Adviesraad voor Bioveiligheid
Conseil consultatif de Biosécurité

Advice of the Belgian Biosafety Advisory Council on the notification B/BE/25/BVW6 of the University of Tokyo for deliberate release in the environment of genetically modified organisms other than higher plants for research and development

Final version : 04/12/2025
Ref. SC/1510/BAC/2025_1365

Context

The notification B/BE/25/BVW6 has been submitted by the University of Tokyo to the Belgian Competent Authority in September 2025 for a request of deliberate release in the environment of genetically modified organisms (GMOs) other than higher plants for research and development according to Chapter II of the Royal Decree of 21 February 2005.

The planned activity concerns a clinical trial with the title : *"A phase 1, placebo-controlled, randomised, participant- and assessor-blind, single-centre study to assess the safety and immunogenicity of 2 dosages of Nipah measles vector vaccine (MV-NiV) administered subcutaneously either a single dose or as 2 consecutive doses at 4-week interval, in healthy nonexposed volunteers, aged 18-40 years".*

The Nipah virus is a dangerous and potentially fatal virus that can infect both animals and humans. Human infection occurs mainly through contact with infected bats or pigs, but the virus can also spread from person to person. Infection can lead to fever, confusion and breathing problems, among other symptoms. In severe cases, encephalitis, a dangerous brain infection, can also occur. There is currently no specific treatment or vaccine available.

The primary objective of this phase I study is to assess the safety and immogenicity of subcutaneous injection of two different dose levels in a single dose or in two consecutive doses of MV-NiV vaccine in healthy, unexposed volunteers aged 18 to 40 years.
The live attenuated virus (LAV)-MV-NiV, includes the full genome of a live-attenuated measles virus (MV) strain from the Edmonsion lineage, genetically engineered to express the glycoprotein G of the Nipah virus.

The clinical study will be conducted at only one clinical study site in Belgium. Up to 60 healthy subjects in Belgium will be included in this Phase I study. This study will be conducted at one clinical site located in Flanders. The national territory is considered as the potential release area of MV-NiV.

The dossier has been officially acknowledged by the Competent Authority on 08 October 2025 and forwarded to the Biosafety Advisory Council (BAC) for advice.
Within the framework of the evaluation procedure, the BAC, under the supervision of a coordinator and with the assistance of its Secretariat, contacted experts to evaluate the dossier. Three experts from the common list of experts drawn up by the BAC and the Service Biosafety and Biotechnology (SBB) of

Sciensano and one expert from the SBB answered positively to this request. The experts assessed whether the information provided in the notification was sufficient and accurate to state that the deliberate release of the genetically modified organism would not raise any problems for the environment, animal health or human health (people coming in contact with the treated patient and/or with the GMO) in the context of its intended use. See Annex I for an overview of all the comments from the experts.

The scientific evaluation has been performed considering following legislation:
- Annex II (principles for the risk assessment) and annex III (information required in notifications) of the Royal Decree of 21 February 2005.
- Commission Decision 2002/623/EC of 24 July 2002 establishing guidance notes supplementing Annex II to Directive 2001/18/EC.
The pure medical aspects concerning the efficacy of the medicinal product and its safety for the treated patients, as well as aspects related to social, economic or ethical considerations, are outside the scope of this evaluation.

On 10 November 2025, based on a list of questions prepared by the BAC, the Competent Authority requested the notifier to provide additional information about the notification. The answers from the notifier to these questions were received by the Competent Authority on 21 November 2025 and transmitted to the secretariat of the BAC on the same day. This complementary information was reviewed by the coordinator and the experts, after which the BAC was able to come to a conclusion with respect to the environmental aspects associated to the proposed clinical trial.

In parallel with the scientific evaluation of the notification, the Competent Authority also made the dossier available on its website for the one-month public consultation foreseen in the above mentioned Royal Decree. The Competent Authority didn't receive any reactions from the public.

Summary of the scientific evaluation

1. The characteristics of the donor, the recipient or parental organism

The donor, recipient and parental organisms were found to be adequately described in the dossier.

2. Information related to the characteristics of the GMO and the medication

Information related to the molecular characteristics of MV-NiV were adequately described in the dossier. The MV-NiV genome has been evaluated for stability over five serial passages under GMP-like conditions, with no dominant reversion mutations or insert instability detected.

3. The conditions of the release

This phase I study will consist of two treatment groups (low or high dose). The GMO will be administered subcutaneously, either as a single dose or as 2 consecutive doses at 4-week interval, in hospital centres. Subjects will be monitored for 6 months to assess treatment side effects and general health.
In non-clinical studies conducted with the MV-NiV vector, no measles vector RNA was detected in any organs or tissues of MV-NiV treated animals at any tested time point, which is consistent with previously published findings on measles-vectored vaccines. During this clinical trial, the timepoints for shedding sample collection (urine, blood and combined nasopharyngeal and buccal swab samples) are 14 days

after each vaccination, plus ad hoc in case of fever and/or measles-like rash symptoms between 5 and 12 days following the day of each vaccination. As shedding of measles vaccine RNA beyond 14 days post vaccination has been reported before, albeit rarely (Washam et al. 2024[1]), the applicant proposes adding an additional sampling timepoint on day 28 following each vaccine administration.

In order to ensure transparency about sensitivity of the tests and to allow correct interpretation of the results, the notifier provided, following BAC's recommendation, the limit of detection (LOD) for RT-qPCR assays performed for biodistribution and shedding analyses.

Since a separate Subject Information Sheet is not standard practice at site, the applicant confirms that specific standard hygiene recommendations and guidance on preventing vector transmission to other people or release into the environment post-vaccination will be provided to participants as an appendix to the Informed Consent Form (ICF).

Taken together, the information related to the conditions of the release were found to be adequately described in the dossier.

4. The risks for the environment or human health

The GMO, (LAV)-MV-NiV, is a recombinant, live-attenuated measles virus (MV) vaccine containing a transgene cassette expressing the glycoprotein G of the Nipah virus. The G protein of Nipah virus, is not functionally active in the absence of the fusion (F) protein and does not mediate viral entry or membrane fusion. The expression of NiV-G protein introduced in the MV-NiV clinical vector does not enhance virulence or immune suppression.
The genetic modification does not introduce antibiotic resistance markers or other sequences that could confer selective advantage in the environment.

