medical facilities are adequate to prevent accidental transmission.

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

As noted previously, there is minimal risk of gene exchange between circulating wild-type and vaccine viruses. Additionally, MEDI-534 replication and survival is reliant on appropriate host organisms. Therefore, no monitoring of other organisms is planned.

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

Not applicable.

5. Duration of the monitoring

Subjects will be followed from administration of study vaccine through the end of the RSV season or 180 days after the final dose of vaccine, whichever is later.

6. Frequency of the monitoring

Frequency of monitoring for safety, immunogenicity and viral shedding is detailed within the study protocol.

I. Information on post-release and waste treatment

1. Post-release treatment of the site

Clinical study sites will be instructed to follow normal site procedures for disposal of biomedical waste.

2. Post-release treatment of the GMOs

Used and unused MEDI-534 droppers will be discarded as biomedical waste and disposed of according to standard clinical site procedures.

3. (a) Type and amount of waste generated

Up to approximately one hundred and twenty-six (126) 0.2 ml dose syringes of MEDI-534 and placebo could be generated as waste. At least two serum samples and nine nasal wash samples will be collected from each individual. In addition, medical examinations will be conducted on each subject as defined by the study protocol.

3. (b) Treatment of waste

Clinical study sites will be instructed to follow normal site procedures for disposal of biomedical waste.

J. Information on emergency response plans

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

Unexpected spread of MEDI-534 would be limited to accidental release of dropper contents (0.2 ml total); therefore, possibility of spread would be minimal. MEDI-534 is susceptible to common disinfectants and physical inactivation is rapidly achieved by UV irradiation and steam sterilization. MEDI-534 does not survive outside of a host at room temperature for more than 8 hours.

2. Methods for removal of the GMO(s) of the areas potentially affected
If decontamination procedures are deemed necessary for any reason, a freshly prepared 1:10 solution of household bleach (~3.5% sodium hypochlorite) and water can be used.

3. Methods for disposal or sanitation of plants, animals, soils, etc. that could be exposed during or after the spread
Administration of MEDI-534 will occur only within contained clinical sites. It is therefore not anticipated that MEDI-534 will come into direct contact with any plants, animals or soils

4. Plans for protecting human health and the environment in the event of an undesirable effect
As described above, extensive procedural controls are in place for the transport, storage, administration, disposal and monitoring of MEDI-534 vaccination for the duration of the clinical study. Should any unexpected undesirable effect occur, MedImmune will follow standard procedures of assessment of the effect and decisions regarding study continuance.

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