If co-infection of MV-NiV and a wild-type or vaccine-strain measles virus were to occur in a single cell, the generation of viable recombinants is considered highly unlikely, given the biological characteristics of measles virus. The Edmonston strain of measles virus possesses a non-segmented, negative-sense RNA genome that replicates exclusively in the cytoplasm. This replication strategy inherently limits the potential for homologous or non-homologous recombination with other viral genomes.

Although no cases of person-to-person transmission have been reported with attenuated measles vectors, as a precautionary measure, individuals enrolled in the trial must not be in close contact for at least 28 days after vaccination with vulnerable populations (i.e., children under 12 months of age, immunocompromised individuals, pregnant or lactating women, and any other individual that, in the judgment of the investigator, might be at increased risk).

Male participants must refrain from donating sperm and use reliable contraception in combination with a highly effective method used by their female partner of childbearing potential from the first vaccination until at least four weeks after the last dose. As precautionary measure, the period of use of highly effective contraception for female participants of childbearing potential was extendend to at least 6 months after the last vaccination.

[1] Washam M.C., et al. Shedding of measles vaccine RNA in children after receiving measles, mumps and rubella vaccination. J Clin Virol. 2024; 173:105696

Following BAC's request, the duration of restriction on tissue, organ, or fluid (e.g., blood, plasma) donation has been harmonized to up to 6 months after receiving the last vaccination.

The BAC concludes that, based on the attenuated nature of the virus, the lack of vector persistence, non-transmissibility, and the biosafety controls, the overall risk posed by MV-NiV to humans and the environment can be considered as low to negligible.

5. The monitoring, control, waste treatment and emergency plans proposed by the applicant

Like the parental Edmonston B strain, the MV-NiV clinical vector is an enveloped virus with low environmental stability. It is highly sensitive to environmental factors including heat, UV light, common disinfectants (e.g. alcohols, hypochlorite, hydrogen peroxide, and detergents) and variable pH.

To maintain chlorine strength and ensure bleach effectiveness, it is essential to prepare the solution just before use to prevent loss of effectiveness over time. The applicant has taken into consideration that freshly prepared sodium hypochlorite solution must be used as a clean-up solution and/or disinfectant.

Since propagation of MV-NiV seems very unlikely, the BAC supports the view that, in terms of risk for the environment or human health, the proposed measures as described in the revised documents are proportionate and adequate in the context of the intended trial provided that the additional requests as outlined in the conditions here below are met.

Conclusion

Based on the scientific assessment of the notification made by the Belgian experts, the Biosafety Advisory Council concludes that it is unlikely that MV-NiV developed as vaccine against Nipah virus will have any adverse effects on human health or on the environment in the context of the intended clinical trial provided that all the foreseen safety measures are followed.

Therefore, the Biosafety Advisory Council issues a **positive advice with the following conditions**:

- The notifier and the investigators must strictly apply the clinical trial protocol, and all the safety instructions as described in the following documents :
    - Latest version of the ICF
    - Latest version of the Protocol
    - SNIF
    - CAF Amend 251120

- As commited by the applicant, some documents still need to be updated as follows in the next amendment opportunity:
    - On top of the good hygiene recommendations described in Appendix A to the ICF, the appendix will be completed with a reminder to participants to closely follow applicable instructions related to contact with vulnerable populations, use of contraception, and tissue/organ/fluid donations, with cross-references to the relevant sections of the ICF.

- The last sentence of Section 2.3.1.4 of the protocol will be amended as follows: “Nonclinical data for MV-NiV did not provide any evidence for excretion of vaccine virus, although potential shedding cannot be completely excluded at this stage of development”.
- The protocol (and other impacted documents) will be updated by adding an additional sampling timepoint on day 28 following each vaccine administration (i.e. D28 and D56 of the study), where participants will be sampled for blood to assess viraemia and nasopharyngeal swab to assess shedding.
- Figure 12 of the IB will be amended to display vaccinated hamsters as “zero” (below the LOD of the NiV-M RT-qPCR assay).

- Any protocol amendment has to be previously approved by the Competent Authority.

- The notifier is responsible to verify that each study centre has qualified personnel experienced in handling infectious material and that the investigator has the required authorizations to perform the clinical trial activities inside the hospital (laboratory, pharmacy, hospital room, consultation room...) according to the Regional Decrees transposing Directive 2009/41/EC on Contained use of genetically modified micro-organisms.

- The Biosafety Advisory Council should be informed within two weeks when the first patient starts the treatment and the last patient receives the last treatment.

- At the latest six months after the last visit of the last patient included in the trial, the notifier must send to the competent authority at the attention of the Biosafety Advisory Council a report with details concerning the biosafety aspects of the project. This report will at least contain:
  - The total number of patients included in the trial and the number of patients included in Belgium;
  - A summary of all adverse events marked by the investigators as probably or definitely related to the study medication;
  - A report on the accidental releases, if any, of MV-NiV.

Dr. ir. Geert Angenon
President of the Belgian Biosafety Advisory Council

Annex I: Compilations of comments of experts in charge of evaluating the dossier B/BE/25/BVW6 (ref. SC/1510/BAC/2025_1318 and SC/1510/BAC/2025_1366

Compilation of comments of experts in charge of evaluating the dossier B/BE/25/BVW6
And comments submitted to the notifier

17 November 2025
Ref. SC/1510/BAC/2025_1318

Mandate for the Group of Experts: Mandate of the Biosafety Advisory Council (BAC) of 18 september 2025.
Coordinator: Jozef Anné (KUL)
Experts: Rik Gijsbers (KULeuven), Anton Roebroek (KULeuven), Nicolas van Larebeke-Arschodt (UGent, VUB), Willy Zorzi (ULiège), Aline Baldo (SBB)
SBB: Sheela Onnockx

INTRODUCTION

Dossier **B/BE/25/BVW6** concerns a notification from the University of Tokyo (Japan) for the deliberate release in the environment of genetically modified organisms other than higher plants according to Chapter II of the Royal Decree of 21 February 2005.
The notification has been officially acknowledged on 08 October 2025 and concerns a clinical trial entitled "*phase 1, placebo-controlled, randomised, participant- and assessor-blind, single-centre study to assess the safety and immunogenicity of 2 dosages of Nipah measles vector vaccine (MV-NiV) administered subcutaneously either a single dose or as 2 consecutive doses at 4-week interval, in healthy nonexposed volunteers, aged 18-40 years".* The trial will involve the use of a live attenuated virus (LAV)-MV-NiV, including the full genome of a live-attenuated measles virus (MV) strain from the Edmonsion lineage, genetically engineered to express the glycoprotein G of the Nipah virus.

♦ INSTRUCTIONS FOR EVALUATION

Depending on their expertise, the experts were invited to evaluate the genetically modified organism considered in the notification as regards its molecular characteristics and its potential impact on human health and the environment. The pure medical aspects concerning the efficacy of the medicinal product and its safety for the treated patient are outside the scope of this evaluation.
The comments of the experts are roughly structured as in
- Annex II (principles for the risk assessment) of the Royal Decree of 21 February 2005
- Annex III (information required in notifications) of the Royal Decree of 21 February 2005
- Commission Decision 2002/623/EC of 24 July 2002 establishing guidance notes supplementing Annex II to Directive 2001/18/EC.

List of comments/questions received from the experts

Remark: The comments below have served as basis for a list of questions that the Competent authority forwarded on 10-11-2025 to the notifier with a request to provide additional information. The comments or remarks highlighted in grey correspond to the questions addressed to the notifier.

2. INFORMATION RELATED TO THE INVESTIGATIONAL MEDICINAL PRODUCT

A.1. Virus from which the clinical vector was derived (parental virus)

(e.g. information on parental virus; phenotypic and genetic markers; host range, zoonotic potential and replication properties of the parental virus ….)

Comment 1

Has evaluated this item and has no questions/comments.

Comment 2

Has evaluated this item and has no questions/comments.

Comment 3

I think there is an error in Figure 1, the arrow starting from “Matrix protein should be directed to the blue ring”

A possible problematic property of the Edmonton B strain is the use of the CD46 receptor enlarging the spectrum of cells that can be infected. The statement “The parental measles virus is not capable of establishing true latency in the natural host” is not consistent with the observation that late nervous system infections can occur. The statement “There are no defined sequence elements within the measles virus genome responsible for latency or reactivation, as measles virus lacks the regulatory genome architecture or nuclear persistence mechanisms required for such a process.” is not consistent with the existence of subacute sclerosing panencephalitis. The problem should be addressed by estimating the likelihood of this phenomenon in the case of the Edmonton B strain and the clinical vector

SBB’s comment:

The “Matrix protein” are represented in figure 1 of the CAF document. According to the description of the figure, the matrix protein (M) should correspond to the blue ring. Therefore, the following question could be sent to the applicant:

In figure 1 of the CAF document representing the structural organization of the Measles virus, the matrix (M) protein corresponds to the blue ring (see description of the figure). However, in the figure, the arrow starting from “Matrix protein” does not point to the blue ring. The applicant is requested to update the figure where applicable.

During viral latency, a virus remains dormant within a host cell, becoming inactive and producing no new viral particles for a period of time. In subacute sclerosing panencephalitis (SSPE), the measles virus persists in a mutated and defective form within the brain, resulting in a chronic persistent infection. The virus undergoes mutations that hinder its ability to produce envelope proteins, which allows it to evade the immune system and replicate slowly. This persistent infection is responsible for the delayed onset of SSPE symptoms several years after an initial measles infection (Mubbashir et al. 2025).

Coordinator's comment:
I think that the sentence "Importantly, no cases of SSPE have been attributed to the attenuated Edmonston strain or its vaccine derivatives, and these strains show markedly reduced neurotropism and replication capacity in neuronal tissues" is sufficient and no other explanation is needed. "Epidemiological data showed that successful measles immunization programmes protect against SSPE and, consistent with virological data, that measles vaccine virus does not cause SSPE. Measles vaccine does not: accelerate the course of SSPE; trigger SSPE or cause SSPE in those with an established benign persistent wild measles infection" see for example https://doi.org/10.1093/ije/dym207

Comment 4
Has evaluated this item and has no questions/comments.

Comment 5
Has evaluated this item and has no questions/comments.

A.2. Pathogenicity
(e.g. pathogenic properties, available treatment methods, attenuation and biological restrictions of the parental virus ….)

Comment 1
Has evaluated this item and has no questions/comments.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3
A possible question is whether the use of CD46 as a cellular receptor does widen the spectrum of cells attacked by the virus. As to homologous recombination, one might wonder whether the endogenous human mechanisms might do the job. The statement "One non-synonymous mutation in the phosphoprotein (K230T) was observed at a low, stable frequency (~10%) and is not expected to impact replication or virulence significantly." Seems to be in contradiction with the genetic stability which is claimed.

SBB's comment:
According to the CAF document, page 10/53, the Edmonston B strain has acquired the ability to also utilize CD46 - a complement regulatory protein that is ubiquitously expressed on most human cells except erythrocytes. This receptor adaptation enhances the strain's ability to infect a broader range of cell types *in vitro*. However, *in vivo*, CD46 expression is tightly regulated and often downregulated following infection, making infected cells more vulnerable to complement-mediated clearance.

According to page 13/53 of the CAF document, the MV-NiV genome has been evaluated for stability over five serial passages under GMP-like conditions, with no dominant reversion mutations or insert instability detected. Only one point mutation in the phosphoprotein (K230T) was observed at a low, stable frequency (~10%) that is not expected to impact replication or virulence significantly. The statement on page 21/53, which reads "the overall sequence remained highly concordant with the pre-MVS sequence throughout all passages, indicating a high degree of genomic stability" contains subjective wording and could be clarified. The applicant could therefore be requested to specify what is

meant by “highly concordant” and “high degree of stability” by providing quantitative data or exact numbers.

At the end of p21/53 the applicant indicates ‘The overall sequence remained highly concordant with the pre-MVS sequence throughout all passages, indicating a high degree of genomic stability.’. This is subjective wording, please provide exact numbers (what means highly concordant, high degree of stability?)

Comment 4
Has evaluated this item and has no questions/comments.

Comment 5
Has evaluated this item and has no questions/comments.

A.3. Ability to colonise
(e.g. transmission routes, survival outside the host….)

Comment 1
Has evaluated this item and has no questions/comments.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3
It is clear that the vaccine virus is not spreading easily, it is however difficult to believe that it is not capable at all, as it is a biologically active virus.

SBB’s comment:
According to page 13 of the CAF document, “attenuated strains used as vaccine vectors (e.g., Edmonston B, Schwarz, Moraten) do not spread efficiently from person to person. In fact, there has never been a documented case of human-to-human transmission of vaccine-derived measles virus strains.” According to page 23 of the CAF document, “like all measles vaccine strains, MV-NiV clinical vector is not transmissible under normal conditions, and no cases of secondary infection due to vaccine virus spread have been reported since its introduction. Therefore, the clinical vector is not expected to be transmissible, either via respiratory droplets or other environmental routes, and does not present a risk of spreading among humans, including close contacts or healthcare personnel”. These statements suggest that transmission is not absolutely impossible.

Coordinator’s comment:
No evidence of human-to-human transmission of the measles vaccine virus has been reported amongst the thousands of clinical samples genotyped during outbreaks or endemic transmission and individual case studies worldwide – see DOI: 10.1016/j.vaccine.2016.03.092

Comment 4
Has evaluated this item and has no questions/comments.

Comment 5
Has evaluated this item and has no questions/comments.

B. Genetic modification and manufacturing of the clinical vector

(e.g. manufacturing process of the vector; characteristics of the cell lines used for production, information on replicating –competent virus…)

Comment 1

Has evaluated this item and has no questions/comments.

Comment 2

Has evaluated this item and has no questions/comments.

Comment 3

What is the function n of the silent mutation in the Niv gene?

What is the effect of the "missense mutation observed at position 13422 in the L gene in the pre-MVS at a frequency of 100% remained at 100% for P1, P3, and P5."

"The detected sequence matched the pre-MVS sequence to a high degree through five passages." This does not mean at all that the biological function of the corresponding virus is maintained. One gets the impression that the genetic stability is insufficient.

"There is no evidence to suggest that the protein has allergenic or cytotoxic properties, nor does it enhance the survival or transmission of the vector virus." Is there any evidence to back up this statement?

"A third mutation at position 13422 in the MeV-L region resulted in an isoleucine-to-valine substitution outside of any known functional domain, and is therefore unlikely to affect viral replication." Is there any evidence to back up this statement?

"There is no evidence that the transgene affects the vector's cellular or tissue specificity." Has this been tested adequately?

SBB's comment:

According to pages 20-21 of the CAF document, the pre-MVS mutation profile and genetic stability was assessed by NGS through five passages. During the whole-genome sequencing, four point mutations in the pre-MVS relative to the original construct were identified.

The applicant may be requested to clarify whether they have assessed the potential impact of these mutations on the parameters evaluated in the environmental risk assessment (ACF section 5.A) and to provide the corresponding analysis.

As to the pathogenic properties of the MV-NiV, the question arises whether the introduction of the glycoprotein G (NiV-G) in the vector has any pathogenic effect, and whether the NiV-G sequence might participate in recombinations with other viral sequences resulting in a pathogenic virus. The function of the NiV fusion (F) protein might be supplied by a similar protein of another virus.

"the potential for recombination between MV-NiV and the parental virus or related strains is not only biologically implausible but also precluded by study design" . It is unlikely that a study design can protect against rare events.

SBB's comment:

According to page 24 of the CAF document, "the potential for recombination between MV-NiV and the parental virus or related strains is not only biologically implausible but also precluded by study design". While careful study design can greatly reduce risks of recombination and other unintended genetic events, it cannot guarantee absolute prevention of extremely rare events. RNA viruses like measles and

Nipah virus have low recombination rates, and engineered constructs are designed to minimize recombination potential. However, no system is completely fail-safe, and rare genetic events, though highly unlikely, cannot be entirely excluded purely by study design. Therefore, the applicant could be required to revised this statement by being more careful.

Coordinator's comment:
I should not include this question, as it will give only a speculative answer as the potential recombination is not possible.

Comment 4
On p15/62 in B_BE_25_BVW6_IB MV-NIV v1.0_Draft 3_Confidential.pdf section2.2: Table2 contains several sections without proper info.

SBB's comment:
As the document is still in draft form, it is not finished and the missing information in Table 2 will likely be added to the final version.

Comment 5
Has evaluated this item and has no questions/comments.

C. Clinical vector

2.13. - 2.16 . Map of the clinical vector and molecular characteristics, coding genes and regulatory sequences, biologic profile of the clinical vector versus parental virus

Comment 1
Has evaluated this item and has no questions/comments.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3
What is the function n of the silent mutation in the Niv gene?
What is the effect of the "missense mutation observed at position 13422 in the L gene in the pre-MVS at a frequency of 100% remained at 100% for P1, P3, and P5."
"The detected sequence matched the pre-MVS sequence to a high degree through five passages."This does not mean at all that the biological function of the corresponding virus is maintained. One gets the impression that the genetic stability is insufficient.
"There is no evidence to suggest that the protein has allergenic or cytotoxic properties, nor does it enhance the survival or transmission of the vector virus." Is there any evidence to back up this statement?
"A third mutation at position 13422 in the MeV-L region resulted in an isoleucine-to-valine substitution outside of any known functional domain, and is therefore unlikely to affect viral replication." Is there any evidence to back up this statement?
"There is no evidence that the transgene affects the vector's cellular or tissue specificity." Has this been tested adequately?
As to the pathogenic properties of the MV-NiV, the question arises whether the introduction of the glycoprotein G (NiV-G) in the vector has any pathogenic effect, and whether the NiV-G sequence might

participate in recombinations with other viral sequences resulting in a pathogenic virus. The function of the NiV fusion (F) protein might be supplied by a similar protein of another virus.

SBB's comment:
See SBB's comment to previous expert's comment in section B

Coordinator's comment:
Indeed

Comment 4
- The MiV-G coding sequence is inserted (not the gene as mentioned in the txt – p20/53 B_BE_25_BVW6_Part 2_CAF_Confidential.pdf). The definition of 'vector' is confusing to me. I reckoned this referred to the clinical DP, the vaccine itself. This is supposed to be an RNA virus, so I do not understand the reference to a plasmid vector (Fig6) and to 'inserted DNA' (B_BE_25_BVW6_Part 2_CAF_Confidential.pdf at p21/53, mid page). For example, in 2.16 'vector production' means something else?
- At the end of p21/53 the applicant indicates 'The overall sequence remained highly concordant with the pre-MVS sequence throughout all passages, indicating a high degree of genomic stability.'. This is subjective wording, please provide exact numbers (what means highly concordant, high degree of stability?)

SBB's comment:
In the CAF document page 20, the following sentence could be corrected by specifying that the insert corresponds to the NiV G coding sequence instead of the NiV G gene : "The reference sequence consists of an MV Edmonston B backbone with a NiV G gene insert between the MV N and P genes"

Coordinator's comment:
Can be added as editorial comment

The second point has been included in previous SBB's comment for question " A.2. Pathogenicity".

Comment 5
Has evaluated this item and has no questions/comments.

2.17. Potential for recombination

Comment 1
Has evaluated this item and has no questions/comments.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3
"the potential for recombination between MV-NiV and the parental virus or related strains is not only biologically implausible but also precluded by study design" . It is unlikely that a study design can protect against rare events.

SBB's comment:

See SBB's comment in section B to expert's comment 3.

Comment 4
Has evaluated this item and has no questions/comments.

Comment 5
Has evaluated this item and has no questions/comments.

2.18. Biodistribution and shedding

Comment 1
Has evaluated this item and has no questions/comments.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3
"one peripheral blood mononuclear cell sample from an animal that received M-M-R® II (not MV-NiV, Edmonston-Enders) was positive at Day 29. This finding was considered unrelated to MV-NiV vector shedding." Was this observation done in a test with MV-NiV?

SBB's comment:
The following question could be sent to the applicant:
According to the CAF non-confidential document (page 24), in the shedding assessment, all excreta samples (including urine, feces, saliva, and sperm) from MV-NiV–treated animals were also negative for vector RNA at all time points tested, with a single exception: one peripheral blood mononuclear cell sample from an animal that received M-M-R® II (not MV-NiV, Edmonston-Enders) was positive at Day 29. The applicant is requested to clarify what is meant by "this finding was unrelated to MV-NiV vector shedding".

Coordinator's comment:
This sentence "…. Unrelated to MV-NiV…" is indeed not clear.

Comment 4
- On p11/62 in B_BE_25_BVW6_IB MV-NIV v1.0_Draft 3_Confidential.pdf the applicant indicates (as part of the summary), as an argument to clarify the results of biodistribution and shedding, the applicant mentions "the absence of signal may have been due to differences in the MV strains, differences in the route of administration, and/or analytical limitations". Please provide more framing here. If analytical limitations are an issue, the conclusion of that same section - "In line with published results, there was no evidence of viral shedding in the repeat-dose study of MV-NiV at any of the tested timepoints." – is not correct. Also, this statement relates solely to the exps described p43 and onwards, but seems contradictory to me to the results shown further in the document (eg Fig3-4 p24-25/62).

SBB's comment:
Information reported in the summary on page 11/62 of the IB are further developed in section 3.2. Biodistribution pages 43-44. If the absence of signal is partly due to analytical limitations, it would be not appropriate to conclude that no viral shedding occurred. Therefore, the applicant might be asked to be more cautious in its conclusion on page 44 of the IB.

Furthermore, as shedding cannot be fully excluded, the risk cannot be considered as non-existent as it is reported in section 5.9 of the environmental risk assessment reported in the CAF (page 44/53). Therefore, the applicant could also be requested to update the following sentence : “As there are no safety concerns associated with MV-NiV clinical vector shedding into the environment, the risk is non-existent.”

Coordinator’s comment:
For information: mentioned in protocol draft 4 (p23/69). “Nonclinical data for MV-NiV did not provide any evidence for excretion of vaccine virus” which confirms that the risk is non-existing”. Better to say “the overall risk posed by MV-NiV to humans and the environment is low to negligible.” As mentioned in CAF p45/53
I should not include this question.
Furthermore;
The attenuated measles virus is fragile and unstable outside the body**.** > environmental contamination is not a practical concern, and confirmed by “During handling and administration, procedures are performed in accordance with standard Biosafety Level (BSL-1) practices.” ?

- On p35/62 in B_BE_25_BVW6_IB MV-NIV v1.0_Draft 3_Confidential.pdf Fig.12, are data missing here? Only the vehicle is shown. I know there was no signal => should be indicated as zero, or below detection limit? Please provide LoD.

SBB’s comment:
The following question could be sent to the applicant :
Figure 12 is intended to show the viral RNA load of NiV-M in nasopharyngeal–buccal swabs from hamsters vaccinated with MMR+MV-NiV, MV-NiV, or administered formulation buffer only (vehicle). However, the figure currently only shows data for the vehicle group. Are data for the vaccinated groups missing? Even if none of the vaccinated hamsters shed viral RNA after challenge, shouldn’t these groups still be included in the figure, indicated as zero or below the detection limit? The limit of detection (LoD) should also be provided in this section.

- LoD is indicated at p44/62. However, one cannot transfer a LoD for a method between labs. Lorin et al 2012 indicated a LoD of 100 copies/reaction. This should be confirmed in the method used by the applicant. Additionally, to crank up sensitivity, ddPCR would be a better alternative? I understand for the animal studies the platform has shown potential for other epitopes/envelopes. But to assess biodistribution and shedding, the most sensitive technology available should be implemented.

SBB’s comment:
In the article from Lorin et al (2012), the limit of detection (LoD) of the RT-qPCR method was reported as 100 copies/reaction. As LoD for a method cannot be directly transferred between laboratories, the applicant could be requested to specify the limit of detection for the biodistribution studies in cynomolgus Macaques (page 43/62). To increase sensitivity, digital droplet PCR (ddPCR) could be considered as a more suitable alternative. While the platform has demonstrated potential for detecting other epitopes or envelopes in animal studies, the most sensitive technology available should be employed to accurately assess viral biodistribution and shedding. The applicant could be requested to clarify whether other methods have been used to determine the biodistribution.

- P44/62 Viral shedding: also here ‘negative’ is used, I would advise to use ‘below LoD’ instead.

- Again, at P52/62: data were negative in cynomolgus macaques, please indicate the limit of detection. All depends on the sensitivity of the assay used. Was a control taken along to confirm all went as expected in the RNA preparation, RT and qPCR?

SBB's comment:

Data in cynomolgus macaques (pages 44 and 52/62) were reported as negative; however, the limit of detection (LoD) is missing. As the interpretation of the results directly depends on the sensitivity of the assay used, the applicant could be requested to specify the LoD and to clarify whether a control has been included to confirm that RNA extraction, reverse transcription, and qPCR all performed as expected.

Comment 5

Shedding of MV-NiV (in urine and buccal and nasopharyngeal samples) and viraemia (presence in blood) will be assessed in the subjects participating in the clinical study. Samples will be collected 14 days after the day of each vaccination and ad hoc in case of symptoms (i.e. fever or rash) within 14 days after the day of each vaccination. Shedding of MV was evaluated in children after receiving MMR and shows that 34.4 % of patients had measles vaccine virus detected from a nasopharyngeal specimen within 30 days following the first dose of MMR. Of these patients, 15.2 % had detection beyond 14 days after immunization with the longest interval being 28.9 days. These data suggest that although most detection will occur within 14 days after vaccination, prolonged detection out to 4 weeks is not an infrequent occurrence. Shedding of measles vaccine RNA beyond 14 days post vaccination has been reported before, albeit rarely. In one study, a child developed rash and fever 29 days post vaccination and a throat sample collected at presentation was positive. In another study, Measles vaccine RNA was detected from nasopharyngeal samples in children more than 100 days after vaccination (Washam et al., 2024).

As it is the first clinical trial in humans with a recombinant MV Edmonston strain B administrated subcutaneously, samples should be collected in several timepoints before 14 days and after 14 days until minimum day 28 post vaccination.

SBB's comment:

Based on the results presented in this article, it could indeed be relevant to suggest the applicant to increase the number of sample collection timepoints for the shedding analysis.

3. INFORMATION RELATED TO THE CLINICAL TRIAL

3.3. Storage of the clinical vector at the clinical site

(e.g. storage location, conditions of storage, …)

Comment 1

Has evaluated this item and has no questions/comments.

Comment 2

Has evaluated this item and has no questions/comments.

Comment 3

Has evaluated this item and has no questions/comments.

Comment 4

Has evaluated this item and has no questions/comments.

Comment 5
No information is available concerning the conditions of storage and the storage location. MV-NiV should be stored in a contained area (in the BSL-1) or according the following guidelines: https://www.biosecurite.be/sites/default/files/stockage_fr.pdf

SBB's comment:
According to section 3.3 of the CAF document, MV-NiV will be stored frozen (≤-65 °C) at the clinical study site in an area with access limited to authorized site personnel.

3.4. Logistics for on-site transportation of the clinical vector

(information on logistics of in-house transportation, characteristics of the container, disinfection procedures, labelling of the containers, ...)

Comment 1
Has evaluated this item and has no questions/comments.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3
Has evaluated this item and has no questions/comments.

Comment 4
Not ERA related. The study is participant- and assessor-blind but at p28/53 it is indicated that all info is provided on the labels?

SBB's comment:
According to the study title, the study corresponds to a participant- and assessor-blind study. However, according to section 3.4, Logistics for on-site transportation of the clinical vector, the "Name and dosage of the IMP" will be reported on the labels that will be directly affixed to each syringe. It is important that the applicant knows how information would effectively remain hidden during administration to the patient if all information are provided on the syringe.

Coordinator's comment:
Interesting remark, but indeed not ERA related, and I suppose that the randomization number will hide the information. In addition, the location where the clinical trial will be carried out has enough experience with clinical trials.

Comment 5
Has evaluated this item and has no questions/comments.

3.5. Reconstitution, finished medicinal product and administration to the patients

(e.g. mode of administration, information on dosing and administration schedule, information on concomitant medication,…)

Comment 1

Has evaluated this item and has no questions/comments.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3
Has evaluated this item and has no questions/comments.

Comment 4
Has evaluated this item and has no questions/comments.

Comment 5
Has evaluated this item and has no questions/comments.

3.6. Measures to prevent dissemination into the environment

(e.g. control measures, PPE, decontamination/cleaning measures after administration or in the case of accidental spilling, waste treatment, recommendation given to clinical trial subjects, …)

Comment 1
Section 3.6 g) Recommendations given to clinical trial subjects to prevent dissemination (page 32 and 33 of CAFs): No details are presented on how clinical trial subjects should avoid dissemination to immunocompromised persons or young infants. Which instructions will be given to them? Potentially they could be provided with a one page take-home information sheet with the relevant instructions.

SBB's comment:
The following requirement could be sent to the applicant:
According to the CAF document pages 32-33, clinical trial subjects will be informed that MV-NiV may biodistribute and shed for a limited period following vaccination. They will be educated on the potential risks associated with possible dissemination to immunocompromised individuals or young infants, as well as on measures to prevent such exposure. However, no details are presented on how clinical trial subjects should avoid dissemination to immunocompromised persons or young infants.
The applicant is requested to clarify what specific instructions will be provided to subjects. Will these include standard hygiene practices such as covering the mouth and nose when coughing or sneezing and washing hands afterwards?
It is strongly recommended to develop a specific information document for patients, which would bring together all information and instructions for patients and patient's family to avoid potential transmission of the viral vector to other people or to the environment, if any, when patients are leaving the hospital setting.
The following information (with their duration) should be reported in this instruction sheet for the patient:

- Which bodily fluids are anticipated to contain viral vector genome (albeit very low levels)
- Instruction on good hygiene to be practiced
- Instructions aimed at limiting contact with vulnerable population i.e. immunocompromised individuals, children under 6 months and pregnant women
- Effective solutions to decontaminate possible contaminated areas, tissues, skin, …
- Restriction on blood, organs, tissue and cells for transplantation donation
- The obligation to use contraceptive methods

Coordinator's comment:

It is to some extent mentioned – see p 32/53 (CAF), but it is indeed be opportune to have this instruction sheet.

Comment 2

In p31/53 of the B_BE_25_BVW6_Part 2_CAF_Confidential document: Please complete the information for the 1% sodium hypochlorite disinfection protocol by indicating that this solution must be freshly prepared. To maintain chlorine strength and ensure bleach effectiveness, it is crucial to prepare the solution just before use to avoid loss of effectiveness over time.

Please consider that although this disinfectant is available for eliminating enveloped RNA viruses, its use should not be considered universal because its corrode or damage stainless steel, aluminium and the most rubbers components of multi-use devices and supports.

SBB's comment:

According to page 31 of the CAF document, in the event of spills or potential contamination, all affected surfaces and materials must be disinfected with a 1% sodium hypochlorite solution. Given that to maintain chlorine strength and ensure bleach effectiveness, it is crucial to prepare the solution just before use to avoid loss of effectiveness over time, the notifier could be requested to complete the information by indicating that this 1% sodium hypochlorite solution must be freshly prepared.

Comment 3

I think it is necessary to exclude persons taking psychotropic drugs, as these persons present a diminished reliability. It should be kept in mind that such persons are sometimes attracted to clinical trials because of the financial compensation.

SBB's comment:

Question related to drug addicts are related to the patient safety and goes beyond the scope of the environmental risk assessment or the biosafety assessment of the proposed trial. However, according to the protocol, the following exclusion criterion is present: "Suspected or current known alcohol or drug abuse according to the investigator's judgement".

Comment 4

- In addition, all personnel should be vaccinated against measles or have documented immunity to minimize health risks in the unlikely event of exposure. Pregnant personnel (or possibly pregnant personnel) is that a problem?
- P33/52 in CAF: participants will be educated about the potential risk in case of dissemination to immunocompromised persons or young infants and how this can be avoided. Please provide which measures will be installed, what will be thought to the participants.

SBB's comment:

- This question is more related to the protection of workers which is outside the scope of this evaluation. Although special consideration should be given to pregnant personnel, as measle virus may pose risks, and appropriate precautions should be implemented.
- A possible question has been proposed in section 3.6 to comment 1

Comment 5

In the event of spills or potential contamination, all affected surfaces and materials must be disinfected with a 1% sodium hypochlorite solution. Sodium hypochlorite is unstable, diluted solutions (1% sodium

hypochlorite) should be prepared extemporaneously, dated and used rapidly (within the week). It should be stored in the dark and protected from heat.

SBB's comment:
A similar question has already been proposed as SBB's comment to comment 2 in the same section

Additional SBB's comment:
According to the CAF non-confidential document page 11, "wild-type measles virus has well-documented pathogenic potential, particularly in unvaccinated populations and vulnerable groups such as young children, immunocompromised individuals, and pregnant women. The SNIF document (page 13) states that vaccinated subjects will be excluded from close contact (within 28 days of vaccination) with vulnerable populations. i.e. immunocompromised individuals and children under 6 months. Given that pregnant women are also identified as a vulnerable group in the CAF document, should they not likewise be included in the SNIF and in the inclusion criterion 7 of the protocol as a population to be avoided by vaccinated subjects? The applicant could be requested to correct the data where applicable or clarify why pregnant women are not listed among the groups requiring protection.

Additional SBB's comment:
According to the SNIF document (page 13), blood, tissue, and organ donation is prohibited for at least 3 months post-vaccination. On page 32 of the CAF document, it is stated that subjects must refrain from any organ or fluid donation (e.g. blood/plasma) for up to three months after receiving the last vaccination. However, on page 33 of the CAF document, subjects are required to commit to not donating blood or organs during the study period (until at least six months after the last MV-NiV vaccination). There appears to be an inconsistency regarding the duration of the prohibition on blood, tissue, and organ donation post-vaccination. Should the restriction period be three months or six months? The applicant could be requested to clarify the correct time frame and update the relevant document(s) accordingly.

Additional SBB's comment:
According to the inclusion criteria, male participants must refrain from donating sperm and either maintain consistent abstinence or use reliable contraception (e.g., condom or vasectomy) in combination with a highly effective method used by their female partner of childbearing potential from the first vaccination until at least four weeks after the last dose. Whereas female participants of childbearing potential are eligible if they agree to use highly effective contraception from 60 days before the first vaccination until at least 6 months after the last vaccination.
The applicant could be requested to clarify the rationale behind the 4 weeks' time frame for restricting contraception and sperm donation for the male and the 6 months' time frame for restricting contraception for the female participants.

3.7. Sampling and further analyses of samples from study subjects

Comment 1
Section 3.7 Sampling and further analyses of samples from study subjects (page 33 of CAFs): BSL-1 requires the use of eye-protection, but is not specified in mentioned PPE in this section.

SBB's comment:
According to the guideline "CDC/NIH – Biosafety in Microbiological and Biomedical Laboratories (BMBL, 6th edition, 2020)", standard practices, safety equipment, and facility specifications recommended for

BSL-1 includes protective eyewear to be worn by personnel when conducting procedures that have the potential to create splashes and sprays of microorganisms or other hazardous materials.
According to page 30 of the CAF, in section b “Personal protective equipment”, personnel involved in handling or administering MV-NiV must wear standard protective equipment, including : gloves, laboratory coat, surgical mask and protective eyewear (in procedures with splash or aerosol risk). These recommendations are consistent with the recommendations provided in the BMBL guideline. The PPE listed on page 33 corresponds to the minimum PPE to be worn when handling the IMP, including a lab coat during sampling and handling, and gloves which will be available and used as appropriate.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3
Has evaluated this item and has no questions/comments.

Comment 4
Has evaluated this item and has no questions/comments.

Comment 5
Has evaluated this item and has no questions/comments.

3.8. Emergency responses plans

Comment 1
Has evaluated this item and has no questions/comments.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3
Has evaluated this item and has no questions/comments.

Comment 4
Has evaluated this item and has no questions/comments.

Comment 5
Has evaluated this item and has no questions/comments.

5. ENVIRONMENTAL RISK ASSESSMENT

Comment 1
Has evaluated this item and has no questions/comments.

Comment 2
Has evaluated this item and has no questions/comments.

Comment 3

“Expression of the NiV-G transgene. No known toxic, oncogenic, or immunosuppressive properties have been associated with its expression in the context of MV-NiV.” What kind of information is available on this topic? Is it only based on mechanistic considerations, or are there relevant observations?

SBB’s comment:
According to section 5.1 of the CAF document (page 38/53), “no known toxic, oncogenic, or immunosuppressive properties have been associated with its expression in the context of MV-NiV”. The applicant could indeed be requested to clarify what information supports this statement. Specifically, is the conclusion based solely on mechanistic considerations, or are there experimental data or relevant observations that substantiate it?

Coordinator’s comment:
The NiV-G transgene itself is considered biologically safe

“Insertional mutagenesis/ genome integration“ “MV-NiV is non-integrating cytoplasmic RNA virus; no mechanism for genome integration “ Has an attempt been made to detect sequences of MV-NiV in DNA of cells ,or animals exposed to MV-NiV?”

SBB’s comment:
The measles virus is a negative single-stranded RNA virus that does not naturally integrate its RNA genome into the host cell's DNA. It replicates within the cytoplasm using its RNA genome as a template.

Comment 4
Has evaluated this item and has no questions/comments.

Comment 5
In the CAF, p44/53 It is indicated that “As there are no safety concerns associated with MV-NiV clinical vector shedding into the environment, the risk is non-existent.” The probability of shedding is low and shedding cannot be excluded. It will be evaluated in the phase I clinical trial. The risk cannot be consider as non-existent.

SBB’s comment:
This comment has been included in the SBB’s comment to comment 4 in section 2.18

Coordinator’s comment:
As mentioned above should be changed to “the overall risk posed by MV-NiV to humans and the environment is low to negligible.”

6. OTHER INFORMATION

Do you have any other questions/comments concerning this notification that are not covered under the previous items?

Comment 1
Has no further questions/comments.

Comment 2
Has no further questions/comments.

Comment 3
As always, this type of developments and experiments has an apprentice sorcerer aspect. I think that the development of this vaccine is important for public health , and so should be pursued. However, it is evident that such an undertaking is accompanied by a tendency to underestimate risks. I believe that it is the duty of members of an advisory body such as the Biosafety Council to view such scientific developments with a critical eye. My assessment should be seen in that context.

Comment 4
In B_BE_25_BVW6_Part 4 Publieksinformatie (Nl).pdf (p6/7) it is mentioned to used 'bereide javel'. Some goes for the other documents. It would be best to be clear with what is meant and indicate a concentration or a specific dilution one is expected to use.

SBB's comment:
See SBB's comment within section 3.6 Measures to prevent dissemination into the environment

Comment 5
Has no further questions/comments.

References

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Adviesraad voor Bioveiligheid
Conseil consultatif de Biosécurité

Compilation of the expert's evaluations of the answers of AskBio on the list of questions for dossier B/BE/25/BVW6

04 December 2025
Ref. SC/1510/BAC/2025_1366

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Experts: Rik Gijsbers (KULeuven), Anton Roebroek (KULeuven), Nicolas van Larebeke-Arschodt (UGent, VUB), Willy Zorzi (ULiège), Aline Baldo (SBB)
SBB: Sheela Onnockx

INTRODUCTION

Dossier **B/BE/25/BVW6** concerns a notification from the University of Tokyo for a clinical trial entitled "Phase 1, placebo-controlled, randomised, participant- and assessor-blind, single-centre study to assess the safety and immunogenicity of 2 dosages of Nipah measles vector vaccine (MV-NiV) administered subcutaneously either a single dose or as 2 consecutive doses at 4-week interval, in healthy nonexposed volunteers, aged 18-40 years".

On 10 November 2025, based on a list of questions prepared by the BAC (SC/1510/BAC/2025_1300), the Competent Authority requested the notifier to provide additional information about the notification. The answers from the notifier to these questions were received by the Competent Authority on 21 November 2025. This complementary information was reviewed by the coordinator and the experts in charge of the evaluation of this notification.

Evaluation Expert 1

According to my evaluation, the notifier addressed correctly and satisfactorily the comments/questions that have been raised.

Evaluation Expert 2

Concerning the answers of the notifier for the clinical trial submitted by the University of Tokyo related to the use of a live attenuated virus (LAV)-MV-NiV vaccine, it could be considered as satisfactory.
I have no additional request or advice

Evaluation Expert 3

I generally agree with the answers the applicant gave to our questions.
Still, as to Q10: the applicant indicates that they will include more detailed instructions as an appendix to the ICF, but that this document is still in draft form, and subject to change (for example to indications to hygiene).
Also the ICF seems not to be in its final form judging from the reply (and is not included in the files).
I'm not sure whether this should and can be approved as such.

In the last paragraph it is indicated that study participants will be instructed orally. I would strongly advice to also provide this info on paper, to ensure that all info is passed on to the participants.

SBB's comment:

As the various points mentioned in question 10 to be included in the document are found either in Appendix A or as inclusion criteria, it does not seem necessary to contact the applicant again. To ensure the appendix is updated as suggested in their answer, we could add a condition to the advice stating that the reminder for participants, covering instructions on contact with vulnerable populations, use of contraception, and tissue/organ/fluid donations, must be included on top of the good hygiene recommendations described in Appendix A to the ICF

Evaluation Expert 4

I have read the answers to questions 9 and 11 and they are satisfactory.